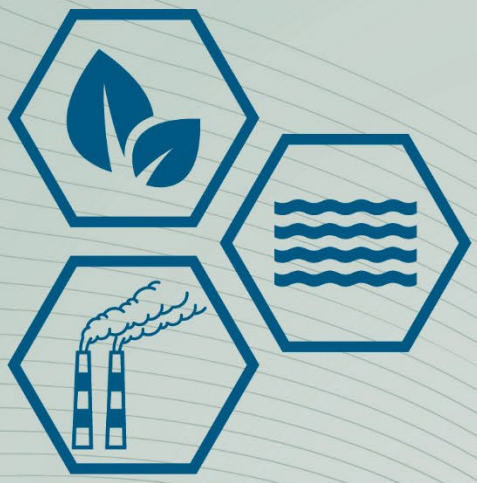

Toxicological Profile for Copper

Draft for Public Comment

April 2022

U.S. Department of Health and Human Services
Agency for Toxic Substances and Disease Registry

CS274127-A

DISCLAIMER

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FOREWORD

This toxicological profile is prepared in accordance with guidelines developed by the Agency for Toxic Substances and Disease Registry (ATSDR) and the Environmental Protection Agency (EPA). The original guidelines were published in the *Federal Register* on April 17, 1987. Each profile will be revised and republished, as necessary.

The ATSDR toxicological profile succinctly characterizes the toxicologic and adverse health effects information for these toxic substances described therein. Each peer-reviewed profile identifies and reviews the key literature that describes a substance's toxicologic properties. Other pertinent literature is also presented, but is described in less detail than the key studies. The profile is not intended to be an exhaustive document; however, more comprehensive sources of specialty information are referenced.

The focus of the profiles is on health and toxicologic information; therefore, each toxicological profile begins with a relevance to public health discussion which would allow a public health professional to make a real-time determination of whether the presence of a particular substance in the environment poses a potential threat to human health. The adequacy of information to determine a substance's health effects is described in a health effects summary. Data needs that are of significance to the protection of public health are identified by ATSDR and EPA.

Each profile includes the following:

(A) The examination, summary, and interpretation of available toxicologic information and epidemiologic evaluations on a toxic substance to ascertain the levels of significant human exposure for the substance and the associated acute, intermediate, and chronic health effects;

(B) A determination of whether adequate information on the health effects of each substance is available or in the process of development to determine the levels of exposure that present a significant risk to human health due to acute, intermediate, and chronic duration exposures; and

(C) Where appropriate, identification of toxicologic testing needed to identify the types or levels of exposure that may present significant risk of adverse health effects in humans.

The principal audiences for the toxicological profiles are health professionals at the Federal, State, and local levels; interested private sector organizations and groups; and members of the public. ATSDR plans to revise these documents in response to public comments and as additional data become available. Therefore, we encourage comments that will make the toxicological profile series of the greatest use.

Electronic comments may be submitted via: www.regulations.gov. Follow the on-line instructions for submitting comments.

Written comments may also be sent to: Agency for Toxic Substances and Disease Registry
Office of Innovation and Analytics
Toxicology Section
1600 Clifton Road, N.E.
Mail Stop S102-1
Atlanta, Georgia 30329-4027

The toxicological profiles are developed under the Comprehensive Environmental Response, Compensation, and Liability Act of 1980, as amended (CERCLA or Superfund). CERCLA section

104(i)(1) directs the Administrator of ATSDR to "…effectuate and implement the health related authorities" of the statute. This includes the preparation of toxicological profiles for hazardous substances most commonly found at facilities on the CERCLA National Priorities List (NPL) and that pose the most significant potential threat to human health, as determined by ATSDR and the EPA. Section 104(i)(3) of CERCLA, as amended, directs the Administrator of ATSDR to prepare a toxicological profile for each substance on the list. In addition, ATSDR has the authority to prepare toxicological profiles for substances not found at sites on the NPL, in an effort to "…establish and maintain inventory of literature, research, and studies on the health effects of toxic substances" under CERCLA Section 104(i)(1)(B), to respond to requests for consultation under section 104(i)(4), and as otherwise necessary to support the site-specific response actions conducted by ATSDR.

This profile reflects ATSDR's assessment of all relevant toxicologic testing and information that has been peer-reviewed. Staffs of the Centers for Disease Control and Prevention and other Federal scientists have also reviewed the profile. In addition, this profile has been peer-reviewed by a nongovernmental panel and is being made available for public review. Final responsibility for the contents and views expressed in this toxicological profile resides with ATSDR.

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VERSION HISTORY

Date	Description
April 2022	Draft for public comment released
September 2004	Final toxicological profile released
October 1990	Final toxicological profile released

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These experts collectively have knowledge of toxicology, chemistry, and/or health effects. All reviewers were selected in conformity with Section 104(I)(13) of the Comprehensive Environmental Response, Compensation, and Liability Act, as amended.

ATSDR scientists review peer reviewers' comments and determine whether changes will be made to the profile based on comments. The peer reviewers' comments and responses to these comments are part of the administrative record for this compound.

The listing of peer reviewers should not be understood to imply their approval of the profile's final content. The responsibility for the content of this profile lies with ATSDR.

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CHAPTER 1. RELEVANCE TO PUBLIC HEALTH

1.1 OVERVIEW AND U.S. EXPOSURES

Copper (Cu) is a chemical element and essential trace mineral that is a reddish metal which occurs naturally in rock, soil, sediment, water, and at low levels, air. The Earth's crust is the primary natural source of copper with an average copper concentration of 50 ppm (Henckens and Worrell 2020). Copper also occurs naturally in all plants and animals, and hence is found in foods and food supplements. The National Academies Institute of Medicine's Recommended Dietary Allowance (RDA) and Tolerable Upper Intake Level (UL) of copper for adult men and women is 900 µg/day and 10,000 µg/day, respectively, but these values vary for children and lactating and pregnant females. In the United States, the geometric mean serum copper level for all adults in the 2015-2016 NHANES was 1146.6 µg/L (18.1 µmol/L). Copper is an essential micronutrient necessary to humans and animals as it is required for adequate growth, lung elasticity, vascular function, neovascularization, neuroendocrine function, and iron metabolism (NRC 2000). However, excess intake of copper can result in toxicity and may adversely interact with certain heavy metals such as zinc. Copper is essential for body system functions, however, there is uncertainty as to the level at which copper becomes toxic. Excess copper exposure can result from external environmental sources such as copper contamination in drinking water, and endogenously from disorders that disturb copper regulation in the body. Copper contaminated water may have a light blue or blue-green color with a metallic, bitter taste (WHO 2004).

Copper is mined in the United States and abroad and is also recovered from scrap which makes up a significant portion of the U.S. copper supply. It is an important commercial metal due to its various properties including corrosion resistance, durability, ductility, malleability, antimicrobial behavior, and electrical and thermal conductivity. Copper and copper compounds are used in several industries including construction, electrical, transportation, and smelting processes. Specific uses of both copper and its compounds include plumbing, electrical wiring, electrical devices, cookware, animal feed, fertilizers, wood preservatives, roofing, and marine antifouling paints (Henckens and Worrell 2020). Due to their antimicrobial properties, copper compounds are used as antimicrobial agents in drinking water treatments, and copper alloys are used in heating, ventilation, and air-conditioning. Copper is also found in ointments and creams as well as multivitamins and dietary supplements. Copper intrauterine devices are a popular form of birth control. Copper nanoparticles, which can be formed naturally or through chemical synthesis, have a variety of uses including as an antibiotic, antimicrobial and anti-fungal agent in plastics, coating, textiles, and pharmaceuticals. The toxicity of copper nanoparticles is distinct from the toxicity of ionic

copper due to their presence in the metallic state and their particle size. This is described in further detail in Section 2.21.

The general public is exposed to copper daily from many sources including air, food, water, and products containing copper. Humans are most likely to ingest copper as its salt but can also be exposed to other forms via inhalation and, to a lesser extent, dermally. In ambient air, the mean copper concentration across 15 U.S. sites ranged from 0.013 to 0.0792 μg/m^3 (EPA 2020a). Concentrations in drinking water can vary widely from ≤0.005 to 10.2 μg/L (see Section 5.5.2). The EPA's action level for dissolved copper in drinking water is 1.3 μg/L. Soluble copper has been reported at various levels in a wide range of food products including fruits, meats, breads, processed foods, dairy, bottled water, and juices, among others. Copper is also measured in blood, urine, hair and nails, and human breastmilk.

1.2 SUMMARY OF HEALTH EFFECTS

Information on the toxicity of copper and copper compounds comes primarily from oral studies in both humans and animals exposed to copper or copper sulfate, the most commonly used compound. Human studies include controlled-exposure and epidemiological studies, and primarily examine the effect of different copper doses from diet and drinking water. Additionally, human studies have also examined the associations of copper in biological fluids or copper in the environment with various health outcomes. Oral toxicity studies evaluate various deficiency or toxicity endpoints, and the gastrointestinal and hepatic systems are the most studied endpoints and appear to be sensitive endpoints of copper exposure. These two endpoints underwent systematic review. There were fewer studies examining inhalation or dermal exposure to copper in humans and animals. A limited number of studies in both humans and animals examined copper toxicity due to inhalation or dermal exposure. The genotoxicity of copper and copper compounds has been evaluated using a variety of species and protocols. Disorders such as Wilson's disease, Indian childhood cirrhosis, and idiopathic copper toxicosis are characterized by excess copper build-up in the body, primarily the liver, and typically result in liver damage. These disorders are further described in Section 2.9. Figure 1-1 and Figure 1-2 summarize the health effects observed in human and animal inhalation and oral studies, respectively. Taken together, the database demonstrates that the most sensitive endpoints for copper toxicity appear to be the gastrointestinal and hepatic systems. A systematic review was conducted on these endpoints. The weight-of-evidence conclusions are defined and summarized in Appendix C.

1. RELEVANCE TO PUBLIC HEALTH

The review resulted in the following hazard identification[1] conclusions:

- Gastrointestinal effects are presumed health effects from exposure to copper.
- Hepatic effects are suspected health effects from exposure to copper.

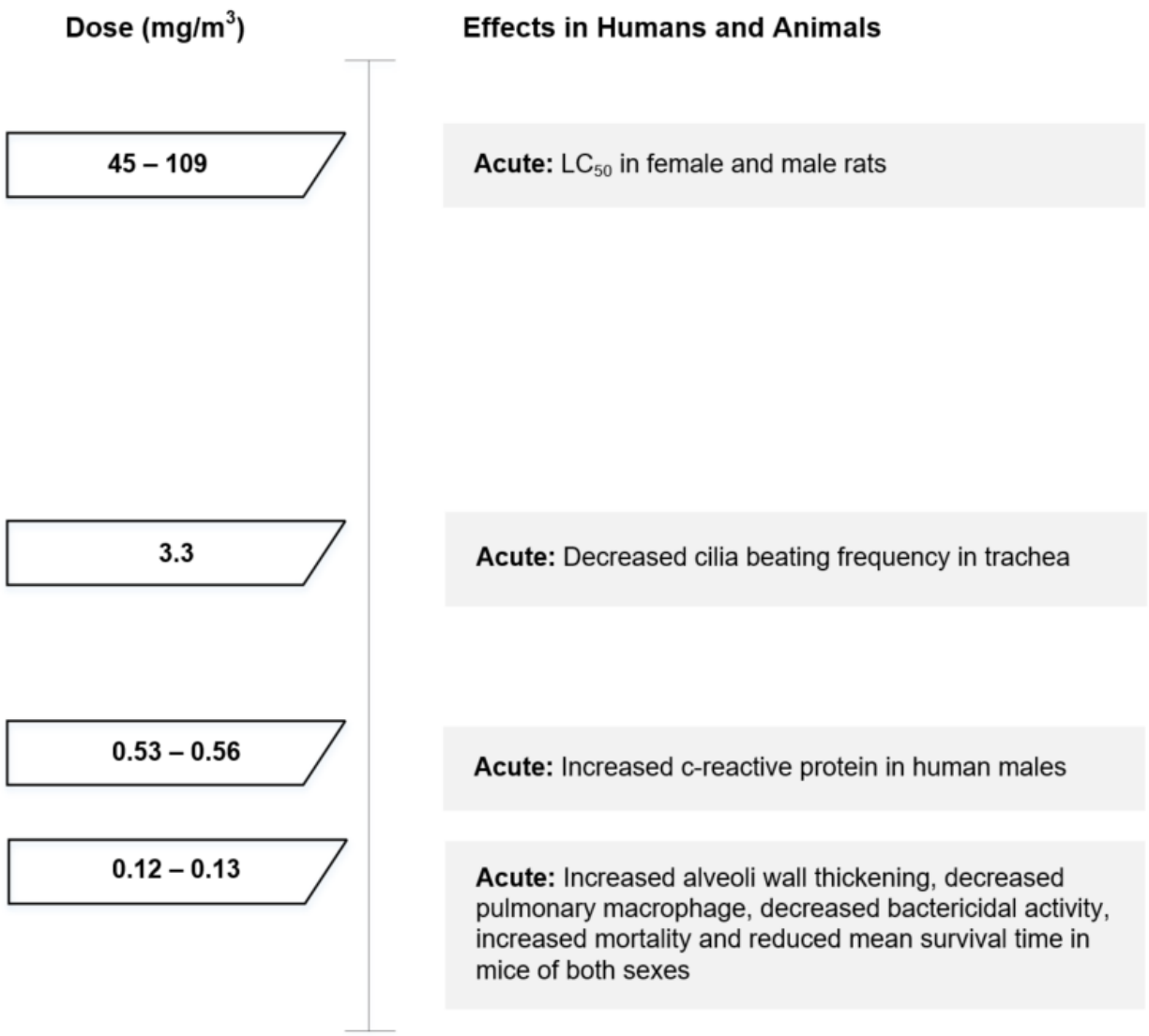
Gastrointestinal Effects. Numerous case studies and epidemiological studies have reported gastrointestinal upset in humans from oral exposure to copper supporting the finding that the gastrointestinal system is a target of copper toxicity. Copper is absorbed rapidly by the stomach and intestine when ingested, and induces abdominal pain, nausea, and vomiting as reported in communities episodically exposed to excess levels of copper in their drinking water (Eife et al. 1999; Knobeloch et al. 1994, 1998; Pizarro et al. 2007) and in some occupational settings (Suciu et al. 1981). A study in humans identified a dose-response relationship between ingestion of drinking water with elevated copper levels and gastrointestinal symptoms (Pizarro et al. 1999). In adults, the incidence of gastrointestinal symptoms (i.e., nausea, vomiting, abdominal pain) was higher in subjects repeatedly exposed to copper doses ranging from 0.07 to 0.17 mg Cu/kg/day (3 to 6 mg Cu/L) (Araya et al. 2003b, 2004; Pizarro et al. 1999, 2001). However, diarrhea does not appear to be associated with exposure at these low copper concentrations (Pizarro et al. 1999, 2001). Females appear to be more sensitive to copper, developing gastrointestinal symptoms at lower doses compared to males (Araya et al. 2004). A study with infants observed no increase in the gastrointestinal symptoms following daily exposure to doses up to 0.319 mg Cu/kg/day (2 mg Cu/L) in drinking water for 9 months (Olivares et al. 1998). Several studies where adults were exposed to single doses of copper ranging from 0.012 to 0.18 mg Cu/kg (4 to 12 mg Cu/L) in drinking water found that nausea is the most reported gastrointestinal symptom (Araya et al. 2001, 2003a, 2003c; Gotteland et al. 2001; Olivares et al. 2001). Vomiting was also reported following exposure to single doses of 0.018 to 0.037 mg Cu/kg (6 to 12 mg Cu/L) (Gotteland et al. 2001; Olivares et al. 2001). Other gastrointestinal effects induced by copper included delayed gastric emptying (Araya et al. 2003a) and increased gastric permeability (Gotteland et al. 2000), both of which were independent of the gastrointestinal symptoms. Evidence in laboratory animals indicates that oral copper exposure results in histological changes, such as ulcerations throughout the gastrointestinal tract, and changes in intestinal microbiome homeostasis at doses ≥2.4 mg Cu/kg/day (Cheng et al. 2020; Kadammattil et al. 2018;

[1] For additional details on the definitions on the hazard identification categories the reader is referred to Appendix C.

Khushboo et al. 2018; NTP 1993; Yamamoto et al. 2004). Pregnant rabbits exposed to high copper doses had diarrhea, stomach hemorrhage, ulcerations, and discolored stomach lining (Munley 2003a, 2003b).

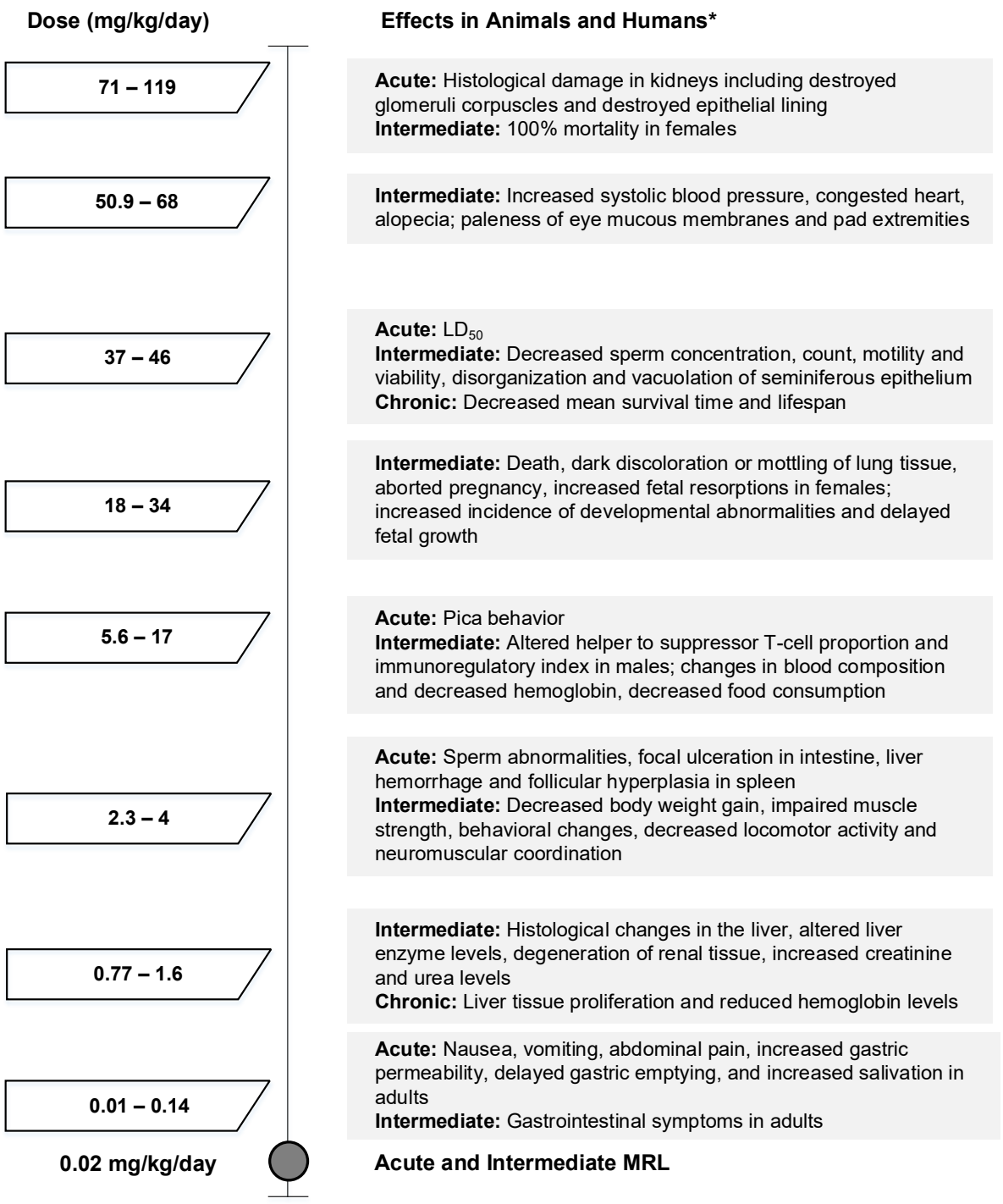
Hepatic Effects. Human case studies report increases in liver enzymes (i.e., alanine aminotransferase, aspartate aminotransferase), liver impairment, jaundice, centrilobular necrosis, and hepatomegaly following exposure to very high doses of copper substances (Ahasan et al. 1994; Akintonwa et al. 1989; Chuttani et al. 1965; Du and Mou 2019; Gamakaranage et al. 2011; Gunay et al. 2006; Lamont and Duflou 1988; Lubica et al. 2017; O'Donohue et al. 1993; Park et al. 2018; Pratt et al. 1985). Controlled-exposure studies, where humans were exposed to lower levels of copper in drinking water, found no alterations or indications of damage to the liver, including studies in infants (Olivares et al. 1998; Zietz et al. 2003a, 2003b) and adults (O'Connor et al. 2003). Individuals with Wilson's disease, Indian childhood cirrhosis, and idiopathic copper toxicosis are particularly susceptible to liver toxicity caused by altered copper homeostasis. These diseases can be exacerbated by excess oral copper intake (i.e. consuming milk boiled or stored in brass vessels) relative to the ability of the liver to safely store copper. Evidence of hepatotoxicity resulting from excess copper exposure primarily comes from laboratory animal experiments, and most of these studies examined rats. Liver effects were seen in doses as low as 1.6 mg Cu/kg/day in experimental animals exposed daily for 30 days, effects included elevated hepatic marker enzymes, lipid damage, and extensive histopathological observations in the liver such as acute swelling of hepatocytes, coagulative necrosis represented by karyolysis of nuclei, and hyperplasia of the epithelial lining of bile ducts (Hashish and Elgaml 2016). Similar or more severe hepatic changes were noted in acute- and intermediate-duration exposure studies. Additional effects include reduced liver weight, massive cellular degeneration, liver hemorrhage from acute-duration exposure (Alhusaini et al. 2018a, 2018b; Kadammattil et al. 2018); and centrilobular necrosis, enlarged liver, jaundice and hepatic lesions from intermediate-duration exposure (Khushboo et al. 2018; Rana and Kumar 1980; Sakhaee et al. 2012; Seven et al. 2018; Suttle and Mills 1966). NTP (1993) noted the effects in the liver were dose-related, and Kumar et al. (2016a) reported that increased severity of histological findings in rats were dose- and duration-related.

Figure 1-1. Health Effects Found in Humans and Animals* Following Inhalation Exposure to Copper



*All effects listed were observed in animals, unless otherwise specified.

Figure 1-2. Health Effects Found in Humans and Animals* Following Oral Exposure to Copper



*All effects listed were observed in animals, unless otherwise specified.

1.3 MINIMAL RISK LEVELS (MRLs)

As presented in Figure 1-3 following acute-duration inhalation exposure, the respiratory and immunological systems are the most sensitive targets of copper toxicity. The inhalation database was inadequate for the derivation of inhalation minimal risk levels (MRLs) for any duration of exposure. The gastrointestinal, hepatic, and neurological systems appear to be sensitive targets of oral copper toxicity, as shown in Figure 1-4. The oral database was adequate for the derivation of acute- duration oral MRL for copper. The acute-duration oral MRL was also used as the intermediate-duration oral MRL. There was insufficient data for the derivation of an oral chronic MRL for copper. MRLs derived for the oral exposure route for copper are summarized in Table 1-1 and are discussed in greater detail in Appendix A.

Figure 1-3. Summary of Sensitive Targets of Copper – Inhalation

The respiratory and immunological systems are the most sensitive targets of copper inhalation exposure.

Numbers in circles are the lowest LOAELs among health effects in animals.

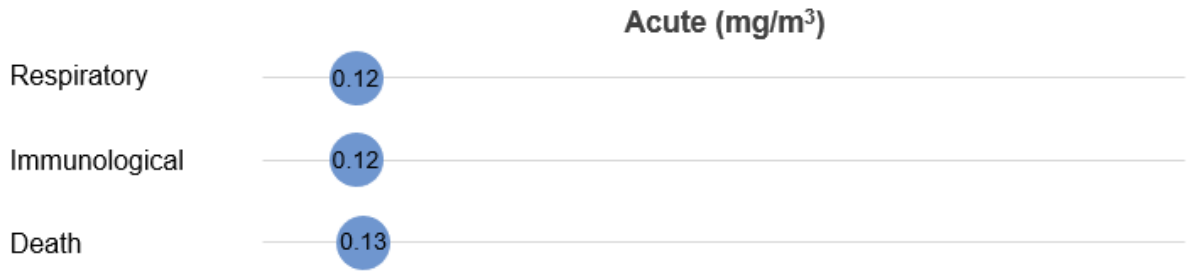


Figure 1-4. Summary of Sensitive Targets of Copper – Oral

The gastrointestinal, hepatic, and neurological systems are the most sensitive targets of copper oral exposure.

Numbers in triangles and circles are the lowest LOAELs among health effect in humans and animals, respectively.

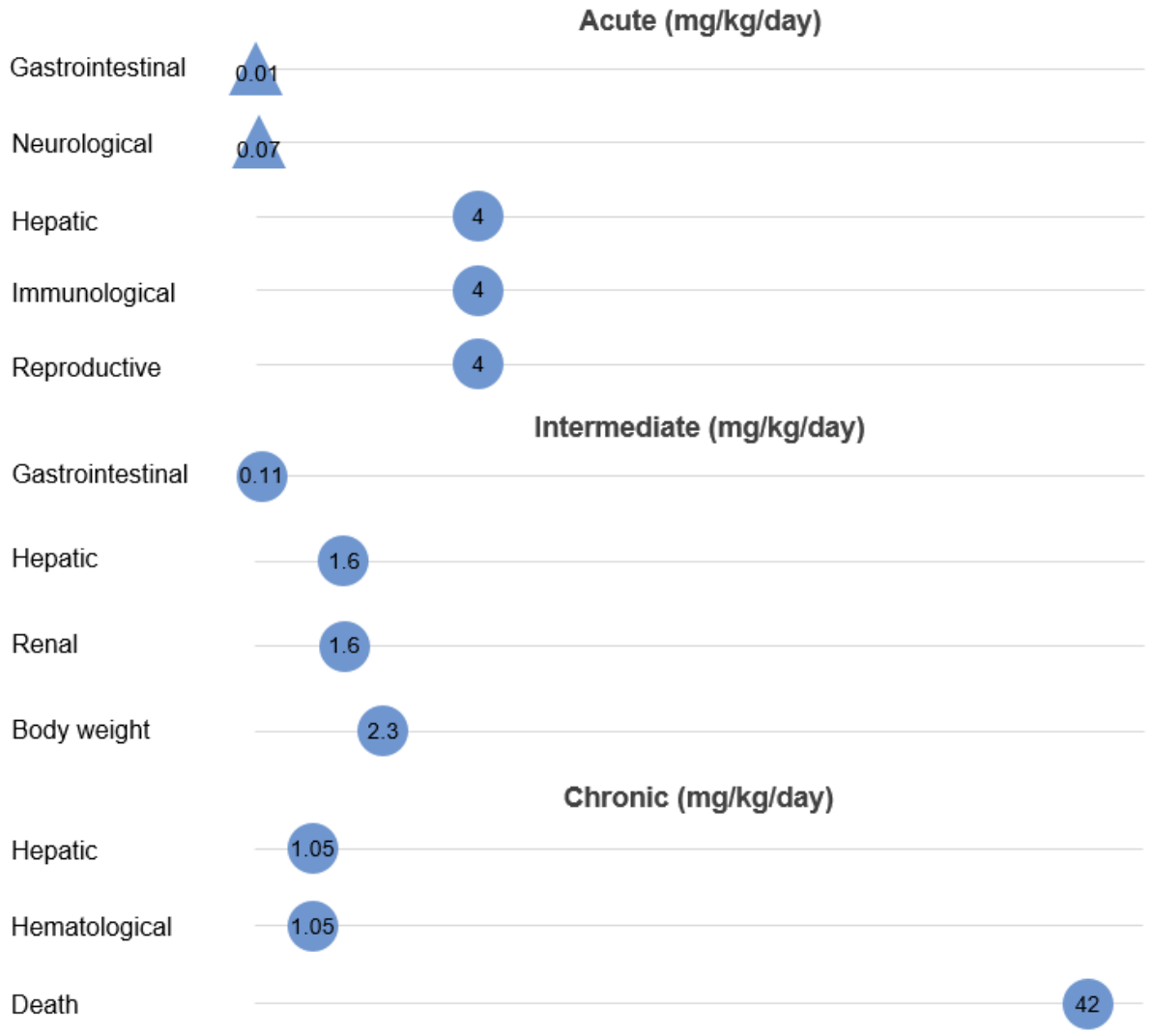


Table 1-1. Minimal Risk Levels (MRLs) for Copper[a]

Exposure duration	MRL	Critical effect	Point of departure	Uncertainty& modifying factor	Reference
Inhalation exposure (mg Copper/m^3)					
Acute	Insufficient data for derivation of an MRL				
Intermediate	Insufficient data for derivation of an MRL				
Chronic	Insufficient data for derivation of an MRL				
Oral exposure (mg Copper/kg/day)					
Acute (provisional)	**0.02**	Gastrointestinal symptoms in women	$BMDL_{10}$: 0.05	UF: 3	Pizarro et al. 1999
Intermediate (provisional)	The provisional acute-duration oral MRL of **0.02 mg/kg/day** is adopted as the provisional intermediate-duration oral MRL.				
Chronic	Insufficient data for derivation of an MRL				

[a]See Appendix A for additional information

BMDL = 95% lower confidence limit on the BMD (subscript denotes benchmark response of exposure dose associated with 10% extra risk); LOAEL = lowest-observed-adverse-effect-level; UF = uncertainty factor

CHAPTER 2. HEALTH EFFECTS

2.1 INTRODUCTION

The primary purpose of this chapter is to provide public health officials, physicians, toxicologists, and other interested individuals and groups with an overall perspective on the toxicology of copper. It contains descriptions and evaluations of toxicological studies and epidemiological investigations and provides conclusions, where possible, on the relevance of toxicity and toxicokinetic data to public health. When available, mechanisms of action are discussed along with the health effects data; toxicokinetic mechanistic data are discussed in Section 3.1.

A glossary and list of acronyms, abbreviations, and symbols can be found at the end of this profile.

To help public health professionals and others address the needs of persons living or working near hazardous waste sites, the information in this section is organized by health effect. These data are discussed in terms of route of exposure (inhalation, oral, and dermal) and three exposure periods: acute (≤14 days), intermediate (15–364 days), and chronic (≥365 days).

As discussed in Appendix B, a literature search was conducted to identify relevant studies examining health effect endpoints. Figure 2-1 provides an overview of the database of studies in humans or experimental animals included in this chapter of the profile. These studies evaluate the potential health effects associated with inhalation, oral, or dermal exposure to copper and copper compounds, but may not be inclusive of the entire body of literature.

Summaries of human epidemiological and ecological studies are presented in Table 2-4, Table 2-5, Table 2-6, and Table 2-7. For these tables, note that the study quality varies and study limitations are included. Animal and human inhalation studies are presented in Table 2-1 and Figure 2-2, and animal and human oral studies are presented in Table 2-2 and Figure 2-3; animal dermal studies are presented in Table 2-3.

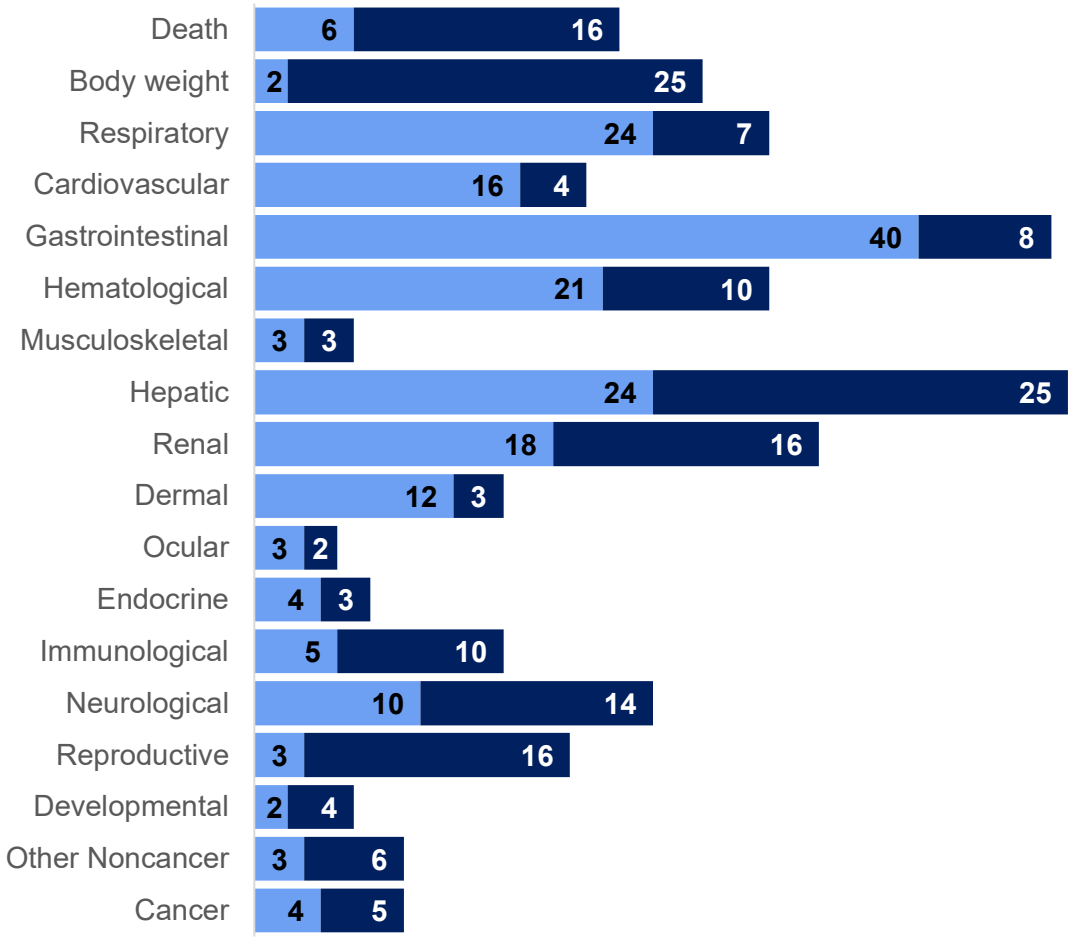
Levels of significant exposure (LSEs) for each route and duration are presented in tables and illustrated in figures. The points in the figures showing no-observed-adverse-effect levels (NOAELs) or lowest-observed-adverse-effect levels (LOAELs) reflect the actual doses (levels of exposure) used in the studies. LOAELs have been classified into "less serious" or "serious" effects. "Serious" effects are those that evoke failure in a biological system and can lead to morbidity or mortality (e.g., acute respiratory distress or death). "Less serious" effects are those that are not expected to cause significant dysfunction or death, or those whose significance to the organism is not entirely clear. ATSDR acknowledges that a considerable amount of judgment may be required in establishing whether an endpoint should be

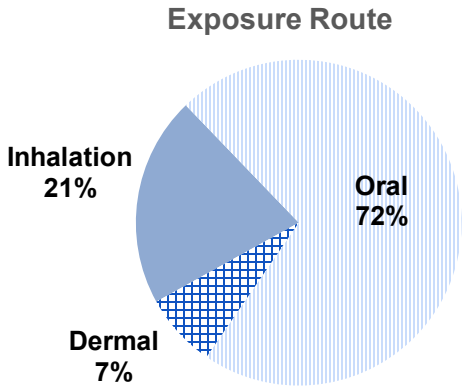
classified as a NOAEL, "less serious" LOAEL, or "serious" LOAEL, and that in some cases, there will be insufficient data to decide whether the effect is indicative of significant dysfunction. However, the Agency has established guidelines and policies that are used to classify these endpoints. ATSDR believes that there is sufficient merit in this approach to warrant an attempt at distinguishing between "less serious" and "serious" effects. The distinction between "less serious" effects and "serious" effects is considered to be important because it helps the users of the profiles to identify levels of exposure at which major health effects start to appear. LOAELs or NOAELs should also help in determining whether or not the effects vary with dose and/or duration, and place into perspective the possible significance of these effects to human health.

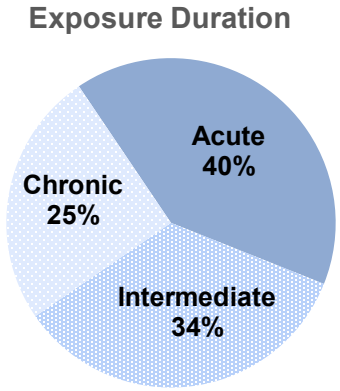
A User's Guide has been provided at the end of this profile (see Appendix D). This guide should aid in the interpretation of the tables and figures for LSEs and MRLs.

Figure 2-1. Overview of the Number of Studies* Examining Copper Health Effects

Most studies examined the potential gastrointestinal and hepatic effects of copper.
More studies have evaluated health effects in **humans** than **animals** (counts represent studies examining endpoint).







*Includes studies discussed in Chapter 2; studies examined multiple endpoints. A total of 161 studies (including those finding no effect) have examined toxicity.

Table 2-1. Levels of Significant Exposure to Copper – Inhalation (mg/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE									
Markert et al. 2016									**Copper metal**
1	HUMAN 5M	3 weeks, 3 periods, 6 hours/ period	0, 0.53	HE	Immuno		0.53		Significant increase in C-reactive protein (p=0.001), which increased the greatest between the 6 and 24th hour after the exposure
Rush 1991									**Copper metal**
2	RAT (Sprague-Dawley) 5M, 5F	4 hours (NS)	11.2, 44.8, 88, 208.8	LX	Death			45 F	LC50
								109 M	LC50
Drummond et al. 1986									**Copper sulfate**
3	MOUSE 23-100B	3 hours	0, 0.56, 1.21, 3.3	CS LE OF	Death			0.56	4.2-5.9 day decrease in mean survival and 54-70% increase in mortality
					Resp	3.3			
					Immuno		0.56		Decreased bactericidal activity of pulmonary macrophage
Drummond et al. 1986									**Copper sulfate**
4	MOUSE 15-24B	1-2 weeks 5 days/weeks 3 hours/day	0, 0.12, 0.13	CS LE OF	Death			0.13	25-31% increase in mortality in both sexes and reduced mean survival time by 1.3-1.5 days
					Resp		0.12		Increased alveolar wall thickness and irregular appearance

Table 2-1. Levels of Significant Exposure to Copper – Inhalation (mg/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Immuno		0.12		Decreased bactericidal activity of pulmonary macrophage
Drummond et al. 1986									**Copper sulfate**
5	HAMSTER 4NS	3 hours	0, 1.21, 3.3	OF	Resp	1.21	3.3		Decreased cilia beating frequency in trachea
Drummond et al. 1986									**Copper sulfate**
6	HAMSTER 4NS	1-2 weeks 5 days/weeks 3 hours/day	0, 0.12, 0.13	HP OF	Resp	0.13			
INTERMEDIATE EXPOSURE									
Johansson et al. 1983									**Copper chloride**
7	RABBIT (NS) 8M	1 month 5 days/week 6 hours/day	0,0.28	OF	Resp	0.28			
					Immuno	0.28			
Johansson et al. 1984									**Copper chloride**
8	RABBIT (NS) 8M	4-6 weeks 5 days/week 6 hours/day	0, 0.28	GN HP	Resp	0.28			

Table 2-1. Levels of Significant Exposure to Copper – Inhalation (mg/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects

[a]The number corresponds to entries in Figure 2-2.

To calculate the copper concentration in studies using copper sulfate, it was assumed that the study used copper sulfate anhydrous (molar mass: 159.61 g/mol) unless otherwise specified that copper sulfate pentahydrate (molar mass: 249.69 g/mol) was used.

B = both sexes; CS = clinical signs; F= female(s); GN = gross necropsy; HE = hematological; HP = histopathology; Immuno = immunological; LE = lethality; LC50 = concentration producing 50% death; LOAEL = lowest-observed-adverse-effect-level; M = male(s); NOAEL = no-observed-adverse-effect-level; NS = not specified; OF = organ function; Resp = respiratory.

Figure 2-2. Levels of Significant Exposure to Copper – Inhalation
Acute (≤14 days)

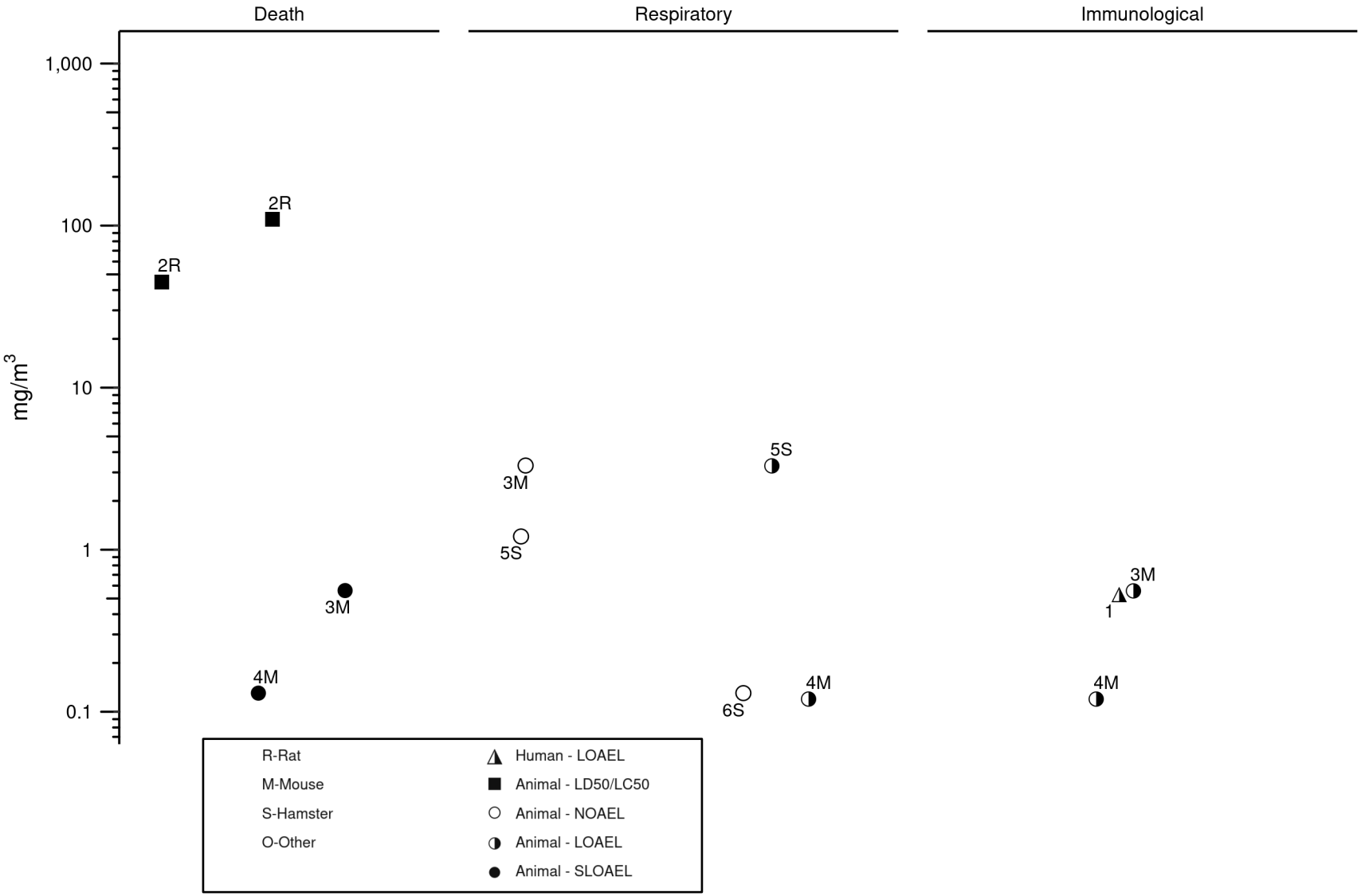


Figure 2-2. Levels of Significant Exposure to Copper – Inhalation
Intermediate (15-364 days)

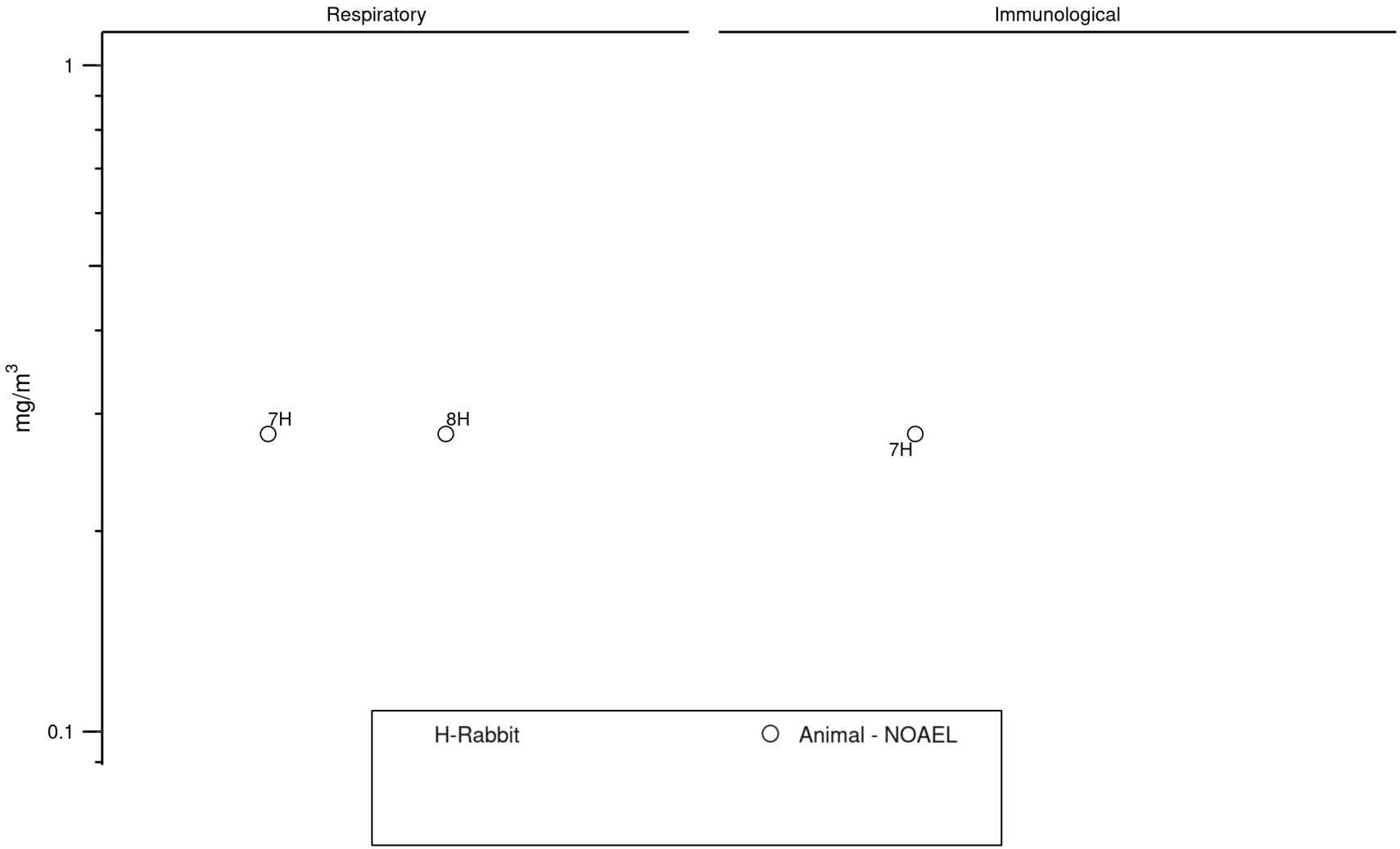


Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE									
Araya et al. 2001**									**Copper sulfate**
1	HUMAN 179B	Once (W)	0, 0.006, 0.012, 0.018, 0.025	CS	Gastro	0.01	0.02		Significantly increased frequency of nausea, 17/179 subjects
Araya et al. 2003a**									**Copper sulfate**
2	HUMAN 15M, 15F	Once (W)	0, 0.046	OF	Gastro		0.05		Nausea in 9/30 subjects and delayed gastric emptying
Araya et al. 2003c**									**Copper sulfate**
3	HUMAN 58-73F	Once (W)	0, 0.03, 0.04, 0.06, 0.08, 0.09, 0.12, 0.18	CS WI	Gastro	0.06	0.09		Nausea in 50/269 subjects
Gotteland et al. 2001**									**Copper sulfate**
4	HUMAN 15M, 16F	Once (W)	0, 0.03	CS OF	Gastro		0.03		Nausea (6/31 subjects) and vomiting (2/31 subjects); 36.5% increase in gastric permeability to sucrose
Olivares et al. 2001**									**Copper sulfate**
5	HUMAN 30M, 31F	once (W)	0, 0.006, 0.012, 0.018, 0.025, 0.031, 0.037	CS	Gastro	0.01	0.01		Nausea in 5/53 participants
Pizarro et al. 1999†									**Copper sulfate**
6	HUMAN 60F	2 weeks daily (W)	0.0006, 0.03, 0.07, 0.1	BI BW CS	Bd wt	0.1			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Gastro	0.03	0.07		Abdominal pain, nausea, and/or vomiting; a $BMDL_{10}$ of 0.05 mg/kg/day was calculated for gastrointestinal symptoms ($BMDL_{10}$=0.05 mg/kg/day)
					Hemato	0.1			
					Hepatic	0.1			
					Neuro	0.03	0.07		Increased salivation in six females
Pizarro et al. 2001†									**Copper sulfate and Copper oxide**
7	HUMAN 45F	1 week daily (W)	0, 0.1	BI CS	Gastro		0.1		Nausea, vomiting, and/or abdominal pain
					Hepatic	0.1			
Alharbi et al. 2018									**Copper sulfate**
8	RAT (albino) 10F	7 days daily (IN)	0, 119	BC BI HP	Renal			119	Destroyed glomeruli corpuscles, hyperplasia of the epithelial cells lining the partial layer of Bowman's capsule, and destroyed epithelial lining of the proximal and distal convoluted tubules
Alhusaini et al. 2018a									**Copper sulfate**
9	RAT (Albino) 6M	7 days daily	0, 119	BC BI HP	Hepatic			119	Indications of liver inflammation; elevated hepatic ALT (+299%), NO (+68%), MDA (+44%), and caspase-3 (+>350%); decreased hepatic GSH (55%), SOD (80%) and IL-10 (>45%)

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Alhusaini et al. 2018b									**Copper sulfate**
10	RAT (Albino) 8M	7 days daily	0, 39.8	BI HP OW	Hepatic			39.8	Massive cellular degeneration and hepatocyte necrosis, unspecified reduction in relative liver weight; elevated AST (25%), ALT (>1000%), LDH (>130%), CRP (500%), hepatic MDA (43%), hepatic NO (62%) and protein expression of COX-2 (>500%); significantly lower hepatic GSH (>50%), and SOD (47%)
Rush 1990a									**Copper metal**
11	RAT (Sprague-Dawley) 5M, 5F	Once (NS)	24, 48, 80, 400	LX	Death			37 F	LD50
								42 M	LD50
Yamamoto et al. 2004									**Copper sulfate**
12	RAT (Wistar) NS	once (G)	0, 2.5, 10	CS, FI	Gastro	10			
					Other noncancer	2.5	10		Induced kaolin ingestion behavior (pica behavior)
Babaei et al. 2012									**Copper sulfate**
13	MOUSE (NMRI) 6F	14 days daily (G)	0, 39.8, 79.6	BC HP	Repro			39.8	Decreased number of antral follicles (45%) and ovarian cell damage

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Kadammattil et al. 2018									**Copper sulfate**
14	MOUSE (Swiss albino) 5NS	7 days daily	0, 0.4, 1, 2, 4	BC BI HP	Gastro	2		4	Intestine showing focal ulceration
					Hepatic	2	4		Increased serum PCC and lower cellularity and hemorrhage in liver
					Renal	4			
					Immuno	2	4		Follicular hyperplasia in spleen
					Neuro	4			
Kadammattil et al. 2018									**Copper metal**
15	MOUSE (Swiss albino) 2NS	Once	39.8	LE	Death			39.8	LD50
Kadammattil et al. 2018									**Copper metal**
16	MOUSE (Swiss albino) M NS	Once	0, 4.0	RX	Repro			4	Increased frequency of folded sperm (morphological anomaly)
Kadammattil et al. 2018									**Copper sulfate**
17	MOUSE (Swiss albino) F NS	7 days daily (day 7 to 12 of pregnancy)	0, 4.0	DX RX	Repro	4			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Rush 1990c									**Copper metal**
18	RATS (Sprague-Dawley) 5M, 5F	Once (NS)	40, 60, 80, 400	LX	Death			118 F	LD50
								94 M	LD50
Yamamoto et al. 2004									**Copper sulfate**
19	SHREW (S. murinus) 4F	once (G)	0, 2.5, 31	CS FI	Gastro	2.5	31		15 episodes of emesis in 4/4 animals
					Other noncancer	31			No altered food consumption
INTERMEDIATE EXPOSURE									
Araya et al. 2003b**									**Copper sulfate**
20	HUMAN 327-355B	2 months Daily (W)	0, 0.042, 0.091, 0.17	BC BI CS	Gastro	0.09	0.17		Gastrointestinal symptoms reported by 19.7% of subjects
					Hepatic	0.17			
Araya et al. 2004**									**Copper sulfate**
21	HUMAN 327-355B	2 months daily (W)	0, 0.055, 0.106, 0.169	CS WI	Gastro	0.06	0.11		Gastrointestinal symptoms in 65/355 subjects
O'Connor et al. 2003†									**Copper sulfate**
22	HUMAN 11M, 11F	6 weeks daily (F)	M: 0.018, 0.058 F: 0.017, 0.067	BC BI BW	Hepatic	0.07 F			
						0.06 M			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Olivares et al. 1998									**Copper sulfate**
23	HUMAN 39-50B	9 months daily (W)	0.0378-0.158, 0.0522-0.319	BC BW CS	Bd wt	0.32			
					Gastro	0.32			
					Hepatic	0.32			
Pratt et al. 1985**									**Copper gluconate**
24	HUMAN 3M, 4F	12 weeks (C)	0, 0.15	BC	Gastro	0.15			
					Hemato	0.15			
					Hepatic	0.15			
Turnlund et al. 2004†									**Copper metal**
25	HUMAN 9M	18 days daily (F)	0.02	BC BI IX UR	Immuno	0.02			
Turnlund et al. 2004†									**Copper metal**
26	HUMAN 9M	18 days daily (F)	0.1	BC BI IX UR	Immuno		0.1		Significantly reduced antibody titer against influenza strain compared to controls; 47-fold increase in the antibody in controls, 14-fold in exposed group
Abe et al. 2008									**Copper gluconate**
27	RAT (Fischer-344) 6-8M	6 weeks daily (F)	0, 62	BW HP	Bd wt	62			
					Hepatic	62			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Arafa et al. 2019									**Copper sulfate**
28	RAT (Wistar) 10M	90 days daily (G)	0, 50.9	BI CS HP	Cardio		50.9		33% increase in systolic blood pressure after 3 months
					Repro			50.9	44%, 84%, and 79% reduction in relative testicular weight, serum testosterone level, and serum LH level, respectively; changes of protein expression in testes
Babaei and Abshenas 2013									**Copper sulfate**
29	RAT (Sprague-Dawley) 12M	56 days daily (G)	0, 79.6	HP OW RX	Repro			79.6	13.5% reduction in testicular weight after 56 days and decreased sperm count, percentage of live spermatozoa and sperm motility ($p<0.001$)
Behzadfar et al. 2017									**Copper sulfate**
30	RAT (Wistar) 7M	21 days daily (W)	0, 19.9, 39.8, 79.6	GN HP NX	Neuro	19.9		39.8	Spatial memory impairment as measured by the Morris water maze test; 52% increase in hippocampal mitochondria lipid peroxidation (MDA formation) and 29% decrease in glutathione
DeVries et al. 1986									**Copper sulfate**
31	RAT (Sprague-Dawley) 8F	11 months daily (W)	0, 46	BI	Neuro		46		25% decrease of 3,4-dihydroxyphenylacetic acid levels in corpus striatum

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Epstein et al. 1982									**Copper acetate**
32	RAT (Sprague-Dawley) 8M	90 days daily (W)	0, 8.6	BW WI BC	Bd wt	8.6			
					Hepatic		8.6		>100% increase in aspartate aminotransferase activity
Haddad et al. 1991									**Copper acetate**
33	RAT (Wistar) 20-42F	60-73 days (W)	0, 130	BI BW DX HP RX	Bd wt	130			
					Hepatic		130		Histological changes in liver including degenerated hepatocytes and focal necrosis
					Renal		130		Histological changes in kidney including cloudy swelling in proximal convoluted tubules
					Develop		130		Delayed growth and development
Hashish and Elgaml 2016									**Copper sulfate**
34	RAT (albino) 8F	30 days daily (F)	0, 1.6	BC BI HP	Hepatic		1.6		Acute cell swelling of hepatocytes and karyolysis of nuclei, mild hyperplasia of portal area lining epithelium of bile ducts

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Renal		1.6		Degeneration of renal tissues and degeneration in lining epithelium of some renal tubules; decreased total protein by >32%, increased urea and creatinine by 50% and >65%; decreased renal CAT, SOD and GSH by 24%, >18, >13% and increased renal MDA by 48%
Kalita et al. 2020									**Copper metal**
35	RAT (Wistar) 6M	1 month daily (G)	0, 25.5	BI BW HP NX	Bd wt	25.5			
					Neuro			25.5	Changes in locomotor activity including reduced distance traveled, time moving, grip strength and reduced latency to fall time on the rotarod test and increased time resting; increased expression of GFAP and caspase-3 in corpus striatum indicating apoptosis
Khushboo et al. 2018									**Copper sulfate**
36	RAT (Wistar) 5M	30 days daily (G)	0, 50.9	BI BW FI HP OW RX UR	Bd wt	50.9			
					Cardio			50.9	Flabby, enlarged, and congested heart
					Gastro			50.9	Thickened stomach wall with corrugated mucosa

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Hepatic			50.9	Enlargement of liver with dark spots and borders swollen, friable and yellow in color; decreased relative liver weight by 30%
					Renal			50.9	Bilateral enlargement of kidney with a dark brown color; decrease in relative kidney weight by 30% and increased urea (+70%) and creatinine (+67%)
					Dermal		50.9		Rough dried skin with alopecia especially in the abdominal region
					Ocular		50.9		Paleness of mucous membranes of eyes and pads extremities
					Immuno		50.9		Congested and enlarged spleen
					Neuro			50.9	Swollen, congested, and edematous brain; slow activity
					Repro			50.9	>30% increase in sperm head abnormalities and >14% increase in sperm tail abnormalities; degeneration of epididymides, disrupted spermatogenesis, irreversible histological changes in testes, tubular and testicular degeneration; decreased relative weight of testes
					Other noncancer		50.9		Reduced food consumption and water intake by 29% and 41%

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Kumar and Sharma 1987									**Copper sulfate**
37	RAT (Albino) 15M	30 days daily (G)	0, 39.8	BC BI BW	Hemato		39.8		Anemia as evidenced by significant reduction in RBCs (52%) and hemoglobin levels (47%)
					Hepatic		39.8		Increased glucose (18%), cholesterol (34%), bilirubin (66%), serum ALT (308%), and decreased total protein levels (60%)
					Renal		39.8		Increased urea levels (161%) indicating kidney damage
Kumar et al. 2015									**Copper sulfate**
38	RAT (Wistar) 18M	30, 60, or 90 days daily (G)	0, 25.5, 50.9	BC BW NX OF	Bd wt			25.5	21.5% decrease in body weight at 90 days
					Hemato		25.5		Reduced hemoglobin by 9-21% at 30, 60, and 90 days
					Hepatic		25.5		Increased ALT, AST, and bilirubin by 190%, 423% and 488%, respectively at 90 days
					Renal		25.5		Increased BUN and BUN/creatinine ratio of 49% and 67%, respectively after 90 days
					Neuro			25.5	Impaired motor coordination and cognitive function including grip strength, latency to fall time, and attention scores

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Kumar et al. 2016a									**Copper sulfate**
39	RAT (Wistar) 18M	30, 60 or 90 days daily (G)	0, 39.8, 79.6	BI BW HP NX	Bd wt		39.8		Unspecified significant weight loss
					Hepatic			39.8	Hepatocellular degeneration and hemorrhage, massive fatty change and centrilobular necrosis, occasional hepatic cell necrosis
					Renal			39.8	Hemorrhage, inflammatory and cellular damage in kidneys, and degeneration of renal intertubular space and Bowmen's capsule
					Neuro		39.8	79.6	LOAEL: gliosis, pyknotic nuclei, and glial nodule formation SLOAEL: Neuronal loss and depleted myelin
Kumar et al. 2016b									**Copper sulfate**
40	RAT (Wistar) 18M	30, 60 or 90 days daily (G)	0, 39.8, 79.6	BC BW HE HP	Bd wt		39.8		Unspecified significant reduction in body weight
					Hepatic		39.8		Reduced TAC (19-33%) and GSH (14-41%), and increased MDA ($p<0.001$) after 30, 60, 90 days
					Renal		39.8		Reduced TAC (14-26%) and GSH (18-48%), and increased MDA ($p<0.001$) after 30, 60, 90 days

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Neuro			39.8	Changes in grip-strength, rotarod test, and Y-maze test; reduced TAC, GSH, and increased MDA ($p<0.01$)
Kumar et al. 2019									**Copper sulfate**
41	RAT (Sprague-Dawley) 5M	16 weeks daily (G)	0, 4.0, 8.0	BC, BW, CS, NX	Bd wt	8			
					Musc/skel		4		Impaired muscle strength in rotarod test
					Neuro			4	Decreased locomotor activity and neuromuscular coordination, reduced catalase and SOD activity in brain tissues ($p<0.0001$), decreased passive avoidance response, less exploration time
Liu et al. 2016									**Copper sulfate**
42	RAT (Wistar) 10M	30 days daily (G)	0, 39.8, 79.6, 159	HP OW RX	Repro		39.8	79.6	LOAEL: Decreased sperm count (14%); 15% and 13% decrease in LH and FSH SLOAEL: Significant 62% reduction in sperm count and 47% increase in sperm malformation rate; significant reduction in testosterone, FSH and LH, by 43%, 23%, and 21%, respectively

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
NTP 1993									**Copper sulfate**
43	RAT (Fischer-344) 5M,5F	15 days daily (W)	M: 0, 10, 29, 36, 45, 96; F: 0, 10, 26, 31, 71	BW CS GN HP WI	Death			71 F	100% mortality
								45 M	100% mortality
					Bd wt	26 F	31 F		16% weight loss
						36 M			
					Resp	31 F			
						36 M			
					Cardio	31 F			
						36 M			
					Gastro	31 F			
						36 M			
					Hepatic	31 F			
						29 M			
					Renal	31 F			
							10 M		Protein droplets in epithelial cells of proximal tubule
					Endocr	31 F			
						36 M			
					Immuno	31 F			
						36 M			
					Neuro	31 F			
						36 M			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Repro	31 F			
						36 M			
NTP 1993									**Copper sulfate**
44	RAT (Fischer-344) 5M,5F	15 days daily (F)	M: 0, 23, 46, 92, 198, 325; F: 0, 23, 44, 93, 196, 285	BW CS FI GN HP OW WI	Bd wt	285 F			
						325 M			
					Resp	285 F			
						325 M			
					Cardio	285 F			
						325 M			
					Gastro	23 F	44 F		Hyperplasia with hyperkeratosis of the squamous mucosa on the limiting ridge separating the forestomach from the glandular stomach
						23 M	46 M		Hyperplasia with hyperkeratosis of the squamous mucosa on the limiting ridge separating the forestomach from the glandular stomach
					Hemato	93 F	196 F		Depletion of hematopoietic cells in bone marrow
						325 M			
					Hepatic	285 F			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
						92 M	198 M		Minimal to mild mononuclear inflammatory cell infiltrate in 4/5 males
					Renal	285 F			
						46 M	92 M		Increased protein droplets in cortical tubules
					Endocr	285 F			
						325 M			
					Immuno	285 F			
						325 M			
					Neuro	285 F			
						325 M			
					Repro	285 F			
						325 M			
NTP 1993									**Copper sulfate**
45	RAT (Fischer-344) 10M,10F	13 weeks daily (F)	M: 0, 8, 16, 33, 66, 140 F: 0, 9, 17, 34, 68, 134	BC BI CS GN HP OW UR	Bd wt	134 F			
						66 M		140 M	24% decrease in bodyweight by end of experiment
					Resp	134 F			
						140 M			
					Cardio	134 F			
						140 M			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Gastro	17 F	34 F		In 7/10 females, hyperplasia of the limiting ridge that forms the junction of the forestomach squamous mucosa with the glandular gastric mucosa
						16 M	33 M		In 10/10 males, hyperplasia of the limiting ridge that forms the junction of the forestomach squamous mucosa with the glandular gastric mucosa
					Hemato	134 F			
						33 M	66 M		Decreases in hematocrit, hemoglobin, reticulocytes, mean cell volume, and mean cell hemoglobin levels and increases in platelet levels
					Hepatic	34 F	68 F		Chronic active inflammation in liver of 6/10 females at 68 mg Cu/kg/day
						16 M	33 M		Chronic active inflammation with focal necrosis in 1/10 males; 112% increase in serum alanine aminotransferase
					Renal	9 F	17 F		Increased BUN by 15% and cytoplasmic alteration in kidneys of 1/10 females
						16 M	33 M		Cytoplasmic alteration in kidneys of 3/10 males
					Endocr	134 F			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
						140 M			
					Neuro	68 F	134 F		Gliosis in brain in 10/10 rats
						66 M	140 M		27% increase in relative brain weight
					Repro	68 F		134 F	Chronic active inflammation of clitoral gland and ovarian cysts in 10/10 rats
						66 M	140 M		Significant 27% increase in relative right testis weight
Rana and Kumar 1980									**Copper sulfate**
46	RAT (Albino) 10M	20 days daily (G)	0, 39.8	BC BW CS GN HP	Bd wt			39.8	>28% lower body weight
					Hemato		39.8		Decreased erythrocyte, hemoglobin, and hematocrit levels by 48%, 38%, and 39%, respectively
					Musc/skel		39.8		Depressed skeletal growth assessed by tail length
					Hepatic		39.8		Centrilobular necrosis and perilobular sclerosis with nuclear edema in liver
					Renal		39.8		Engorgement of uriniferous tubules, necrosis of the tubules, nuclear pyknosis and cell proliferation in medullary region
					Other noncancer		39.8		Change in paw color from pink to white

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Sakhaee et al. 2012									**Copper sulfate**
47	RAT (Wistar) 20M	8 weeks daily (G)	0, 39.8, 79.6	BC BI HP RX	Hepatic			39.8	Hepatic lesions including cell swelling in hepatocytes, centrilobular hepatocellular necrosis, mild bile retention, multifocal hepatitis and presence of apoptotic bodies
					Renal			39.8	Renal lesions with mild tubular necrosis and hyaline cast formation in renal tubules
					Repro			39.8	54%, 48%, and 60% decrease in sperm concentration, motility, and viability, respectively
Seven et al. 2018									**Copper sulfate**
48	RAT (Sprague-Dawley) 6M	21 days daily (G)	0, 199	BI, BW, HP	Bd wt	199			
					Hepatic			199	Degenerative and necrotic changes in the liver
					Renal			199	Kidneys showed degeneration and necrosis of mostly the proximal and a minority of distal tubules in the cortex
					Immuno		199		Increased serum tumor necrosis factor alpha (TNF-α) level, 1.55 times over control levels
					Other noncancer		199		21% decrease in food consumption

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Tian et al. 2019									**Copper sulfate**
49	RAT (Sprague-Dawley) 10M	30 days daily (W)	0, 79.6	BI BW HP OW	Bd wt		79.6		10-15% decrease in body weight and reduced body weight gain
					Hepatic		79.6		Significantly increased AST and ALT by 67% and 70%, respectively
Babaei et al. 2012									**Copper sulfate**
50	MOUSE (NMRI) 6F	35 days daily (G)	0, 39.8, 79.6 mg/kg/day	BC HP	Repro			39.8	Significant decrease in number of ovarian follicles (>80%) and corpus luteum (88%), and ovarian cell damage
Cheng et al. 2020									**Copper chloride**
51	MOUSE (Kunming) 12F	90 days daily (W)	0, 2.4	BW HP	Bd wt	2.4			
					Gastro			2.4	Increased histological lesions of cecum and rectum including increased thickness of outer muscularis, widened submucosa, and severe atrophy of central lacteal; changes in rectal microbial gut bacteria composition

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Kheirandish et al. 2014									**Copper sulfate**
52	MOUSE (NMRI) 15M	56 days daily (G)	0, 79.6	GN HP	Repro			79.6	Shrinkage of seminiferous tubules and moderate to severe degeneration of germinal layers, significantly decreased Sertoli cell nuclei diameter, and epithelial height and significantly less meiotic index and spermatogenesis
Kvietkauskaite et al. 2004									**Copper sulfate**
53	MOUSE (BALB/c) 10M	19 weeks ad libitum (W)	0, 5.6, 10.7	BC BI BW HE HP OW	Bd wt	5.6	10.7		10.3% reduction in body weight
					Hemato	10.7			
					Hepatic		5.6		13.6% decrease in total liver protein
					Immuno		5.6		Decreased percent of natural killer (CD4*CD8) and suppressor (CD8*CD4) cells and altered immunoregulatory index
Lu et al. 2009									**Copper sulfate**
54	MOUSE (Kunming) 8M	16 weeks daily (W)	0, 0.08	HP, NX	Neuro	0.08			
NTP 1993									**Copper sulfate**
55	MOUSE (B6C3F1) 5M,5F	15 days daily (W)	M: 0, 10, 24, 58, 133, 367; F: 0, 15, 36, 62, 174, 330	BW GN HP WI	Death			62 F	3/5 died
								58 M	1/5 died

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Bd wt	36 F		62 F	27% weight loss in 2/5 mice
						24 M	58 M		16% weight loss
					Resp	36 F			
						24 M			
					Cardio	36 F			
						24 M			
					Gastro	36 F			
						24 M			
					Hepatic	36 F			
						24 M			
					Renal	36 F			
						24 M			
					Endocr	36 F			
						24 M			
					Neuro	36 F			
						24 M			
					Repro	36 F			
						24 M			
NTP 1993									**Copper sulfate**
56	MOUSE (B6C3F1) 5M,5F	15 days daily (F)	M: 0, 43, 92, 197, 294, 717; F: 0, 53, 104, 216, 398, 781	BW CS FI GN HP OW WI	Bd wt	781 F			
						717 M			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Resp	781 F			
						717 M			
					Cardio	781 F			
						717 M			
					Gastro	104 F	216 F		2/5 females had minimal hyperplasia with hyperkeratosis of the squamous mucosa on the limiting ridge of the forestomach at its junction with the glandular gastric mucosa
						92 M	197 M		3/5 males had minimal hyperplasia with hyperkeratosis of the squamous mucosa on the limiting ridge of the forestomach at its junction with the glandular gastric mucosa
					Hepatic	781 F			
						197 M			
					Renal	781 F			
						717 M			
					Neuro	398 F	781 F		Unspecified significant increase in relative brain weight
						197 M	294 M		Unspecified significant increase in relative brain weight

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
NTP 1993									**Copper sulfate**
57	MOUSE (B6C3F1) 10M,10F	13 weeks daily (F)	M: 0, 44, 97, 187, 398, 815; F: 0, 52, 126, 267, 536, 1058	BW CS FI GN HP OW	Bd wt	267 F	536 F	1,058 F	LOAEL: 13% decrease in bodyweight SLOAEL: 25% decrease in bodyweight
						97 M	187 M	815 M	LOAEL: 11% decrease in bodyweight SLOAEL: 21% decrease in bodyweight
					Resp	1,058 F			
						815 M			
					Cardio	1,058 F			
						815 M			
					Gastro	126 F	267 F		In 5/10 females, hyperplasia of forestomach mucosa
						97 M	187 M		In 2/10 males, hyperplasia of forestomach mucosa
					Hepatic	1,058 F			
						815 M			
					Renal	1,058 F			
						815 M			
					Endocr	1,058 F			
						815 M			
					Neuro	126 F	267 F		10% increase in relative brain weight
						97 M	187 M		13% increase in relative brain weight

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Repro	536 F	1,058 F		cyst in clitoral gland in 8/10 rats
						97 M	187 M		12% increase in relative right testis weight
Sakhaee et al. 2014									**Copper sulfate**
58	MOUSE (NMRI) 12M	42 days daily (G)	0, 79.6	BC BI GN HP	Hepatic		79.6		AST and ALT significantly elevated ($p<0.05$) 2.8 and 3.6 times greater than controls
					Repro			79.6	Significantly decreased sperm concentration, motility, and viability by 56%, 71%, and 67%, respectively, and degenerative changes of the seminiferous tubules
Sakhaee et al. 2016a									**Copper sulfate**
59	MOUSE NMRI 6M	28 days once every 2 days (GW)	0, 39.8	HP RX	Repro			39.8	60%, 37%, and 41% decrease of sperm count, motility, and vitality, respectively; depletion and vacuolation of seminiferous epithelium
Sakhaee et al. 2016a									**Copper sulfate**
60	MOUSE NMRI 6M	42 days once every 2 days (GW)	0, 39.8	HP RX	Repro			39.8	61%, 39%, and 39% decrease of sperm count, motility, and vitality, respectively; degeneration of the seminiferous tubules

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Sakhaee et al. 2016b									**Copper sulfate**
61	MOUSE (NMRI) 6M	42 days ad libitum (W)	0, 39.8	HP RX	Repro			39.8	48%, 43% and 43% decrease in sperm count, motility, and viability, respectively, and disorganization and vacuolation of seminiferous epithelium
Wu et al. 2020									**Copper sulfate**
62	MOUSE (ICR) 60NS	42 days daily (G)	0, 4, 8, 16	HP	Hepatic			4	Increased incidence of granular and vacuolar degeneration in hepatocytes and increased rate of hepatic apoptosis
Seffner et al. 1997									**Copper metal**
63	GN PIG (albino) 5-8NS	6 months daily (IN)	<1.04, 6.6 for 4 weeks then 9.6 for	BI BW HE HP OW	Bd wt	9.6			
					Hepatic	9.6			
					Renal	9.6			
					Endocr	9.6			
					Immuno	9.6			
					Neuro	9.6			
Munley 2003a									**Copper hydroxide**
64	RABBIT (New Zealand) 5F	23 days daily (G)	0, 7.5, 15, 30	BW FI LX	Death			30	2/5 rabbits
					Bd wt	30			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Munley 2003a									**Copper hydroxide**
65	RABBIT (New Zealand) 5-9F	GD 7-28 daily (G)	BW CS DX FI GN HP RX	0, 7.5, 15, 30	Death			30	2/8 rabbits
					Bd wt	30			
					Resp	30			
					Gastro		7.5		Diarrhea in 1/8 rabbits
					Hepatic	30			
					Renal	15		30	One animal died from hemolytic event causing hemoglobin nephropathy and likely renal failure; other dead animal had moderately autolyzed small liver
					Endocr	30			
					Immuno	30			
					Repro	15		30	Slight increase in fetal resorptions, total mean per litter of 1.3 resorptions compared to 0.3 resorptions in controls
					Other noncancer	7.5	15		22% reduction in mean food consumption
Munley 2003a									**Copper hydroxide**
66	RABBIT (New Zealand) 29-64 M,F	GD 7-28 daily (G)	0, 7.5, 15, 30	DX GN	Develop	15		30	12% reduction in mean fetal weights; 4/29 rabbits with omphalocele (protrusion of intestines at the umbilicus)

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Munley 2003b									**Copper hydroxide**
67	RABBIT (New Zealand) 21-22F	GD 7-28 daily (G)	0, 6, 9, 18	BW CS FI GN LX OW RX	Death			18	3/22 rabbits
					Bd wt	18			
					Resp	9		18	Dark discoloration or mottling of lung tissue in 3/21 rabbits
					Gastro		6	18	LOAEL: Increased incidence of diarrhea (5/22 rabbits) SLOAEL: Stomach hemorrhage and/or ulceration in 3/21 rabbits; diarrhea in 2/21
					Hepatic	9		18	Pale liver in 3/21 rabbits
					Renal	18			
					Neuro	9	18		Weakness preceding death in one animal that died during exposure
					Repro	9		18	2 fetuses aborted on gestation day 22
					Other noncancer	6	9		Significant 17% decrease in food consumption
Munley 2003b									**Copper hydroxide**
68	RABBIT (New Zealand) 126-159 M,F	GD 7-28 daily (G)	0, 6, 9, 18	BW DX	Develop	9		18	Increased incidence of delayed skull ossification occurring in 5 fetuses among 2 litters and incidence of supernumerary ribs (110/126 fetuses); significant incidence of hemivertebra (2 fetuses)

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Shen et al. 2005									**Copper metal**
69	RABBIT (White Chinese Big Ear) 5M	50 days daily (W)	0, 16	BC BW HE	Bd wt	16			
					Hemato		16		Changes in blood composition including unspecified decreases in neutrophils, eosinophils, platelets, monophils, and basophils
					Hepatic		16		Unspecified increase of LDL and decrease of TG and VLDL
Kline et al. 1971									**Copper sulfate**
70	PIG (Hampshire-Yorkshire) 12NS	88 days ad libitum (F)	0.1, 1.7, 2.3, 2.7	BC BW FI HE	Bd wt	1.7	2.3		17% reduction in body weight gain
					Hemato	2.7			
Suttle and Mills 1966a									**Copper carbonate**
71	PIG (NS) 6F	46 days daily (F)	0, 16.5	BC BI BW	Bd wt			16.5	Decreased weight gain by 22%
					Hemato		16.5		28% decrease in hemoglobin levels
					Hepatic		16.5		Jaundice in 2 of 6 animals
Suttle and Mills 1966a									**Copper carbonate**
72	PIG (NS) 6F	49 days daily (F)	0, 18.7	BC BI BW	Bd wt			18.7	Decreased weight gain by 27%

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Hemato		18.7		25% decrease in hemoglobin levels measured at 6 weeks, and 12% increase in erythrocyte count
					Hepatic		18.7		Severe transient jaundice in 5 of 6 animals between weeks 3 and 6; aspartate aminotransferase activity elevated by >100%
CHRONIC EXPOSURE									
Araya et al. 2012									**Copper gluconate**
73	MONKEY (Tufted Capuchin) 2M, 2F	3 years daily (F)	0, 0.7 increased to 1.05 over first 2 months	BI BW CS FI HP OF	Bd wt	1.05			
					Hemato		1.05		Significantly lower hemoglobin (6%)
					Hepatic		1.05		300% increase in Ki67 positive cells indicative of tissue proliferation induction after 36 months
					Other noncancer	1.05			
Araya et al. 2012									**Copper gluconate**
74	MONKEY (Tufted Capuchin) 2M, 2F	3 years daily (Milk)	0, 0.49 increased to 0.77 over first 2 months	BI BW CS FI HP OF	Bd wt	0.77			
					Hemato	0.77			
					Hepatic	0.77			

Table 2-2. Levels of Significant Exposure to Copper – Oral (mg/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Other noncancer	0.77			
Massie and Aiello 1984									**Copper gluconate**
75	MOUSE (C57BL/6N) 8M	850 d (W)	0, 4.2, 8.5, 42	BW CS	Death			42	14.4% decrease in mean survival time and 12.8% decrease in maximum lifespan
					Bd wt	42			

[a]The number corresponds to entries in Figure 2-3.
[b]Used to derive a provisional acute oral minimal risk level of 0.02 mg/kg/day; the $BMDL_{10}$ of 0.05 mg/kg/day was divided by an uncertainty factor of 3 (3 for human variability). The acute oral minimal risk level was also adopted as the intermediate oral provisional minimal risk level.

**The previous EPA reference bodyweight for adults of 65kg was used to calculate the dose.
†The bodyweight reported by the study was used to calculate the dose.

To calculate the copper dose in studies using copper sulfate, it was assumed that the study used copper sulfate anhydrous (molar mass: 159.61 g/mol) unless otherwise specified that copper sulfate pentahydrate (molar mass: 249.69 g/mol) was used.

ALT = alanine aminotransferase; ALP = alkaline phosphatase; AST = aspartate aminotransferase; BC = serum (blood) chemistry; Bd wt = body weight; BI = biochemical indices; BUN = blood urea nitrogen; BW = body weight; Cardio = cardiovascular; COX-2 = cyclooxygenase 2; CRP = c-reactive protein; CS = clinical signs; Develop = developmental; DX = developmental toxicity; Endocr = endocrine; F= female(s); (F) = feed; FI = food intake; FSH = follicle stimulating hormone; (G) = gavage – not specified; Gastro = gastrointestinal; GFAP = glial fibrillary acidic protein; GGT = γ-glutamyl transferase; GN = gross necropsy; GSH= glutathione; (GW) = gavage in water vehicle; HE = hematological; Hemato = hematological; HP = histopathology; IL-10 = interleukin-10; Immuno = immunological; (IN) = ingestion; LD50 = dose producing 50% death; LDH = lactate dehydrogenase; LDL = low-density lipoprotein; LE = lethality; LH = luteinizing hormone; LOAEL = lowest-observed-adverse-effect-level; M = male(s); MDA = malondialdehyde; Musc/skel = musculo/skeletal; Neuro = neurological; NO = nitric oxide; NOAEL = no-observed-adverse-effect-level; NS = not specified; NX = neurological function; OF = organ function; OW = organ weight; PCC = protein carbonyl content; RBC = red blood cell(s); Repro = reproductive; Resp = respiratory; RX = reproductive toxicity; SOD = superoxide dismutase; TAC = total antioxidant capacity; TG = teratogenicity; UR = urinalysis; VLDL= very low-density lipoproteins; (W) = water; WI = water intake

Figure 2-3. Levels of Significant Exposure to Copper – Oral
Acute (≤14 days)

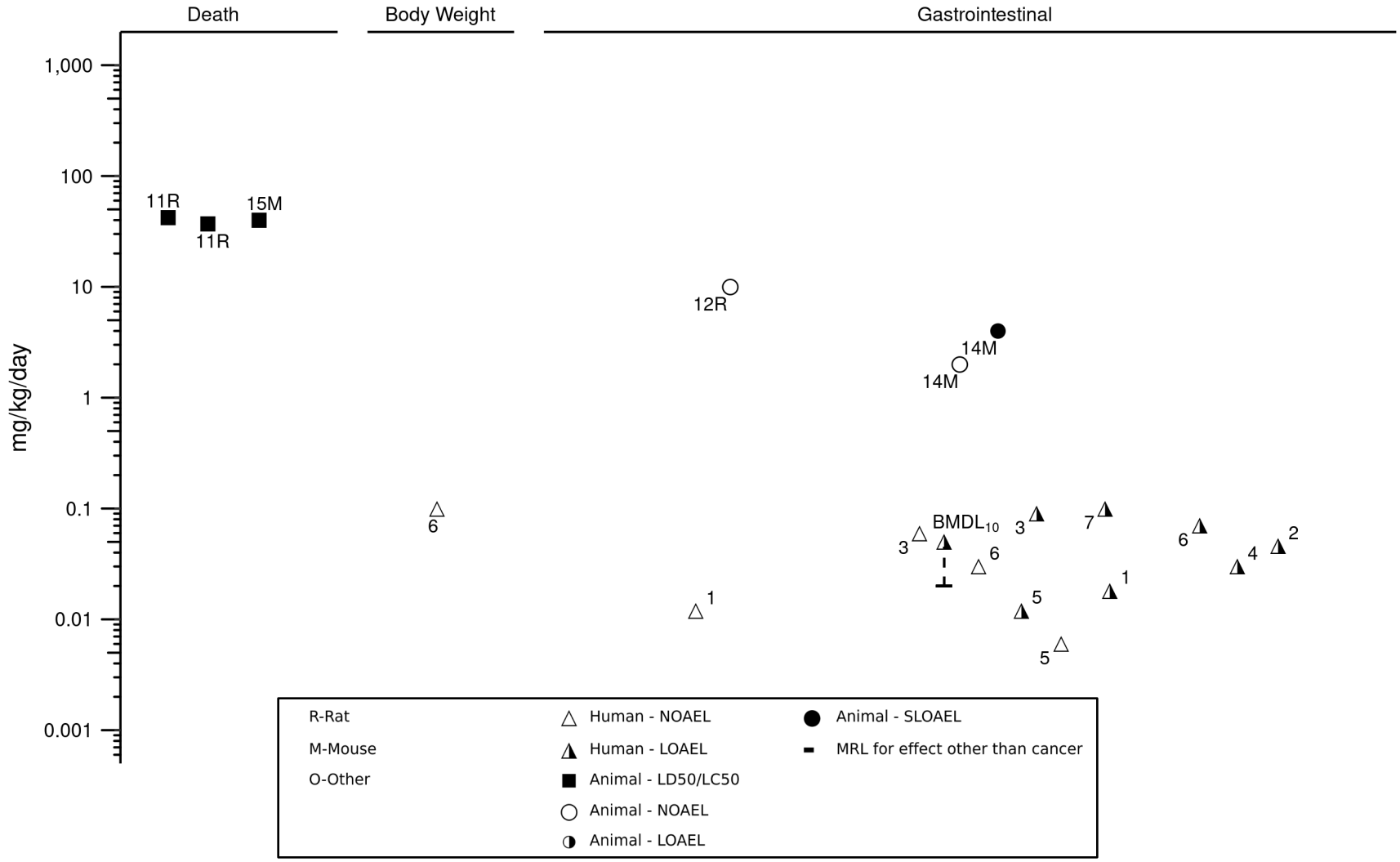


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Acute (≤14 days)

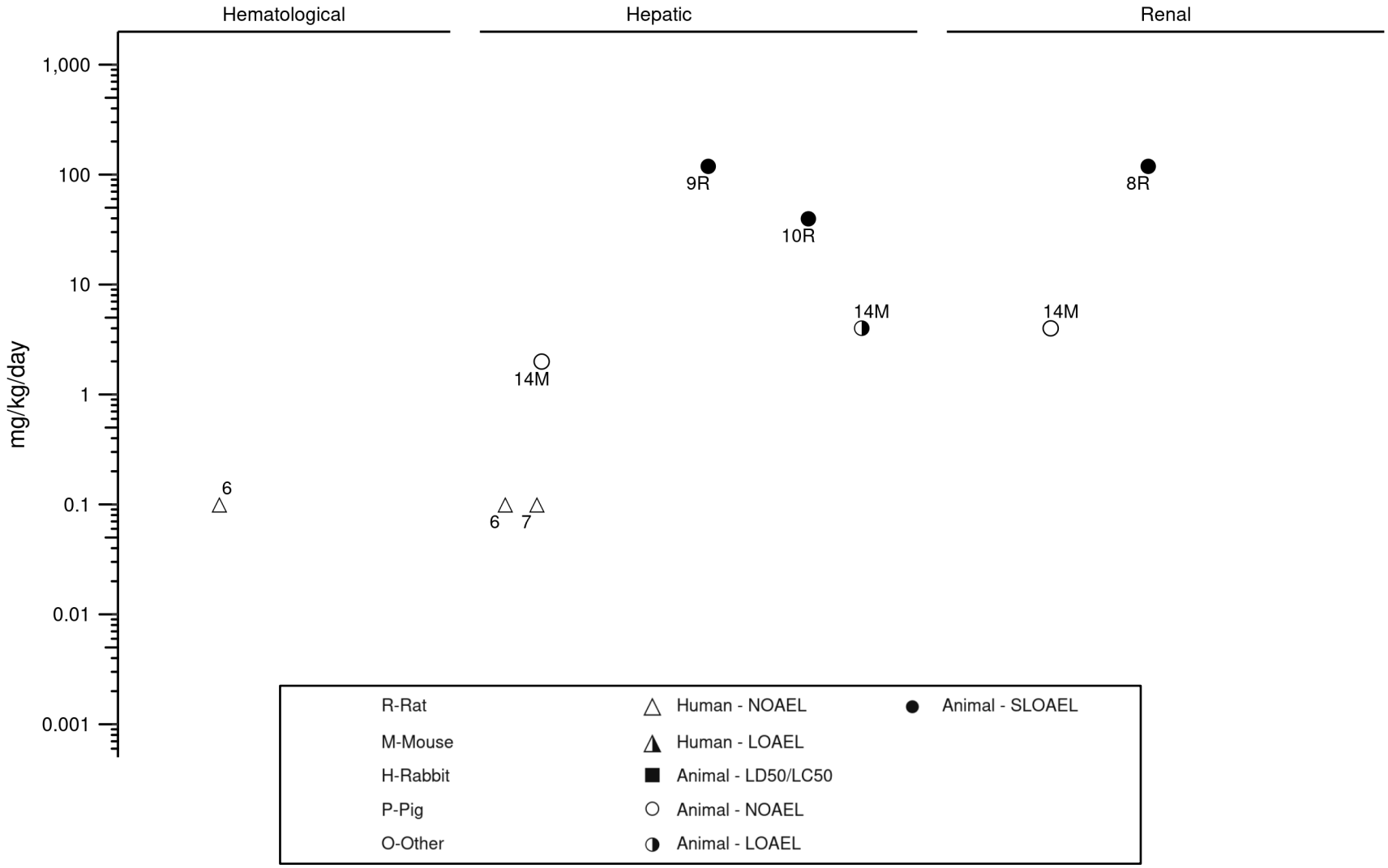


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Acute (≤14 days)

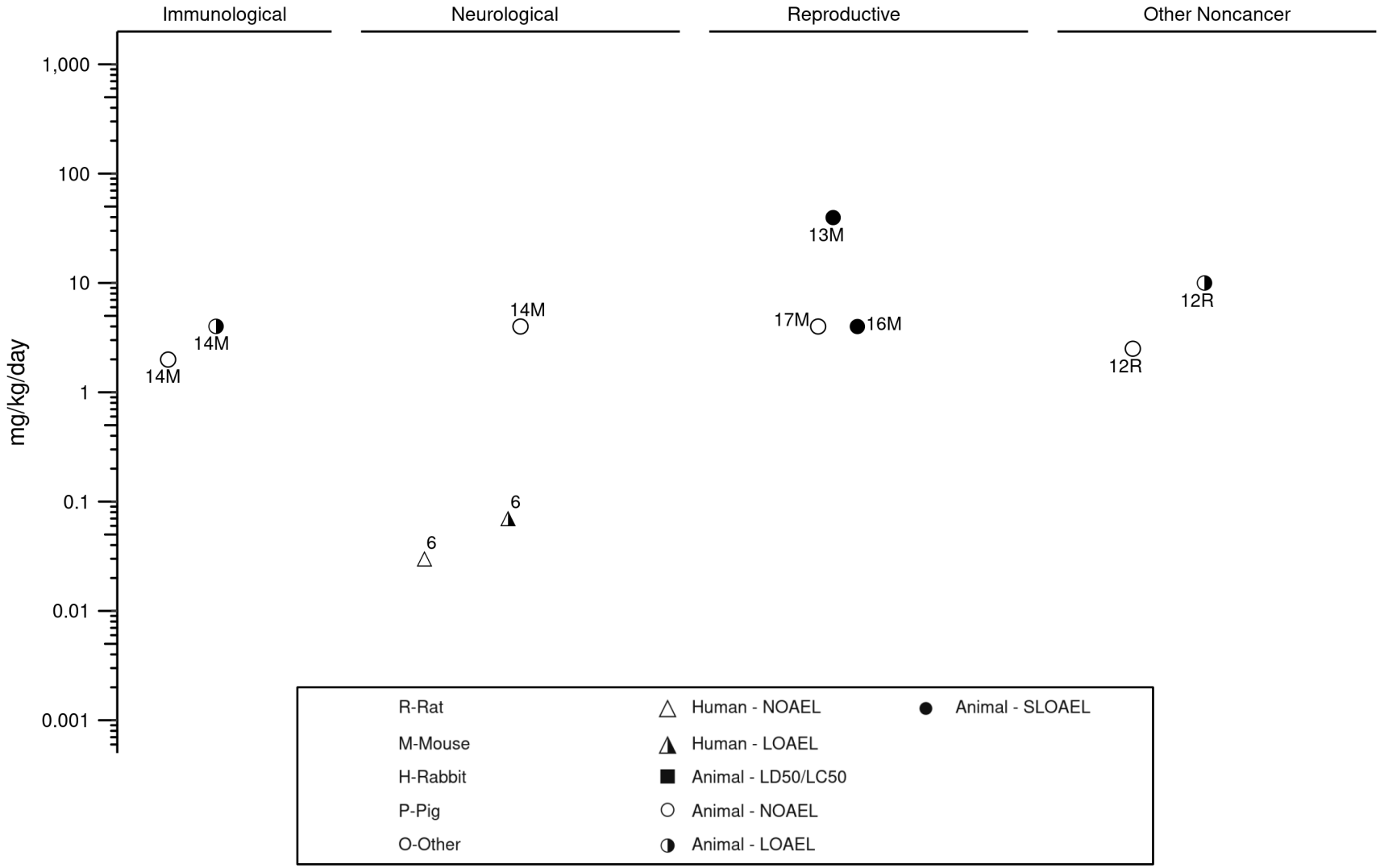


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

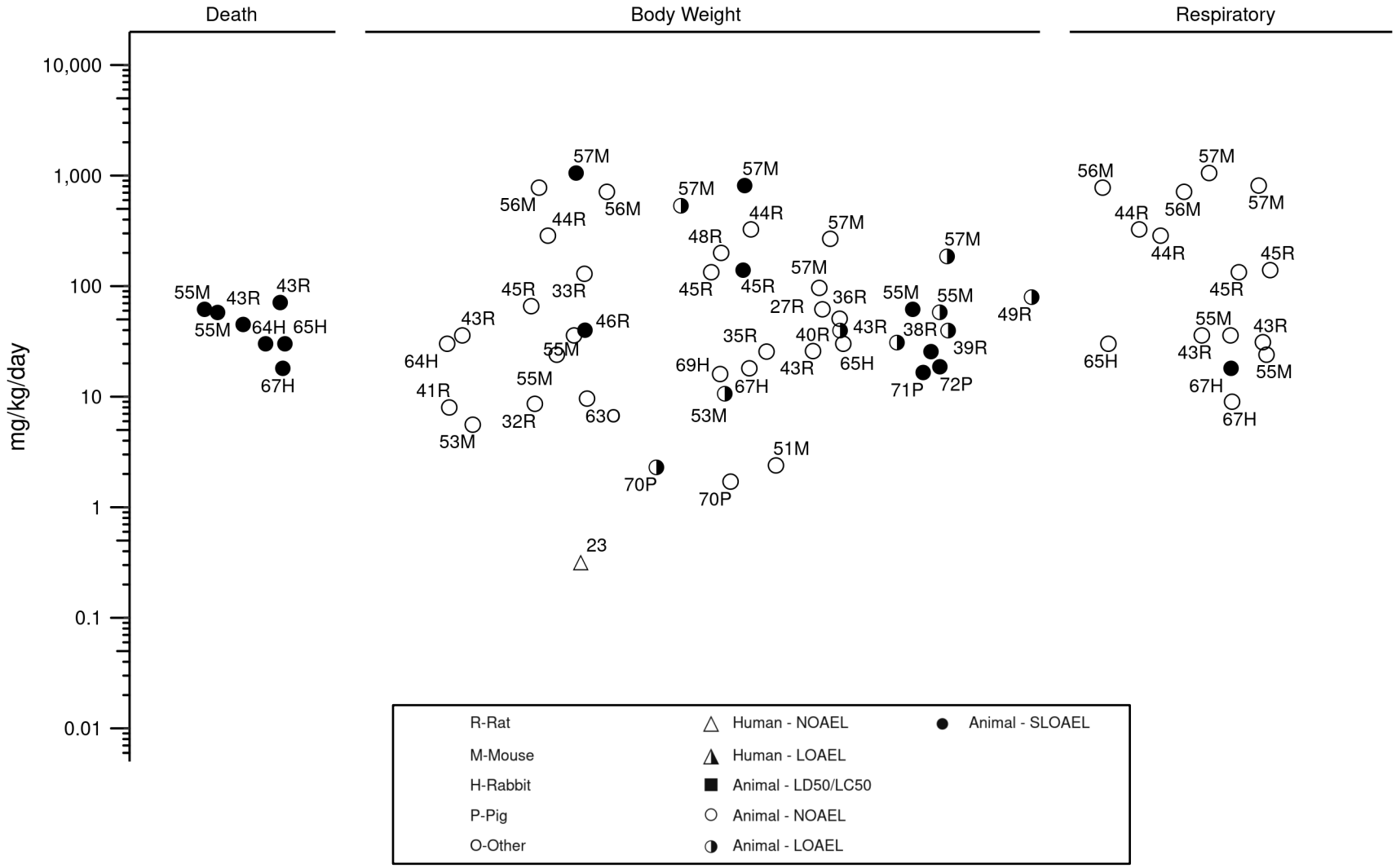


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

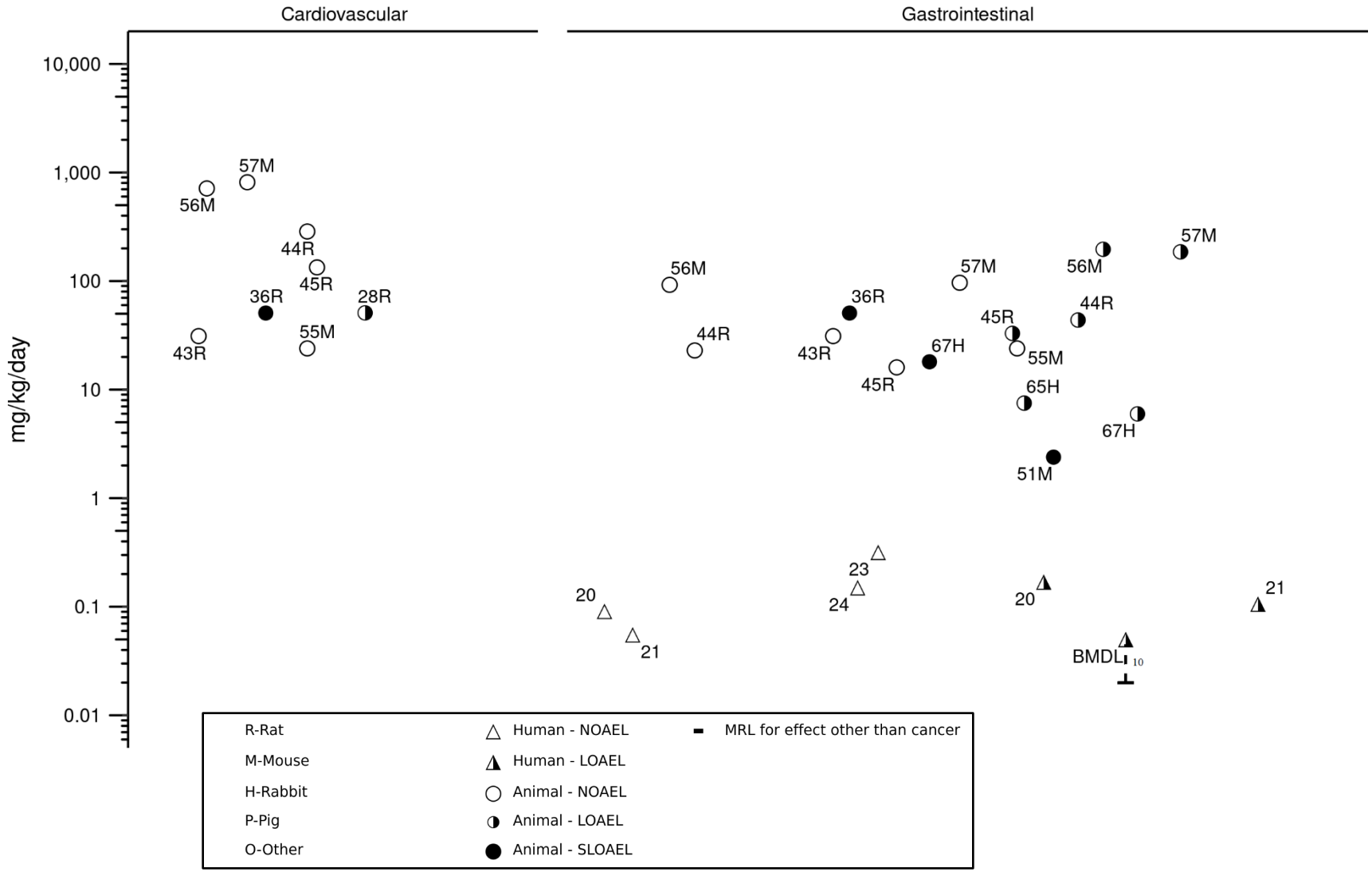


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

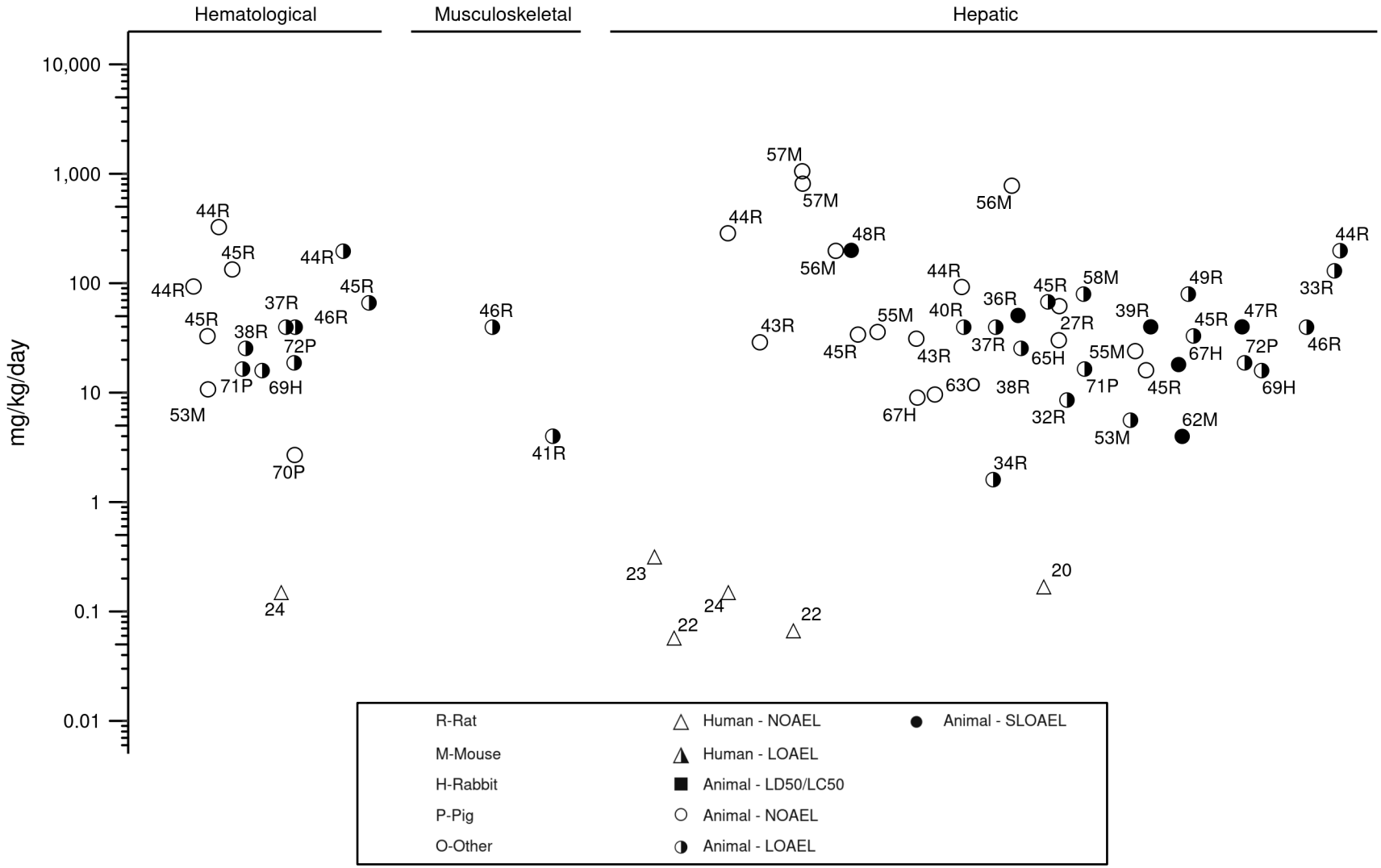


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

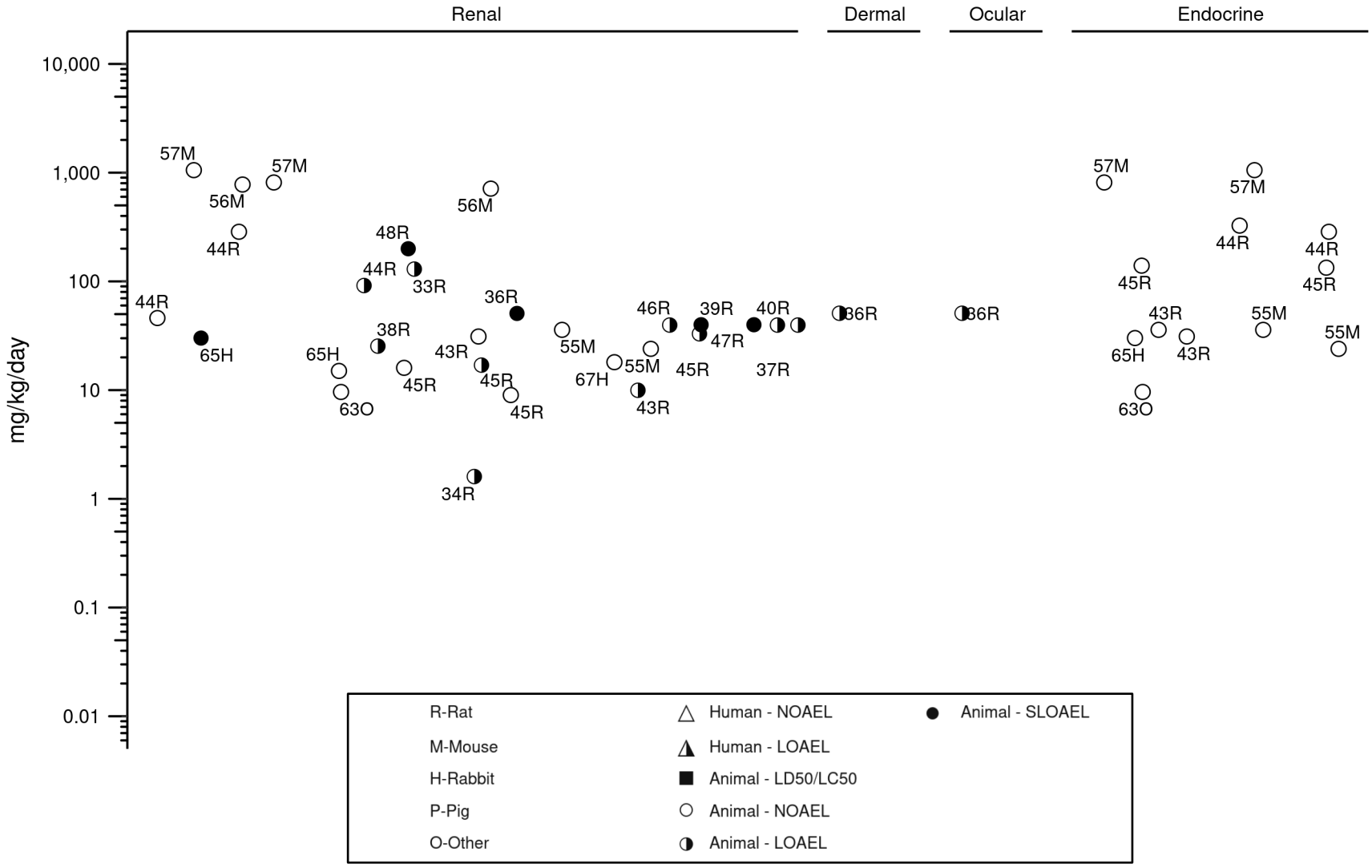


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

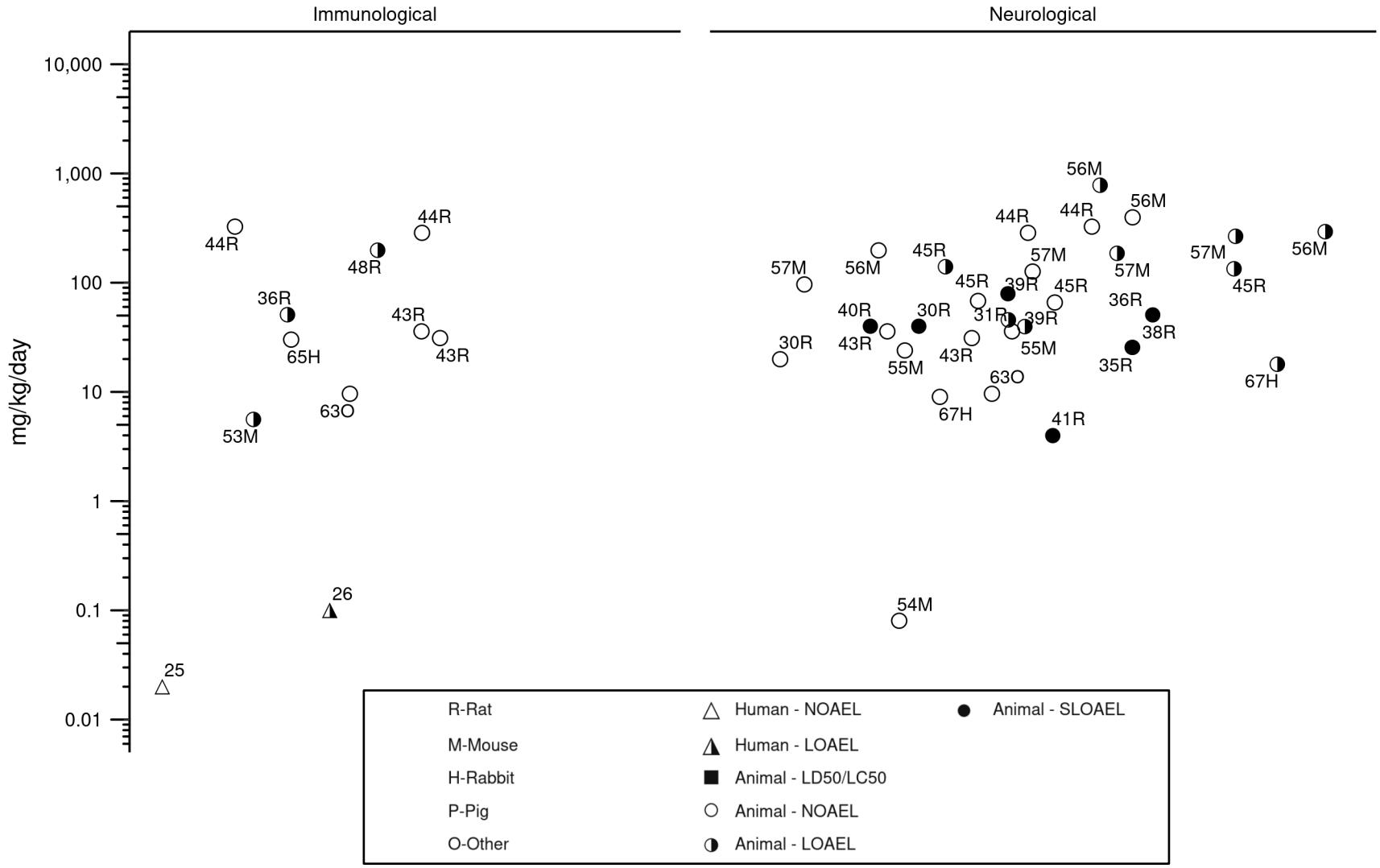


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Intermediate (15-364 days)

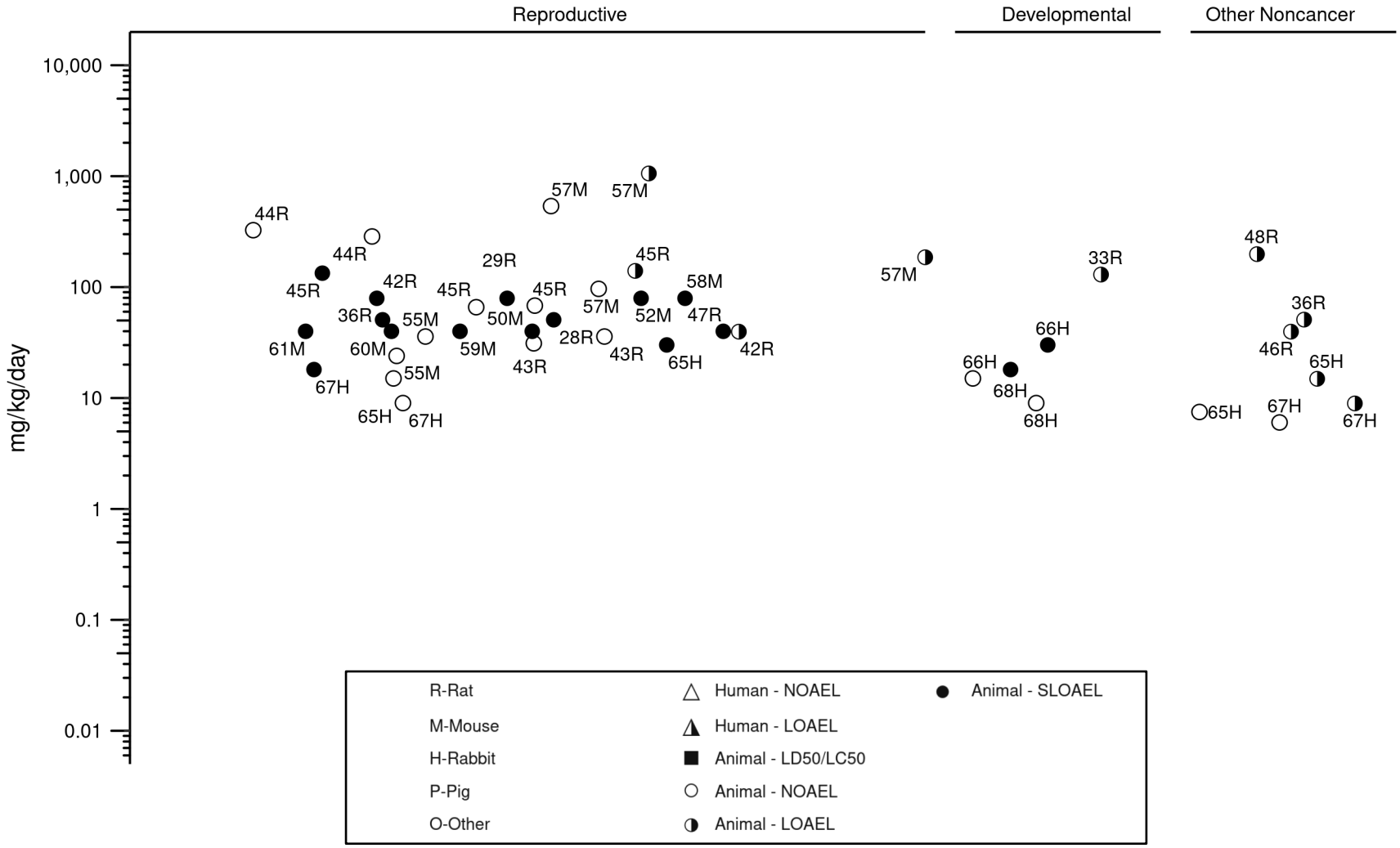


Figure 2-3. Levels of Significant Exposure to Copper – Oral
Chronic (≥365 days)

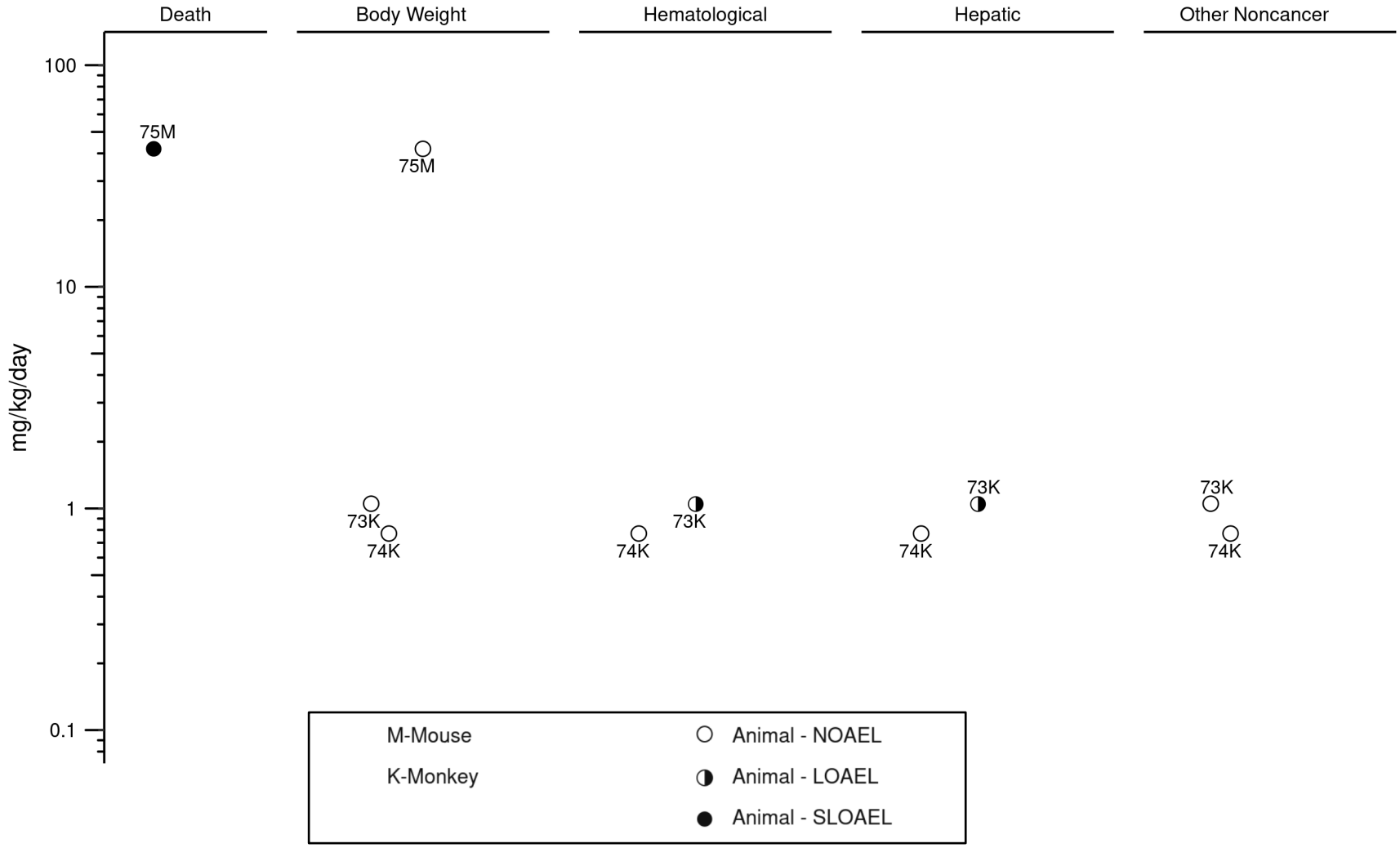


Table 2-3. Levels of Significant Exposure to Copper – Dermal

Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE								
Kuhn 1989b								**Copper oxide**
RABBIT (New Zealand) 5M, 5F	24 hours	1613	LX	Death			1,613	One rabbit died during the exposure period
Rush 1990b								**Copper metal**
RABBIT (New Zealand) 5M, 5F	24 hours	160	LX	Death	160			
INTERMEDIATE EXPOSURE								
Hagemann 1992								**Copper 8-Quinolinolate**
RAT (Albino) 5M, 5F	6 hours/day 5 days/week 4 weeks (NS)	0, 9, 36, 181	BC BW CS FI HE LX OW	Death	181			
				Bd wt	181			
				Resp	181			
				Cardio	181			
				Gastro	181			
				Hemato	181			
				Musc/skel	181			
				Hepatic	181			
				Renal	181			
				Dermal	181			
				Ocular	181			
				Endocr	181			
				Immuno	181 F			

Table 2-3. Levels of Significant Exposure to Copper – Dermal

Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					36 M	181 M		Increased occurrence of necrotic thymic lymphocytes found in higher incidence (5/5 males)
				Neuro	181			
				Repro	181			
				Other noncancer	181			

To calculate the copper concentration in studies using copper sulfate, it was assumed that the study used copper sulfate anhydrous (molar mass: 159.61 g/mol) unless otherwise specified that copper sulfate pentahydrate (molar mass: 249.69 g/mol) was used.

F = female(s); LE = lethality; LOAEL = lowest-observed-adverse-effect level; M = males(s); NOAEL = no-observed-adverse-effect level

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
Multiple Endpoints		
Suciu et al. 1981 **Study Type:** Cohort study of workers who worked in copper sieving operators in 1970 to 1973. The study examined 100, 97, 75, and 97 workers in 1970, 1971, 1972, and 1973, respectively. 51% of workers were 30-39 years of age. A control of 20 healthy non-exposed people were used only as reference for serum copper levels. Medical examinations were performed for the exposure cohort only, and serum copper was measured for both the exposure cohort and controls.	**Exposure:** Inhalation of copper. Copper concentration in air measured. In 1971, concentrations up to 464 mg Cu/m^3 were measured. In 1972, the measured maximum concentration was 132 mg Cu/m^3 and in 1973 it was 111 mg Cu/m^3. Duration of individual exposures varied; most workers were exposed for 6-9 years. Copper serum concentrations were used to classify exposure and workers were divided into 3 groups based on their serum levels, including one "normal" group (normal value of serum copper: 80-120 µg/100 mL serum). **Inclusion/Exclusion Criteria:** Primarily followed-up with workers directly in contact with copper/copper dust. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** Among all years, headaches were the most reported effect, followed by irritability, vertigo, and perspiration. Neurological observances included memory deficits, disturbed concentration, emotional changes and altered motor reactions. Workers examined in 1973 reported paresthesia, spontaneous limb pain, disturbed sensitivity, and disturbances in reflexes. 15.5% of workers who sieved copper presented with emphysematous thorax,5% had bronchial rales, and 18.5% reported dyspnea. 24.5% of workers had functional changes in capillary hypertony. In 1970, 16% of workers had arterial hypertension and by 1973 only 6% had arterial hypertension and palpitations. Workers showed anorexia, nausea, vomiting, hypochondrial pain, abdominal distension, discomfort and sometimes diarrhea. No hepatic cytolysis was observed. Thymol turbidity test showed that 22% of workers had markers for inflammatory conditions. During dermatological examination, 43 workers showed fissured palmo-plantar hyperkeratosis and 44 workers had green impregnation with copper derivatives of the squamous epithelium and of nails. 16% of workers showed sexual impotence. **Limitations:** No control group was used to compare clinical observations. Lifestyle factors not considered, such as smoking, and local exposure

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
		levels. Selection criteria for workers who participated is not stated, thus there is the potential for selection bias.
Death		
Valdes et al. 2012 Ecological study in Santiago, Chile (population 6 million), 1998-2007. National data on daily mortality counts (cause-specific) were assessed.	**Exposure:** Inhalation of copper in ambient air particulate matter. Air pollution data collected from 1998 to 2008 from one air quality monitoring site located in a residential area of Santiago. Median monthly mean copper in $PM_{2.5}$ was 0.75 ng/m^3 (IQR 0.36 ng/m^3). **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** Day of the week, long-term time trends, temperature, relative humidity, interaction between $PM_{2.5}$ and monthly averages of mean ratios of copper to $PM_{2.5}$ mass.	**Outcomes:** $PM_{2.5}$ copper concentration was positively associated with cause-specific mortalities, including cardiovascular, respiratory, COPD, and cerebrovascular mortalities. PM-adjusted percent increases in cause-specific mortalities per 10 µg/m^3 increase in 2-day average copper in $PM_{2.5}$ were as follows: cardiovascular: 1.24% (95% CI: 0.44, 2.05); respiratory: 2.09% (95% CI: 0.75, 3.45); COPD: 2.64% (95% CI: 0.32, 5.02); cerebrovascular: 1.44% (95% CI: 0.07, 2.82). **Limitations:** $PM_{2.5}$ measurements were not collected consistently throughout the year, and daily $PM_{2.5}$ data were only available April-September. Averaging elemental concentrations over months may have masked some of the variation. $PM_{2.5}$ data were collected from only one location in the study area and may underestimate pollution as study authors noted this area as “one of the main green areas.” The study only considered short-term impacts of copper exposure via $PM_{2.5}$ inhalation on CVD mortality using day of or 2-day average measurements. Total $PM_{2.5}$ was also associated with each cause-specific mortality endpoint that the authors analyzed and may be a confounder.
Respiratory		
Boogaard et al. 2013 **Study Type:** Prospective cohort study of Dutch residents from 12	**Exposure:** Inhalation of copper in ambient air based on elemental composition of particulate matter. Air monitoring data from 12 locations (8 urban streets and 4 suburban background location) in 2008 and 2010.	**Outcomes:** When adjusted for covariates, reductions in traffic-related air pollutants, such as copper, was associated with a statistically

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
locations. Participants had to be at least 4 years of age. Spirometry tests were only done on participants older than 6 years, and along with airway resistance test these were conducted. Participants also completed questionnaire on respiratory symptoms. Results were compared against pollution data in these areas from 2008 and 2010. 661 participants completed both baseline and follow-up examinations.	Six 1-week samples collected over two 6-month periods in both years. 2008 and 2010 mean copper concentration in air for all 12 sites only differed by 0.01 mg/m^3. Mean copper concentration in air in 2008 for all 12 sites = 29.8 m/m^3. **Inclusion/Exclusion Criteria:** Participants had to be ≥4 years old for airway resistance test, and ≥6 years old for spirometry test. **Covariates Considered/Other Regression Adjustments:** Sex, age, number of cigarette(s)/day, level of education, having cold at time of exam, and any difference in time of year that exams (baseline vs. follow-up) were performed (to account for diurnal fluctuation in respiratory health). In other models, smoking status (yes/never/former), passive smoking, bedroom carpet, animals in home, occupational gas exposure, NO_2 concentrations, and temperature were accounted for. Air samples were measured for and analyses were stratified by PM_{10}, $PM_{2.5}$, soot, NO_x, and Cr and, Fe within PM, in addition to Cu in PM.	significant improvement in forced vital capacity (% change in FVC between 2008 and 2010 = -0.88) **Limitations:** Low response rate of about 10% presents possible selection bias, and there is no information on non-respondents. PM_{10}, $PM_{2.5}$, soot, NO_x, and other elements within PM were not accounted for in the model relating copper to respiratory functions, which could be a source of confounding.
Lavigne et al. 2019 **Study Type:** Ecological study of people living in wards in the London and Oxford area of England (population 13.6 million), 2008-2011. Cardiovascular mortality, respiratory mortality, and lung cancer incidence assessed.	**Exposure:** Inhalation of copper in ambient air particulate matter. Copper in $PM_{2.5}$ and in PM_{10} were obtained from air monitoring data from 20 sites from 2010-2011, and land use regression models were developed to predict $PM_{2.5}$ and PM_{10} elemental composition for the study population. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** Age, sex, ward level tobacco expenditure (pounds/week/inhabitant), % of Asian and White populations at the ward level, the 2007 index of	**Outcomes:** There were 48,483 respiratory deaths that occurred in the study area during the study timeframe. Copper in $PM_{2.5}$ was associated with a slightly increased risk of respiratory mortality (RR = 1.003 per IDR, 95% CI: 0.998, 1.009). Copper in PM_{10} fraction had a very small protective association (RR = 0.988, 95% CI: 0.978, 0.998). Cardiovascular and lung cancer outcomes reported below. **Limitations:** Exposure misclassification is possible due to the ecological study design. The study was

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
	multiple deprivation (IMD) was used as a relative measure of area-level deprivation, which serve as a proxy for socio-economic factors.	reliant on registry data and did not have access to individual-level covariate information other than age and sex. The study did not adjust for co-exposure to multiple pollutants and found that PM components were highly correlated.
Cardiovascular		
Badaloni et al. 2017 **Study Type:** Retrospective cohort study of 1,249,108 residents of Rome. Mortality data was assessed.	**Exposure:** Inhalation of copper in ambient air particulate matter. $PM_{2.5}$ and PM_{10} were measured at 20 sampling sites for 14 days in warm, cold, and intermediate seasons in 2010. Sites were chosen to represent the spatial distribution of residential addresses. Land-use regression models used this data to estimate annual average concentrations of pollutants at the baseline residential addresses of study subjects. Mean copper (± SE) in PM_{10} was 57 ± 26.7 $\mu g/m^3$. Mean (± SE) copper in $PM_{2.5}$ was 15 ± 4.2 $\mu g/m^3$. **Inclusion/Exclusion Criteria:** Subjects were participants in the Rome Longitudinal Study (RoLS) 2001 census administrative cohort, which included all Rome residents aged 30+ years who had lived in Rome for at least 5 years and did not reside in a prison, hospital, or nursing home. Subjects were excluded if exposure data were unavailable. **Covariates Considered/Other Regression Adjustments:** Sex, date of birth, marital status, place of birth, education level, occupation, socioeconomic position of census block, co-pollutants (e.g., NO_2).	**Outcomes:** 59,434 participants died from CVD and 22,234 died from IHD. Copper in $PM_{2.5}$ was associated with increased risk of CVD and IHD in unadjusted and total PM mass-adjusted models (total PM-mass adjusted HR for CVD = 1.04, 95% CI: 1.01, 1.07; PM-adjusted HR for IHD = 1.08, 95% CI: 1.04, 1.13 per 5th–95^{th} percentile range increments). Copper in PM_{10} was also associated with increased risk of CVD and IHD in unadjusted and PM-adjusted models (total PM mass-adjusted HR for CVD = 1.05, 95% CI: 1.02, 1.07; PM mass-adjusted HR for IHD = 1.08, 95% CI: 1.03, 1.13 per 5th–95^{th} percentile range increments). **Limitations:** Exposure misclassification is possible because exposure was measured retroactively (regression models were based on measurements in 2010 and applied to residential addresses dating back to 2001). Residential mobility during follow-up was not accounted for. Individual risk factors, such as smoking, physical activity, and diet, were not accounted for.
Lavigne et al. 2019 **Study Type:** Ecological study of people living in wards in the	**Exposure:** Inhalation of copper in ambient air particulate matter. Copper in $PM_{2.5}$ and PM_{10} was obtained from air monitoring data from 20 sites from 2010-2011, and land use regression models were	**Outcomes:** There were 108,478 CVD deaths in the study area during the study timeframe. Copper in $PM_{2.5}$ was associated with increased risk of death

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
London and Oxford area of England (population 13.6 million), 2008-2011. Cardiovascular mortality, respiratory mortality, and lung cancer incidence assessed.	developed to predict $PM_{2.5}$ and PM_{10} elemental composition for study population. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** Age, sex, tobacco weekly expenditure at the ward level, % of Asian and White populations at the ward level, the 2007 index of multiple deprivation as a relative measure of area-level deprivation	by CVD (RR = 1.005 per IDR, 95% CI: 1.001, 1.009). Respiratory and lung cancer outcomes reported in separate entries. **Limitations:** Exposure misclassification is possible due to the ecological study design. The study was reliant on registry data and did not have access to individual-level covariate information other than age and sex. The study did not adjust for co-exposure to multiple pollutants.
Occelli et al. 2020 Retrospective cohort population-level study with cases with data obtained from the French WHO-MONICA population-based coronary heart disease (CHD) registry for the Lille urban area (population 220,000), 2008-2011.	**Exposure:** Inhalation of copper in ambient air particulate matter (PM_{10}). Air pollution data was obtained using air quality monitoring data from 2009. In addition, bioaccumulation of copper in lichen from 2003-2009 was used as an indicator of historic copper levels in ambient air. Study authors developed a composite environmental score (SEnv) for cumulative exposure to air pollution. Each neighborhood was given a SEnv score. Median copper in lichen was 19.2 µg/g (IQR: 15.2, 27.5). **Inclusion/Exclusion Criteria:** CHD cases where secondary prevention measures (medical intervention) were used were excluded. **Covariates Considered/Other Regression Adjustments:** Age, sex, area-level social deprivation, neighborhood spatial structure.	**Outcomes:** SEnv was positively associated with CHD risk (p=0.0151), and copper concentration in lichen was positively associated with SEnv (SP = 0.47, p<0.0001). Relative risk of CHD was 17% higher in neighborhoods in the highest tertile for SEnv compared to those in the lowest (RR = 1.17, 95% CI: 1.05, 1.31). **Limitations:** Exposure misclassification is likely because exposure was measured at the neighborhood level and did not account for possible commuting. Individual CHD risk factors, such as smoking and diet, were not accounted for. Exposure and outcome information were collected retrospectively and for different time periods (outcome was information collected from 2008-2011 and exposure information was measured in 2009).
Ostro et al. 2008 **Study Type:** Ecological study of residents of 6 California counties, 2000-2003. Obtained data from	**Exposure:** Inhalation of copper in ambient air particulate matter. Daily copper in $PM_{2.5}$ was modeled using time-series regression. Mean copper in $PM_{2.5}$ was 0.007 µg/m^3 (IQR 0.007 µg/m^3).	**Outcomes:** Copper in $PM_{2.5}$ was positively associated with CVD mortality among Hispanic subjects in both the 1 and 3 lag day models. Copper in $PM_{2.5}$ was not associated with any change in CVD mortality among white subjects.

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
the state on the daily counts of cardiovascular mortality from areas of interest.	**Inclusion/Exclusion Criteria:** Counties for which 180+ days of $PM_{2.5}$ monitoring data were available from 2000-2003 were included. **Covariates Considered/Other Regression Adjustments:** Time of year, temperature, humidity, day of the week, number of lag days. Analyses were stratified by subject ethnicity and education level.	Copper in $PM_{2.5}$ was positively associated with CVD mortality among non-HS graduates in the 1 lag day model only. Copper in $PM_{2.5}$ was not associated with any change in CVD mortality among HS graduates. **Limitations:** The study has low statistical power due to the small sample size (~350 observations over 4 years). Because of the ecological study design, exposure and outcome were not linked for individual subjects. The study did not adjust for long-term time trends in copper concentrations in $PM_{2.5}$, so results may be sensitive to the sampling period. The study also did not control for total $PM_{2.5}$ in the association between Copper in $PM_{2.5}$ and mortality, which may be a source of confounding. Covariate information was limited to ethnicity and education level.
Ostro et al. 2015 **Study Type:** Prospective cohort population-level study of 101,884 current and former female teachers and administrators in California, 2001-2007. Participants had been previously mailed questionnaires and state mortality data was reviewed.	**Exposure:** Inhalation of copper in ambient air particulate matter. Copper in $PM_{2.5}$ and ultrafine (UF) particles was modeled for each subject on a monthly basis using the emissions inventory in California from 2001-2007. Mean copper in $PM_{2.5}$ was 0.5 $\mu g/m^3$ and mean copper in UF was 0.03 $\mu g/m^3$. **Inclusion/Exclusion Criteria:** Subjects in the California Teachers Study with follow-up information for years 2001-2007. **Covariates Considered/Other Regression Adjustments:** Age, race, marital status, smoking status, pack-years of smoking, body mass index, lifetime physical activity, alcohol consumption, average daily dietary intake of fat, fiber, and calories, menopausal status, hormone replacement therapy,	**Outcomes:** 1,085/101,884 subjects died from ischemic heart disease (IHD). Copper in $PM_{2.5}$ and UF was positively associated with IHD mortality. The hazard ratio for increase in risk of IHD mortality per 10 $\mu g/m^3$ increase in copper in $PM_{2.5}$ was 1.09 (95% CI: 1.04, 1.15). The hazard ratio for increase in risk of IHD mortality per 10 $\mu g/m^3$ increase in copper in UF was 1.06 (95% CI: 1.03, 1.09). Associations of cardiovascular mortality for copper were reported (HR = 1.03; 95% CI: 1.00, 1.05). **Limitations:** The study has limited generalizability because enrollment was limited to women. Enrolled women all shared the same occupation (schoolteachers or administrators), so the study also cannot be generalized to all women. Incidence of IHD was low in this cohort, so risk estimates may

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
	family history of myocardial infarction and stroke, use of blood pressure medication.	be unstable. Exposure models did not account for possible synergism between co-pollutants or total $PM_{2.5}$.
Wang et al. 2017 **Study Type:** Time-series population-level analysis in Pudong New Area of Shanghai, China (population 2.96 million in 2016), 2014-2016. Daily counts of CVD death from 2014-2016 in the district collected.	**Exposure:** Inhalation of copper in ambient air particulate matter. Daily $PM_{2.5}$ measurements were obtained from the Shanghai Environmental Monitoring Center from 2014-2016. Mean copper in $PM_{2.5}$ was 0.02 ± 0.02 SD μg/m^3. **Inclusion/Exclusion Criteria:** Subjects were permanent residents of the Pudong New Area of Shanghai from 2014-2016. **Covariates Considered/Other Regression Adjustments:** Calendar day, seasonality of CVD mortality, long-term CVD mortality trends, and concentrations of O_3, SO_2, NO_2, and CO in $PM_{2.5}$. $PM_{2.5}$ total mass was controlled for.	**Outcomes:** Previous 2-day $PM_{2.5}$ copper concentration was positively associated with cardiovascular mortality. The percent increase in cardiovascular mortality associated with an IQR increase in $PM_{2.5}$ and copper were 1.53% (95% CI 0.37%, 2.69%) before adjusting for other gaseous pollutants. The association remained positive and significant after adjusting for O_3, SO_2, NO_2, and CO concentrations **Limitations:** Exposure measurement error is likely because exposure data was collected from a fixed-site monitor. The study only considered short term effects of copper in $PM_{2.5}$ on daily CVD mortality.
Neurological		
Pujol et al. 2016 **Study Type:** Cross-sectional study of 2,836 children aged 8-12 years from schools in Barcelona, Spain who completed behavioral testing to test motor function. A subgroup of 236 children had a 3D MRI, functional MRI, and diffusion tensor imaging (DTI) to test brain repercussions.	**Exposure:** Air samples were collected from all schools that participants attended to calculate years air pollution levels. Samples were collected twice during 1-week periods separated by 6 months in warm and cold weather months through 2012 and 2013. Indoor air in the classrooms and outdoor air in the school courtyards was measured. The primary source of copper was road traffic, followed by industrial activity. **Inclusion/Exclusion Criteria:** Children with dental braces. DTI images were excluded when image degradation was detectable.	**Outcomes:** Higher copper exposure was associated with poorer motor performance, which was significant for reaction time (p=0.006; β=2.2). This relationship was observed among the main study group and subgroup. Among the subgroup, copper exposure was associated with a higher proportion of gray matter in the brain tissue (striatum). No other significant alterations were seen on the MRI. In the DTI, copper was associated with increased neural tissue fractional anisotropy.

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
		Limitations: Potential head movement in children during imaging may affect the quality. Self-selection bias as parents opted into the study.
Developmental		
Pedersen et al. 2016 **Study Type:** Retrospective birth cohort study of 34,923 mother-infant pairs from 8 European cohorts, 2008-2011. Birth outcomes obtained from either birth records or from parental reports.	**Exposure:** Inhalation of copper in ambient air particulate matter. PM composition was measured at various European sites from 2008-2013. Mean copper in $PM_{2.5}$ was 3.4 μg/m^3 ± 2.1 SD. Mean copper in PM_{10} was 14.0 μg/m^3 ± 10.6 SD. **Inclusion/Exclusion Criteria:** Data were pooled from 8 European birth cohorts and limited to singleton births from 1994-2008. **Covariates Considered/Other Regression Adjustments:** Maternal alcohol consumption, smoking active and passive smoking during pregnancy, age, pre-pregnancy weight, education level, annual household income, parity, height, infant season of conception, gestational age, sex.	**Outcomes:** Copper in $PM_{2.5}$ and PM_{10} were not associated with mean birth weight in term births (β = 10, 95% CI: -8, 27 and β = 8, 95% CI: -4, 19, respectively). Copper in $PM_{2.5}$ and PM_{10} were not associated with odds of low birth weight (LBW) among term births (OR = 1.08, 95% CI: -0.81, 1.44) and OR = 1.13, 95% CI: -0.92, 1.39, respectively). **Limitations:** Exposure data was collected retrospectively (air quality measurements from 2008-2013 were applied to birth outcomes that occurred from 1994-2008), so temporality cannot be inferred. Subjects' exposure status was determined by home address, and the study did not account for commuting.
Cancer		
Lavigne et al. 2019 **Study Type:** Ecological study of people living in wards in the London and Oxford area of England (population 13.6 million), 2008-2011. Cardiovascular mortality, respiratory mortality, and lung cancer incidence assessed.	**Exposure:** Inhalation of copper in ambient air particulate matter. Copper in $PM_{2.5}$ and PM_{10} was obtained from air monitoring data from 20 sites from 2010-2011, and land use regression models were developed to predict $PM_{2.5}$ and PM_{10} elemental composition for study population. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** Age, sex, tobacco weekly expenditure at the ward level, % of Asian and White populations at	**Outcomes:** There were 24,849 incident cases of lung cancer in the study area during the study timeframe. Copper in $PM_{2.5}$ was associated with increased lung cancer incidence (RR = 1.092 per IDR, 95% CI: 0.943, 1.225). Copper in PM_{10} was associated with increased lung cancer incidence (RR = 0.998 per IDR, 95% CI: 0.912, 1.091). Cardiovascular and respiratory outcomes are reported in separate entries. **Limitations:** Exposure misclassification is possible due to the ecological study design. The study was

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations
	the ward level, the 2007 index of multiple deprivation as a relative measure of area-level deprivation	reliant on registry data and did not have access to individual-level covariate information other than age and sex. The study did not adjust for co-exposure to multiple pollutants and found that PM components were highly correlated.
Raaschou-Nielsen et al. 2016 Prospective cohort study of 245,782 people using pooled data from 14 cohorts across 8 European countries, 1997-2008. Incidence of lung cancer analyzed for each cohort the local study centers.	**Exposure:** Inhalation of copper in ambient air particulate matter, both $PM_{2.5}$ and PM_{10}. Air pollution measurements were taken at 250 locations for each cohort from October 2008-May 2011 and land use regression was used to estimate PM composition at subjects' baseline addresses. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** Sex, calendar time, smoking status, smoking intensity, smoking duration, time since quitting smoking, environmental tobacco smoke, occupation, fruit intake, marital status, education level employment status, and area-level employment status. Models were fit with one-pollutant at a time then two-pollutant model fits for each element and concentration of particulate matter, NO_2, and NO_x.	**Outcomes:** There were 1,878 incident cases of lung cancer among study subjects. Copper in $PM_{2.5}$, but not PM_{10}, was associated within increased risk of lung cancer (HR for copper in $PM_{2.5}$: 1.25, 95% CI: 1.01, 1.53, and HR for copper in PM_{10}: 1.14, 95% CI: 0.96, 1.35); however, the effect was somewhat attenuated when controlling for Fe in $PM_{2.5}$. In two-metal models with Fe and Cu, an increase in risk of lung cancer was seen for the main effect of copper in PM2.5 and PM10. **Limitations:** Exposure was assessed retrospectively (air quality data from 2008-2011 was applied to outcome data from 1997-2008). Covariate information was collected only at baseline. Mean subject age ranged from 43-73, so generalizability to younger age groups may not be limited. Use of regression modeling to estimate exposure likely resulted in some exposure misclassification. Because misclassification may be present in varying degrees with each contaminant, when elements are correlated with each other, the misclassification of one element in relation to lung cancer may affect the misclassification present in the other estimate of element and lung cancer risk; this effect may be present in the Cu-Fe relationship explored above.

ARF = acute renal failure, BMI = body mass index, CI = confidence interval, CHD = coronary heart disease, CVD = cardiovascular disease, EAF = electric arc furnace, FEF = mid-expiratory flows, FVC = forced vital capacity; HR = hazard ratio; IDR = interdecile range, IHD = ischemic heart

Table 2-4. Epidemiological Studies of Humans Exposed to Copper in Air

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, and Covariates Considered/Other Regression Adjustments	Outcomes and Limitations

disease, IMD = index of multiple deprivation, MND = motor neuron disease, PM = particulate matter, RR = relative risk, SMR = standardized mortality ratio, UF = ultrafine

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Gastrointestinal			
Araya et al. 2001 Randomized, prospective, double-blind, controlled study of 179 healthy adults, exposed to copper sulfate added to their drinking water or placebo, after overnight fasting.	**Exposure:** Weekly dose for 5 weeks. Copper sulfate bolus added to drinking water and screened for symptoms for 24 hours after dose. **Adjustments:** none	Copper as copper sulfate added to drinking water: 0, 2, 4, 6, 8 mg Cu/L, and corresponding total copper dose calculated by study were 0.4, 0.8, 1.2, and 1.6 mg Cu, respectively. Total daily doses were 0.006, 0.012, 0.018, and 0.025 mg Cu/kg/day.	Clear dose-response relationship observed with gastrointestinal symptoms and nausea; and nausea and vomiting appeared related. Gastrointestinal (GI) symptoms appeared at 6 mg Cu/L. **Limitations:** Recall bias (self-reporting of symptoms)
Araya et al. 2003a Randomized, double-blind, controlled study of 30 healthy adults, 15 women (mean age 33 years) and 15 men (mean age 37 years), exposed to copper added to their drinking water or placebo, after overnight fasting.	**Exposure:** Each participant underwent two trials receiving, in random order, either placebo or the water-copper sulfate solution and was observed immediately after for 2 hours, then followed up with 24 hours after to check for symptoms. **Adjustments:** none	Copper as copper sulfate added to drinking water: 0 mg Cu/L (placebo), 10 mg Cu/L (0.046 mg Cu/kg).	9 subjects had nausea after receiving copper solution. Presence of copper in the stomach caused delayed gastric emptying presenting as a copper-induced significant delay in decreasing antral area. **Limitations:** Recall bias (self-reporting of symptoms)

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Araya et al. 2003b Randomized, double-blind community-based study of 1,365 adults in Santiago, Chile where participants were exposed by adding copper to their daily drinking water for 2 months. All participants shared the same water source, and all had copper pipes. Of the participants, 240 participants (60 from each dose group) provided a blood sample. Mean age: 32.9 years ± 12.5. Range of ages: 20-55 years.	**Exposure:** Participants were randomized into 4 groups and added the provided copper solution (as copper sulfate) into 10 liters of drinking water daily to be consumed throughout the day. Subjects reported symptoms daily in a diary and were survey for daily consumption. Acute gastrointestinal symptoms reported in Araya et al. 2004. This study reported results from blood sampling. **Adjustments:** None.	Copper added to drinking water (mg/L): <0.01, 2, 4, or 6. Equating to doses of: 0, 0.042, 0.091, or 0.17 mg Cu/kg/day.	The percentage of participants reporting gastrointestinal symptoms at 6 mg Cu/L (19.7%) was significantly greater compared to controls. Among 240 participants, no exposure-related changes in indicators of copper status and homeostatic regulations, and liver function not affected. **Limitations:** Water consumption and symptoms were self-reported (recall bias)
Araya et al. 2003c Randomized, double-blind, 3x3 factorial (volume x dose) study of 269 adult females (aged 18-60 years) from four different international sites (70 in Santiago, Chile; 68 in North Dakota, U.S.; 58 in Coleraine, Northern Ireland; 73 in Shanghai, China).	**Exposure:** 3 x 3 two-way factorial design had participants given doses of in volumes of either 100,150, or 200 ml bottled drinking water. A control and high dose group also added. Participants fasted the night before visiting the testing center, 3 hours within waking, once a week for 11 successive weeks. Immediately after participants completed a symptoms questionnaire. **Adjustments:** none	Range of copper concentrations, as copper sulfate, among controls, 3x3 factorial and high dose (mg Cu/L): 0, 2, 2.6, 4, 5.3, 5, 8, 12, which equate to doses of 0, 0.03, 0.04, 0.06, 0.08, 0.09, 0.12, 0.18 mg Cu/kg.	Incidence of nausea increased with dose at all study sites and typically within 15 minutes after water consumption. Nausea incidence decreased with time after ingestion. Probability of all gastrointestinal symptoms increased with copper dose, with highest probability of 0.25 (0.2, 0.3) in 1.6 mg Cu group within 15 minutes of ingestion.

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Araya et al. 2004 Randomized, double-blind community-based study of 1,365 adults in Santiago, Chile where participants were exposed by adding copper to their daily drinking water for 2 months. All participants shared the same water source, and all had copper pipes. Mean participant age: 36.9-38.5 years Sample size: 327-355 adults per exposure group	**Exposure:** Participants were randomized into 4 groups and added provided copper solution into 1 liter of drinking water daily that had to be consumed throughout the day. **Adjustments:** Censored subjects were included and counted per event when their information was incomplete. Data were stratified by time. Covariates included sex, age, total daily fluid volume, volume of water ingested as plain water, and volume of water ingested as mixed fluids.	Copper added to drinking water (mg/L): <0.01, 2, 4, or 6, which equate to doses of 0, 0.055, 0.106, or 0.169 mg Cu/kg/day. End of study concentration in water: 0.05, 2.02, 3.71, or 5.77 mg/L	The risk of gastrointestinal effects increased as copper exposure increased. Women had a higher risk of gastrointestinal symptoms at a lower copper dose than men. At 4 mg/L, women showed an increased risk of gastrointestinal symptoms (RR: 1.53, CI: 1.02, 2.05), while men showed an increased risk of symptoms at 6 mg/L (1.9; CI: 1.02, 2.79). **Limitations:** Water consumption and symptoms were recorded daily by one person in each household, meaning that the data were self-reported, and the timing of exposure compared to symptom onset is unknown.
Buchanan et al. 1999 Retrospective cohort study and nested case-control study in Nebraska. Levels in 1993 had exceeded action level at the time of 1.3 mg/L. Study authors interviewed households who had been exposed to different levels of copper in drinking water. 148 participating households with 451 individuals in cohort study	**Exposure:** Cohort of 182 households were selected from communities where the Nebraska Department of Health had tested the drinking water for copper in 1993. Household members were interviewed about water consumption habits and occurrence of gastrointestinal effects. Water of all participants then measured in August 1994. The cases were identified from these interviews, and then chosen for the nested case-control study,	Households with drinking water copper concentrations >3 mg/L (n = 60), copper concentrations between 2 and 3 mg/L (n = 60), and copper concentrations >1.3 mg/L (n = 62) 1993 Drinking water concentrations (mg/L): ≤1.3, 2.0-2.9, ≥3.0 1994 Drinking water concentrations (mg/L	No significant increase in risk of gastrointestinal symptoms was associated with drinking water copper concentrations greater than 3 mg/L (RR = 1.03; 95% CI: 0.43, 2.49) or 2-3 mg/L (RR = 0.50; 95% CI: 0.18, 1.41) compared to concentrations <1.3 mg/L. **Limitations:** Recall bias (self-reported symptoms)

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
34 individuals in nested-cohort study	with more health interviews and water was tested. **Adjustments:** Risk factors excluded from analysis for affecting <3 participants included medical history of alcoholism, anemia, cancer, high blood pressure, kidney and/or liver problems, and pregnancy; household water system information; previous water contamination with lead or iron; controlled for clustering of people in homes	≤1.3, 1.3-2.9, ≥3.0	
Eife et al. 1999 Retrospective cohort of 29 patients that suffered from nausea, vomiting, colic, and diarrhea (27 from Germany, 1 from Austria, and 1 from USA), 9 adults and 20 children (14/20 children were infants). All patients lived in homes with copper plumbing and reported clinical signs indicating chronic copper intoxication.	**Exposure:** Copper pipes were leaching copper into the water supply. Serum copper ranges in participants by age: 2–7 months: 18.6–35.4 µmol/L 3 years: 61.1 µmol/L 8–16 years: 15.3–24.2 µmol/L “adults”: 13.1–16.4 µmol/L **Adjustments:** none	Concentration in drinking water: 0.1–16.9 mg Cu/L tap water	The authors concluded that the gastrointestinal effects observed in these patients were associated with their copper exposure, along with other systemic diseases of chronic copper intoxication including hepatopathy and natural-killer-cell deficiency. **Limitations:** Study recruitment was inconsistent and involved parent reports, neighbor reports, doctor referrals, and self-reports introducing recall bias. Serum and urine copper levels were only measured in 12/29 patients. Other potential causes of gastrointestinal symptoms do not appear to have been evaluated.
Gotteland et al. 2001	**Exposure:** Controlled exposure of copper added to 200 ml of drinking water, and	Copper added to drinking water (as copper sulfate): 0 mg Cu/L (controls)	There was a copper related increase in gastric permeability to sucrose, but this was not related to gastrointestinal symptoms.

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Randomized, double-blind study of 31 healthy adults, 15 males, and 16 females (age range: 20-63 years) exposed to copper in drinking water and observed after in a controlled setting for 5 hours and 15 minutes. Participants recorded severity of gastrointestinal symptoms.	then consumption of 200 ml of water with no copper. Equates to a dose of 0.03 mg Cu/kg. **Adjustments:** none	10 mg Cu/L (total copper dose of 2 mg)	After copper ingestion, 22.6% of participants reported gastrointestinal symptoms that were reported as significantly more intense than control participants. **Limitations:** Self-reported perception of symptom intensity
Knobeloch et al. 1994 Study II Between January and June of 1992, a Wisconsin community in newly built homes reported flu-like symptoms attributed to copper-contaminated drinking water tested in the home and distribution system (27 adults and 15 children).	Exposure: Elevated copper in drinking water Adjustments: none	Concentration in drinking water at first draw: 0.16 – 0.65 mg/L	In residents of new homes, Cu concentrations in first draw (rather than flushed) of tap water was associated with an increased relative risk of gastrointestinal upset (RR: 5.25; CI: 1.85, 14.91). Symptoms include diarrhea and abdominal pain. **Limitations:** Recall bias (self-reported symptoms)
Knobeloch et al. 1994 Study III A Wisconsin community was found to have elevated copper levels in their drinking water. 60 residents (ages 15 months to 91 years; mean: 52 years) living in 37 homes responded to health surveys reporting their residence history, age, water	**Exposure:** Elevated copper in drinking water **Adjustments:** none	Concentration in drinking water: 0.09 – 5.3 mg/L	Time of residence at the home (<1 year or >1 year) was associated with an increased relative risk of gastrointestinal symptoms (RR: 8.30, CI: 2.21, 31.09). **Limitations:** Recall bias (self-reported symptoms)

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
consumption habits, and any symptoms.			
Knobeloch et al. 1994 Study IV In 1992, water from water fountain and bathroom sinks in a Wisconsin university building had high levels of copper as water appeared blue in color. Health questionnaires, water consumption, and time in the building was recorded from employees. 41 respondents (17 women, 14 men), age range 27-67 years, mean age: 47.5 years).	**Exposure:** Building water with elevated copper levels **Adjustments:** none	Daily intake from drinking water: 0 – 3.8 mg Cu/day	Daily copper intake of 0.6-3.8 mg Cu/day was associated with increased relative risk of gastrointestinal symptoms (RR: 4.95, CI: 1.56, 15.75), compared to a daily copper intake of 0-0.55 mg Cu/day. Drinking water primarily from the fountain (higher copper concentrations) was associated with increased relative risk of gastrointestinal symptoms (RR: 4.64, CI: 0.66, 32.55) **Limitations:** Recall bias (self-reported symptoms), non-response bias, and small sample size
Knobeloch et al. 1994 Study V Residents of an apartment building in Wisconsin reported bitter-tasting water and asked the local health department to inspect the water supply.19 residents (13 adults and 6 children) filled out health surveys including information on residential history, water use, and health status.	**Exposure:** household tap water with elevated copper levels **Adjustments:** none	Daily intake from drinking water: 0.5 – 8.1 mg Cu/day (mean: 3.4 mg Cu/day)	Relative risk of gastrointestinal upset was higher in children (<18 years) compared to adults (>18 years) (RR: 1.14, CI: 0.73,1.78), but higher copper dose was not associated with higher risk of gastrointestinal upset (RR: 0.74, CI: 0.45, 1.23). **Limitations:** Recall bias (self-reported symptoms) and small sample size

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Knobeloch et al. 1998 Study 1 Elevated copper levels were found in new or recently remodeled homes of 188 people from 89 families in Wisconsin. Residents were given health surveys to report symptoms and drinking water source.	**Exposure:** household tap water with elevated copper levels **Adjustments:** none	Concentration in drinking water: 0.013 – 4.3 mg/L	A daily intake of >3 mg Cu/day was associated with an increased risk of diarrhea (RR: 1.83, CI: 1.13, 58.36), indigestion (RR: 2.17, CI: 1.09, 4.03), and cramps (RR: 4.98, CI: 2.17, 11.46) when compared to those with <3 mg Cu/day. Symptoms reported include indigestion, diarrhea, abdominal cramps and/or nausea. **Limitations:** symptoms were self-reported via questionnaire
Knobeloch et al. 1998 Study 2 In 1996, residents of a mobile home park in Wisconsin reported “blue water” with a metallic taste and gastrointestinal symptoms. New copper pipes had been added to the water distribution system. 13 families completed health surveys (22 adults and 16 children aged 1-17 years).	**Exposure:** water samples were collected from 10 homes **Adjustments:** age of the home, use of water treatment devices	Concentration in drinking water: 0.4 – 3.7 mg/L	No statistical analysis was done to measure the relationship between copper ingestion and health outcomes. Park residents reported diarrhea (29% of respondents) and upset stomachs (50%). Copper levels were not correlated with age of home, and symptoms rates were higher among children than adults. **Limitations:** Recall bias (symptoms were self-reported via questionnaire) and small sample size
Olivares et al. 2001 61 adult participants from Santiago, Chile (age range: 25 to 60 years) were exposed to added copper in either water or an orange-flavored juice.	**Exposure:** Participants were randomized into 6 groups each week, and then given water or juice with added copper sulfate. Thus, concentrations were tested in almost all participants, resulting in 47-61 adults in each exposure group.	Copper (as copper sulfate) concentration in water or juice: 0, 2, 4, 6, 8, 10, 12 mg Cu/L. Doses in water equate to 0, 0.006, 0.012, 0.018, 0.025, 0.031, 0.037 mg Cu/kg.	Significant increase in incidence of nausea at ≥4 mg/L in water. Significant increase in incidence of vomiting at ≥6 mg/L in water. In juice, threshold for nausea at 8 mg/L. No age or gender related differences observed. **Limitations:** Recall bias (self-reporting of symptoms)

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Participants came in once a week for 12 exposures.	**Adjustments:** none		
Pettersson et al. 2003 Swedish cohort study from two municipalities with repeatedly high copper concentrations in drinking water. 430 families responded to questionnaires, and 430 young children in the study, aged 9 months to 21 months.	**Exposure:** 4703 samples of tap water from the children's homes were used to measure their exposure to copper. Surveys, a short questionnaire on water consumption habits, and parent-reported symptoms were used to determine gastrointestinal health outcomes. **Logistic regression adjustments**: Ages, number of siblings, family history of allergy, number of daily meals obtained by breastfeeding	Concentration in drinking water: 0.01 to 5 mg/L	No statistically significant increase in diarrhea or vomiting was found for higher copper intake. **Limitations**: The data on the gastrointestinal health outcome was self-reported via survey (recall bias).
Pizarro et al. 1999 Prospective, double-blind, Latin square design study in 60 adult women who were randomly assigned 4 concentrations of copper in their drinking water. Participants were split into 4 groups of 15 people; every participant was exposed to each copper concentration and served as their own control. After each 2-week exposure	**Exposure:** Each group was assigned a different sequence of exposure concentrations. Groups consumed water containing 0, 1, 3, or 5 mg/L ionic copper as copper sulfate pentahydrate for a 2-week period followed by a 1-week rest, then consumed the next concentration in sequence. Each dose was tested in each participant.	Concentrations of 0, 1, 3, or 5 mg/L of copper sulfate pentahydrate equated to daily copper intakes of 0.04 1.74, 4.68, and 7.94 mg Cu/day. Doses calculated using the study reported body weight of 64 kg. Doses were 0.0006, 0.03, 0.07, and 0.1 mg Cu/kg/day.	Twelve subjects reported symptoms of abdominal pain, nausea, and/or vomiting; the incidences were 3/60, 1/60, 10/60, and 9/60 in the 0, 0.03, 0.07, and 0.1 mg Cu/kg/day groups, respectively. Nine subjects reported 12 episodes of diarrhea with or without abdominal pain; no association was found between copper concentration in water and diarrhea. There were no significant differences in serum copper, ceruloplasmin, hemoglobin, or liver enzymes levels between the groups.

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
period, participants reported symptoms and blood was taken at the beginning and towards the end of study.	**Adjustments:** none		**Limitations:** There was no certainty that participants prepared and consumed the water as instructed. Participant consumption of the experimental groups decreased from weeks 1-2 to weeks 11-12, which likely lowered copper exposure and subsequent risk of developing gastrointestinal symptoms. Subjects may have also adapted to high copper concentrations over study period.
Pizarro et al. 2001 Prospective, double-blind study in 45 adult women who worked at home, were neither pregnant or lactating, and lived in Santiago, Chile. Subjects randomized into 3 groups of 15 people, and exposure based on Latin square design. Subjects reported any gastrointestinal symptoms daily on a questionnaire.	**Exposure:** Each group was assigned a different sequence of proportions of copper sulfate to copper oxide (0:5, 1:4, 2:3, 3:2, 5:0 mg/L). Total duration was 9 weeks, five 1-week exposure period with 1-week breaks in between. The total concentration at a given time was 5 mg Cu/L. **Adjustments:** none	Dose of 0.1 mg Cu/kg/day was calculated using 1.2 L of water consumption and body weight of 62.7 kg.	Nine subjects reported diarrhea (with or without abdominal pain and vomiting); seven of the 10 episodes of diarrhea occurred during the first half of the study period and the incidence was not related to ratio of copper sulfate to copper oxide. No time effect was observed for nausea, abdominal pain, and vomiting. Eleven subjects reported abdominal pain, nausea, or vomiting; this incidence is significantly higher than the incidence during the periods the subjects ingested plain tap water. The incidences of gastrointestinal symptoms (excluding diarrhea) for each copper sulfate to copper oxide ratio (0:5, 1:4, 2:3, 3:2, and 5:0) were 5, 3, 3, 2, 6, respectively. No differences in activities of liver enzymes between groups. **Limitations:** Subjects were not asked to record when they consumed the water. Subjects may have consumed the water with meals with may have reduced gastrointestinal symptoms.
Pizarro et al. 2007	**Exposure:** Categorized into 3 groups: group 1 (district with	Dose not reported. Sampled households	The odds of GI symptoms were significantly increased in participants who were younger than

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Retrospective cohort study of 1,778 families (6,782 people) and their drinking water supplies in Talca, Chile following government reports of gastrointestinal symptoms in the population.	copper-based plumbing where residents reported health effects, n=2,613), group 2 (districts with copper-based plumbing where residents reported no health effects, n=2,515), and group 3 (district with polyvinyl chloride plumbing where residents reported no health effects, n=1,654). **Logistic Regression Adjustments:** dichotomous variables for sex, age (<12 vs ≥12 years), pipe replacement or recent construction, water intake (<0.6 vs ≥0.6 L/d), bottled water consumption (<0.2 vs =0.2 L/d), year home was built (<1996 vs ≥1996), time spent at home (<16 vs ≥16 h/d), and whether participant was the first member of the household to get up in the morning.	showed a mean stagnant water concentration of 0.5 ±0.32 mg Cu/L. Measurement 6 months later showed a mean concentration of 0.57 ±0.36 mg Cu/L.	12 years (OR: 1.468, 95% CI: 1.178,1.832), female (OR: 1.226, 95% CI: 1.010, 1.490), lived in a home built during or after 1966 (OR: 1.279, 95% CI=1.005, 1.626), or who drank less than 200 mL of bottled water per day (OR: 1.668, 95% CI: 1.273, 2.185). Additionally, the odds of GI symptoms were significantly increased in participants living in a home with copper plumbing reporting health complaints compared to participants with copper plumbing reporting no health complaints (OR: 1.589, 95% CI: 1.187, 2.128) or compared to participants with plastic plumbing reporting no health complaints (OR: 1.73, 95% CI: 1.301, 2.301). **Limitations:** The health survey and water sampling were conducted months after the original reports of symptoms. Only a subsample of the homes was selected for Cu tap water concentration measurements. GI symptoms were self-reported, and parents reported symptoms for children younger than 12. Residents of homes without copper pipes drank significantly more tap water. Additionally, residents with copper pipes reported significantly more symptoms unrelated to copper ingestion (ex. allergies, bronchitis, emotional stress) compared to residents without copper pipes.
Hepatic			
Dieter et al. 1999 Retrospective case-control of 103 pediatric patients in	**Exposure:** Patient postal codes were used to collect information from local water suppliers on drinking water	Estimated concentration in drinking water: Low: <1 mg Cu/L Medium: 1-2 mg Cu/L	No statistical analysis was performed on the data. 8/103 cases of ECC (7.8% of patients) occurred in patients with high copper exposure. 5 of those 8 cases occurred in children with high liver

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
Germany with confirmed cases of early childhood cirrhosis (ECC) for the years 1984-1994.	quality. For patients whose homes used a private well, researchers relied on self-reported information from the parents regarding the presence of copper plumbing and the use of tap water. Patients were divided into groups of low, medium, high, or very high copper exposure. Additional information about the patients was identified using the patient records. **Adjustments:** none	High:2-3 mg Cu/L Very High: >3 mg Cu/L	copper and copper plumbing/acid well water. Connection with copper exposure considered probable. **Limitations**: Given the retrospective nature of this study, the temporality of exposure may be inaccurate. Additionally, only 53 parents (51.5%) responded to the request for information about their family's water supply and use. No statistical risk calculation was conducted.
Zietz et al. 2003a A cohort study of infants 8-10 months old living in households with copper plumbing. Water samples were collected from 2,944 houses in Berlin, Germany. Sample were taken and submitted by participants.	**Exposure:** Mean copper concentrations measured in tap water from the home. Families with 0.8 mg Cu/L or more in their water and whose infants ingested tap water were recommended for pediatric exams. Blood copper levels of 79 to 250 ug/dL for infants exposed to elevated copper levels. **Adjustments:** none	Mean copper concentration was 0.44 mg Cu/L and 0.56 mg Cu/L for each of the two composite samples	No signs of liver disease were found in the exposed infants during their pediatric exams. No dose relation between liver serum parameters and copper intake. **Limitations:** Some participants were lost to follow up. Composite copper concentrations limited the ability to draw conclusions about copper dosage.
Zietz et al. 2003b A cohort study of infants up to 12 months old from households	**Exposure:** Copper concentrations measured in household tap water	Concentration in stagnated drinking water: >0.01 – 6.40 mg/L	No signs of liver malfunction were found in the infants.

Table 2-5. Summary of Health Outcomes in Humans Exposed to Copper in Drinking Water

Reference and study population	Exposure	Cu Dose or Exposure	Outcomes
with copper plumbing, from mothers who had given birth in or near Gottingen, Germany. 1,674 households participated, and 172 households were studied more closely, and 14 infants were examined by a pediatrician. Family medical history was obtained.	Blood copper levels of 82 to 220 µg/dL, among 11 infants. **Adjustments:** none	Concentration in daytime samples: >0.01 – 3.00 mg/L	**Limitations:** Pediatrician information and blood samples were only available for a very small subset of infants.
Developmental			
Longerich et al. 1991 Case-control study of 56 women in Newfoundland, Canada who had recently given birth to infants. 28 women (cases) gave birth to infants with neural tube defects (NTD) and 28 women (matched controls) gave birth to infants without NTDs.	**Exposure:** Trace elements, including copper, were measured in the women's drinking water **Adjustments:** A Student's t-test was run to compare the mean copper level in the drinking water of participants, but no adjustments were made during the analysis.	Mean water concentrations: 210 ± 285 ppb (controls), 290 ± 367 ppb (cases)	While the mean copper concentration was higher for cases, no significant difference in average drinking water copper concentration was observed between mothers that gave birth to infants with NTD and mothers that gave birth to infants without NTD. **Limitations**: Drinking water samples were collected once and assumed to be indicative of exposure during pregnancy. Mothers were matched by age of their infants, but no other consideration of potential confounding appears to have been made.

CI = confidence interval, ECC = early childhood cirrhosis, GI = gastrointestinal, NTD = neural tube defects, RR = relative risk

Table 2-6. Copper Exposure from Environmental Media and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
Musculoskeletal		
Yang et al. 2016 **Study Population:** Cross-sectional study of 122 rheumatoid arthritis (RA) patients in a medical center in central Taiwan. Patient age ranged from 16-89 years; 34 males and 88 females.	**Exposure:** Possible ingestion of copper in farm soils. Copper levels in farm soils were obtained from a national database and averaged at the town/precinct level. Each town/precinct received a grade for copper concentration as follows: Grade 1 (most severe): >23.83 mg/kg; Grade 2: 15.43-23.83 mg/kg; Grade 3: 7.03-15.43 mg/kg; Grade 4 (least severe): 0-7.03 mg/kg. Blood copper was also measured in 39 subjects. **Inclusion/Exclusion Criteria:** Subjects had to be formally diagnosed with RA. In the comparison group, gout patients had to have had at least one attack and ankylosing spondylitis (AS) patients were formally diagnosed. **Covariates Considered/Other Regression Adjustments:** In subjects for whom blood copper was measured, multiple regression models were adjusted for age, sex, smoking, hemoglobin level, and an array of other heavy metals.	**Outcomes:** RA patients living in Grade 1 towns had significantly higher mean WBC count than those living in Grade 4 towns and significantly higher mean platelet count and erythrocyte sedimentation rate (ESR) than those living in Grade 2, 3, and 4 towns. RA patients living in Grade 1 towns also had significantly higher mean overall RA disease activity score than those living in Grade 3 and 4 towns. RA patients for whom blood copper was measured had higher blood copper levels than gout patients, AS patients, and steel plant workers of similar backgrounds. Among RA patients, blood copper was significantly positively correlated with white blood count (WBC) count, ESR, platelet count, and rheumatoid factor-IgM. **Limitations:** The cross-sectional study design prevents conclusions about a causal relationship between copper concentration in soil or blood copper and RA outcomes. The study did not account for other known determinants of RA outcomes, including genes, viral infections, and vitamin D deficiency. The small sample size (especially for blood copper measurement) limited the statistical power, possibly attenuating the observed association between copper levels in farm soil and blood copper levels in subjects.
Neurological		
Sánchez-Díaz et al. 2018 **Study Type:** Ecological study using mortality data from the National Statistics Institute of Spain (n=9434), 2007-2016.	**Exposure:** Copper released into river basins at 235 sites in Spain from 2007-2015. Exposure was measured at the municipality level. Exposed municipalities were considered those to be within a 20-km river section downstream of at least one	**Outcomes:** The standardized mortality ratio (SMR) for motor neuron disease (MND) was 12.7% higher in copper-exposed municipalities than in unexposed municipalities (IRR = 1.127, 95% CI: 1.075, 1.182).

Table 2-6. Copper Exposure from Environmental Media and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
	site where a copper release was made into the river during the considered time period. **Inclusion/Exclusion Criteria:** none **Covariates Considered/Other Regression Adjustments:** None.	**Limitations:** Exposure misclassification is likely because of the ecological study design (subjects' exposure status was determined by their municipality of residence). The study did not consider exposure to copper from other sources or demographic information for subjects or municipalities. Possible synergistic or antagonistic effects of mixing metals in water, such as cadmium, on MND mortality was not considered, only the effect of isolated metals.
Shen et al. 2014 **Study Type:** Ecological study in 26 provinces and 3 municipal districts of mainland China, 1991-2000. Mortality data of locations analyzed.	**Exposure:** Copper concentration in soil. Mean copper concentration in the "A" soil horizon 24.04 mg/kg ± 6.18 SD. Mean copper concentration in the "C" soil horizon) was 24.96 mg/kg ± 6.65 SD. Mean copper concentration in the A and C soil horizons combined was 49.00 mg/kg ± 12.44 SD. **Inclusion/Exclusion Criteria:** Subjects had to be age 40 or older. **Covariates Considered/Other Regression Adjustments:** Age, gender.	**Outcomes:** Copper in soil was positively associated with relative risk of Alzheimer's Disease (AD) mortality. Compared to regions where copper in soil was <20 mg/kg, regions with the highest copper soil levels (60-80 mg/kg) had an RR of 2.634 (95% CI: 2.626, 2.642). **Limitations:** Due to the ecological study design, exposure data were collected at the county level, so exposure misclassification is likely. Besides age and gender, the study did not adjust for other genetic and environmental risk factors for AD mortality. The study found a positive correlation between zinc and copper correlations in soil (r=0.699) but did not adjust for it in statistical models.
Cancer		
Whitehead et al. 2015 Case-control study of copper levels in carpet dust and childhood acute lymphoblastic leukemia (ALL). Case data were obtained from participants in the California Childhood Leukemia	**Exposure:** Copper loading in carpet dust (possible ingestion), collected between 2001-2006. Median copper loading in carpet dust was 110 μg/m^2 for cases (IQR: 48-220) and 130 μg/m^2 for controls (IQR: 50-290). Subjects were split into quartiles for level of copper loading as follows: <50 μg/m^2, 50-130 μg/m^2, 130-290 μg/m^2, and ≥290 μg/m^2.	**Outcomes:** Odds of child ALL did not significantly differ between copper loading quartiles in unadjusted or adjusted models. **Limitations:** Exposure measurement occurred, on average, 1.34 years after ALL diagnosis (or reference date for controls), so temporality cannot be inferred. The study did not account for other potential sources of metal

Table 2-6. Copper Exposure from Environmental Media and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
(CCL) Study (142 cases and 187 controls).	**Inclusion/Exclusion Criteria:** Subjects were enrolled in the CCL Study between Dec 1999-Nov 2007, ages 0-7 years old, and occupying the same home they occupied at the time of diagnosis (or similar reference date for controls). **Covariates Considered/Other Regression Adjustments:** Age at diagnosis (or reference date for controls), sex, race/ethnicity, annual household income, season of dust sampling, year of dust sampling (2001-2003 or 2004-2006).	exposure (inhalation, breastfeeding, diet, etc.). Biological intake of metals was not measured. Dust measurements could not account for variability in dust accumulation in the homes.

AD = Alzheimer's Disease, ALL = acute lymphoblastic leukemia, AS = ankylosing spondylitis, CCL = California Childhood Leukemia, CI = confidence interval, ESR = erythrocyte sedimentation rate, IRR = incidence rate ratio, MND = motor neuron disease, RA = rheumatoid arthritis, RR = relative risk, SMR = standardized mortality ratio, WBC = white blood count

Table 2-7. Serum Copper Levels and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
Cardiovascular		
Ford 2000 **Study Type:** Prospective cohort study of 4,574 adults aged 30-75 who participated in NHANES II from 1976-1980. Mortality data from coronary heart disease reported in NHANES (1976-1992) reviewed.	**Exposure:** Measured **c**opper concentration in blood serum. **Inclusion/Exclusion Criteria:** Subjects were NHANES participants from 1976-1992. Subjects who had prevalent heart disease, no serum copper measurement, missing covariate data, or were lost to follow up were excluded. **Covariates Considered/Other Regression Adjustments:** Age, sex, race, education level, smoking status, systolic blood pressure, serum cholesterol, serum high density lipoprotein cholesterol, body mass index, recreational activity, non-recreational activity, history of diabetes, white blood cell count.	**Outcomes:** 151/4,574 subjects died from coronary heart disease (CHD). Age-adjusted serum copper was 5% higher in subjects who died from CHD than in those who did not (129.8 µg/dl ± 3.7 SD versus 122.9 µg/dl ± 0.5 SD, p=0.072). Odds of death by CHD increased by 6% per 1 µmol/liter increase in serum copper in all models (multiple adjusted OR = 1.10, 95% CI: 1.05, 1.14). Hazard ratios for death by CHD and serum copper quartile showed that subjects in quartiles 3 and 4, but not 2, had significantly higher risk of death by CHD compared to quartile 1 (Q2 OR = 1.84, 95% CI: 0.93, 3.66; Q3 OR = 2.14, 95% CI: 1.21, 3.77; Q4 OR = 2.87, 95% CI: 1.57, 5.25). **Limitations:** Because deaths were identified by matching National Death Index information to Social Security files, some records may have been incorrectly matched or failed to match. Death certificates may be inaccurate. Participants were assumed to be alive if they could not be identified as being deceased.
Reproductive		
De Craemer et al. 2017 **Study Type:** 3 cross-sectional studies of Flemish adolescents (FLEHS I, II, and III), 2002-2015 (n = 1,659 for each study). Participant blood serum, urine, and hair sampled by study researchers.	**Exposure:** Copper levels in blood. Geometric mean blood copper was 699 µ/l (95% CI: 692, 706) for FLEHS I, 821 µ/l (95% CI: 810, 831) for FLEHS II, and 888 µ/l (95% CI: 873, 903) for FLEHS III. **Inclusion/Exclusion Criteria:** Subjects were adolescents aged 14-15. **Covariates Considered/Other Regression Adjustments:** Age, body mass index,	**Outcomes:** Blood copper was negatively associated with production of sex hormones, including E2, fE2, T, fl, and LH, and positively associated with SHBG in both FLEHS I and II cohorts (p<0.05 in all cases). In FLEHS I, blood copper was negatively associated with odds of male genital development (OR = 0.82, 95% CI: 0.692, 0.967). In FLEHS II, blood copper was negatively associated with the age of menarche (OR = -0.264, 95% CI: -0.387, -0.142). In FLEHS III, blood copper was negatively associated with odds of male pubic hair (OR =

Table 2-7. Serum Copper Levels and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
	contraceptive pill usage (females only), smoking status, hour of blood collection, fasting, passive smoking at home, urbanization, season, illness in the last 14 days, weekly alcohol consumption.	0.376, 95% CI: 0.23, 0.591) and male genital development (OR = 0.411, 95% CI: 0.251, 0.649). **Limitations:** The cross-sectional study design only accounts for very recent copper exposure and prevents inference of temporality between blood copper and sexual maturation.
Kasperczyk et al. 2016 Cross-sectional study of sperm quality in 65 fertile men in the southern region of Portland. Semen samples collected by study researchers.	**Exposure:** Copper concentration in seminal plasma (CuS). Participants were grouped into 2 exposure categories: low exposure (Cu-L = 10.2-21.7 μ/dl) and high exposure (Cu-H = 21.8-228 μ/dl). **Inclusion/Exclusion Criteria:** Subjects were males who were healthy, non-smoking, free of drug consumption (including antioxidant medications). **Covariates Considered/Other Regression Adjustments:** Age.	**Outcomes:** TOS Total oxidant status (TOS) was 243% higher, Mn-superoxide dismutase (SOD) activity was 125% higher, and median IL-10 level was 144% higher in the Cu-H group compared to the Cu-L group. Copper exposure was positively correlated with TOS and IL-10 and negatively correlated with granulocyte colony stimulating factor (G-CSF) and granulocyte-macrophage CSF (GM-CSF). Copper exposure was not significantly associated with sperm count, volume, motility, or morphology. **Limitations:** Cross-sectional study design only accounts for very recent copper exposure and prevents inference of temporality between copper exposure and sperm parameters. The sample size was small.
Developmental		
Yang et al. 2020 **Study Type:** Cross-sectional study of 734 mother-infant pairs in Wuhan, China, 2014-2015. Hospitals medical records of participants examined for birth outcomes.	**Exposure:** Serum copper concentrations in umbilical cord blood collected at birth/ **Inclusion/Exclusion Criteria:** Subjects were pregnant women who were residents in Wuhan city with a single gestation who were willing to take complete questionnaires and provide umbilical cord blood samples at delivery.	**Outcomes:** As a continuous variable, log-transformed copper in cord serum was linked with decreased birthweight z-score in all infants (β = -0.49, 95% CI: 0.69, -0.29) and term birth infants (β = -0.47, 95% CI: -0.67, -0.27). In quantile regression analysis, the association between increased copper concentration in cord serum and decreased birth weight was stronger below the 50th percentile for birthweight z-score.

Table 2-7. Serum Copper Levels and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments:	Outcomes and Limitations
	Covariates Considered/Other Regression Adjustments: Maternal age, annual household income, pre-pregnancy BMI, parity, passive smoking during pregnancy, maternal weight gain during pregnancy, fetal sex, gestational age.	**Limitations:** The cross-sectional study design prevents the conclusion of a causal relationship between copper exposure in utero and birthweight. Dietary information was not collected from mothers, so various forms of arsenic (known to have varying effects on birthweight) in cord blood could not be distinguished and so could not be adjusted for.

ARF = acute renal failure, BMI = body mass index, CI = confidence interval, CHD = coronary heart disease; CVD = cardiovascular disease, EAF = electric arc furnace, FEF = mid-expiratory flows, FVC = forced vital capacity, IDR = interdecile range, IHD = ischemic heart disease, IMD = index of multiple deprivation, MND = motor neuron disease, PM = particulate matter, RR = relative risk, SMR = standardized mortality ratio, UF = ultrafine

2.2 DEATH

Inhalation

No toxicity studies were located regarding death of humans following inhalation exposure to copper. Copper in $PM_{2.5}$ has been associated with increased risk of cardiovascular disease mortality (Badaloni et al. 2017; Lavigne et al. 2019; Wang et al. 2020; Ostro et al. 2008; Valdes et al. 2012) and ischemic heart disease mortality (Badaloni et al. 2017; Lavigne et al. 2019, Ostro et al. 2015; Valdes et al. 2012), and cerebrovascular disease mortality (Valdes et al. 2012). Ostro et al. (2008, 2015) specifically noted an increased risk of mortality from cardiovascular disease among Hispanics, and mortality from ischemic heart disease among a cohort of teachers in California (Ostro et al. 2015); however, it is unclear whether it is the copper that is causing these effects, rather than $PM_{2.5}$, which is independently associated with adverse cardiovascular health outcomes, including cardiovascular mortality (EPA 2019). Increased risk of coronary heart disease death was associated with elevated serum copper levels among adult participants from NHANES (Ford 2000). Further study-specific information may be found in Table 2-4; it should be noted that the quality of these studies differs, and study limitations are included.

Acute inhalation LC_{50} values were 45 and 109 mg Cu/m^3 for female and male rats, respectively, following a 4-hour exposure to a copper-containing herbicide (Rush 1991). No death was reported in rats exposed for 4 hours to 1,662 mg Cu/m^3 as copper oxide aerosols (Holbert 1990). In mice exposed to copper sulfate with a Streptococcus aerosols challenge, mortality ranged from 54–70% higher compared to controls, and the mean survival time decreased by 4.2–5.9 days following a single 3-hour exposure to 0.56 mg/m^3 (Drummond et al. 1986). Mortality was one of several indicators that showed impaired immune response to the challenge resulting from copper sulfate exposure (see Section 2.14). When exposed to 0.13 mg/m^3 for 3 hours/day, 5 days/week for 2 weeks, mortality was 25-31% higher and survival time was 1.3–1.5 days lower, compared to controls (Drummond et al. 1986).

Oral

Several case studies have reported death following ingestion of large doses of copper sulfate (Sharma 2011; Gupta et al. 2018; Griswold et al. 2017; Chuttani et al. 1965). For example, death by cardiac arrest following ingestion of copper sulfate crystals was reported in two case studies, one involved a 26-year-old man who intentionally ingested an unknown amount of copper sulfate crystals, and another was a situation where a 60-year-old man accidentally ingested 15–18 mg of copper sulfate as crystals (Gupta et al. 2018; Griswold et al. 2017). In a case series, 7 of 48 individuals admitted with copper sulfate poisoning died (Chuttani et al. 1965). The deaths occurring within 24 hours of ingestion were attributed to

shock, and deaths after 24 hours were likely due to hepatic and/or renal complications. Study authors reported a significant correlation between whole copper levels and the severity of manifestations among the 48 individuals (Chuttani et al. 1965). Deaths, likely due to central nervous system depression and hepatic or renal failure, have also been reported in individuals ingesting "spiritual green water," which contains ≥100 mg Cu sulfate/L (Akintonwa et al. 1989). A prospective cohort study found that the odds of death from cardiovascular disease increased by 6% per 1 µmol/L increase in serum copper (Ford 2000). The study used serum data collected from individuals who participated in a medical examination for NHANES II (1976-1980) and were followed though 1992 (NHANES III). Vital status was determined by matching participant information to Social Security Administration data and the National Death index. The NHANES serum copper levels most likely reflect a combination of dietary intake and occupational exposures (Ford 2000).

An oral LD_{50} for mice was reported as 39.8 mg Cu/kg, however, only 2 mice were tested (Kadammattil et al. 2018). Oral LD_{50} values of 42 and 37 mg Cu/kg were reported in male and female rats, respectively, exposed to an herbicide with 8% elemental copper (Rush 1990b). Following exposure to an algaecide containing 8% elemental copper, oral LD_{50} values of 94 and 118 mg Cu/kg were reported for male and female rats, respectively (Rush 1990c). Increased mortality was observed in rats fed a diet containing 4 mg/g of added copper (133 mg Cu/kg/day) for 1 week, compared to controls (Boyden et al. 1938). Reduced food intake, possibly the result of taste aversion, contributed to the deaths. No death was reported in 10 rats from a single oral dose of 4,034 mg Cu/kg/day as copper oxide (Kuhn 1989a).

Oral exposure animal studies examining copper lethality reported mix results following intermediate- or chronic-durations. Among 10 rats orally exposed to 31 mg Cu/kg/day in water for 15 days, 100% mortality was reported before the end of study period (NTP 1993). However, no death was reported in rats when exposed for 15 days in feed to doses up to 325 mg Cu/kg/day. This difference of vehicle is further demonstrated in the 13-week study where no death was reported in male and female rats exposed daily to 140 and 134 mg Cu/kg/day in feed, respectively (NTP 1993). Similar results were seen in mice. One of five male mice died following exposure to 58 mg Cu/kg/day in water for 15 days (NTP 1993). When exposed to copper in feed, no death was reported in male and female mice exposed daily to 717 to 781 mg/kg/day for 15 days or to 815 to 1058 mg Cu/kg/day for 13 weeks (NTP 1993). Lifetime exposure of mice to 42 mg Cu/kg/day as copper gluconate in drinking water resulted in an average 12.8% reduction of the maximum lifespan (from 986 to 874 days) and an average 14.4% decrease in their mean survival time (Massie and Aiello 1984). Following exposure of 5 New Zealand white rabbits to 30 mg Cu/kg/day as copper hydroxide via gavage for 23 days, 2 were found dead. In the same report, 2 of 8 pregnant rabbits died following exposures on gestation days 7 to 28 (Munley 2003a). In a similar study, 3 of 22

pregnant New Zealand white rabbits were reported dead when exposed to 18 mg Cu/kg/day via gavage over gestation days 7 to 28 (Munley 2003b). No deaths were reported at 15 mg Cu/kg/day (Munley 2003a) or at 9 mg Cu/kg/day (Munley 2003b).

Dermal

No studies were found regarding death in humans following dermal exposure to copper.

One New Zealand white rabbit was reported dead after a 24-hour dermal exposure to 1,613 mg Cu/kg as copper oxide applied to its shaved dorsal epithelial (back skin) surface (Kuhn 1989b). The study author concluded the acute dermal LD_{50} to be >1,613 mg Cu/kg. No death was reported in New Zealand white rabbits exposed to an herbicide (8% elemental copper) with a dose of 160 mg Cu/kg (Rush 1990a). No death was reported in rats dermally exposed to ≤181 mg Cu/kg, as copper 8-quinolinolate, for 6 hours/day, 5 days/week for 4 weeks, applied to the shaved back skin surface (Hagemann 1992).

Other Routes

One case study reported death by multi-organ failure in a 22-year-old man who intentionally intravenously injected approximately 1 g copper sulfate dissolved in water into his right arm (Behera et al. 2007). Another case study reported death by hypoxia and multi-organ failure in a 29-year-old pregnant woman who intentionally exposed her vaginal tissues to an unknown amount of copper sulfate dissolved in water (Motlhatlhedi et al. 2014).

2.3 BODY WEIGHT

Inhalation

No studies were located regarding body weight effects in humans or animals following inhalation exposure to copper.

Oral

Authors did not observe any effects on body weight in a drinking water study in human females exposed to a daily dose of up to 0.1 mg Cu/kg/day following a 2-week exposure period (Pizarro et al. 1999). In addition, no changes in body weight were reported in infants following a daily dose of up to 0.319 mg Cu/kg/day in drinking water for 9 months (Olivares et al. 1998).

No body weight effects were reported in rats following exposure to 50.9 mg Cu/kg/day for 30 days

(Khushboo et al. 2018). Intermediate-duration dietary exposure studies reported 12–27% decreases in body weight gain and 10-28 % decreases of body weight in rats following exposure to 25.5–140 mg Cu/kg/day for 15–91 days (Kumar et al. 2015, 2016a, 2016b; NTP 1993; Rana and Kumar 1980; Tian et al. 2019); in mice following exposure to 10.7–398 mg Cu/kg/day for 15–133 days (Kvietkauskaite et al. 2004; NTP 1993); and in pigs following exposure to 16.5–18.7 mg Cu/kg/day for 46–49 days (Suttle and Mills 1966). An unspecified decrease in body weight gain was observed in pigs exposed to 2.3 mg Cu/kg/day for 88 days (Kline et al. 1971). No effects on body weight were reported in rats at doses of 8–325 mg Cu/kg/day (Abe et al. 2008; Epstein et al. 1982; Kalita et al. 2020; Kumar et al. 2019; NTP 1993; Seven et al. 2018); in mice at doses of 2.4–781 mg Cu/kg/day (Cheng et al. 2020; Kvietkauskaite et al. 2004; NTP 1993); in pigs at 1.7 mg Cu/kg/day (Kline et al. 1971); in rabbits at 6-30 mg Cu/kg/day (Munley 2003a, 2003b; Shen et al. 2005); and in guinea pigs at 9.6 mg Cu/kg/day (Seffner et al. 1997). Additionally, no exposure-related changes in maternal body weight were seen in rats exposed to 130 mg Cu/kg/day for up to 73 days, prior to mating and during gestation (Haddad et al. 1991). Normally an increase in maternal body weight is expected during gestation. A chronic study found no biologically significant body weight effect in mice exposed to 42 mg Cu/kg/day as copper gluconate in drinking water (Massie and Aiello 1984). A 2-year chronic study in monkeys also found no effects on body weight following exposure to 0.77–1.05 mg Cu/kg/day (Araya et al. 2012).

Dermal

No studies were found regarding body weight effects in humans following dermal exposure to copper.

One dermal toxicity study in albino rats did not report any significant differences in body weight or food intake between groups following dermal exposure to 0, 9, 36, or 181 mg Cu/day for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). The substance, copper 8-quinolinolate was applied as an ointment to the shaved back skin surface.

2.4 RESPIRATORY

Inhalation

In humans, copper is a respiratory irritant. Workers exposed to copper dust have reported symptoms such as coughing, sneezing, thoracic pain, and a runny nose (Askergren and Mellgren 1975; Suciu et al. 1981). In an occupational study of 75–100 workers involved with sieving copper dust, lung radiographs revealed linear pulmonary fibrosis, and in some cases, nodulation (Suciu et al. 1981). During the first year of operation, the workers were exposed to an estimated average concentration of 464 mg Cu/m^3; the

exposure levels declined each year due to introduction of mechanization and added protective measures for workers. By the third year, the levels were estimated to average 111 mg Cu/m^3. For reference, the permissible exposure limit (PEL) by OSHA for an 8-hour time weight average (TWA) exposure to copper dusts and mists in general industry is 1 mg/m^3 (OSHA 2020a).

Among sheet metal workers exposed to patina dust (copper-hydroxide-nitrate, copper-hydroxide-sulfate, copper silicate, copper oxide), 6 of the 11 examined workers displayed increased vascularity and superficial epistatic vessels in the nasal mucosa (Askergren and Mellgren 1975); however, copper exposure levels were not reported. Two case studies reported non-occupational respiratory effects following copper inhalation in humans. A 2-year-old female child developed an acute respiratory distress syndrome with dyspnea, bilateral hyperinflation, and interstitial infiltrates of the lungs following inhalation of copper dust (Donoso et al. 2007). These effects were further indicated by cyanosis on the patient. A 24-year-old man developed a deviated septum with persistent sinus pressure and rhinorrhea after spilling molten copper on his face shield and inhaling the associated fumes (Gibson et al. 2011). Multiple studies examined the associations between copper levels in particulate matter and associated respiratory outcomes among general populations. A decline of copper in particulate matter was associated with improved forced vital capacity in a prospective cohort study in the Netherlands (Boogaard et al. 2013). Increased risk of allergic sensitization was positively associated in children exposed to the copper in airborne PM_{10} aerosols in either their current home address or home address at birth (Gehring et al. 2015). Increased odds of asthma symptoms were positively associated with the estimated copper concentrations in PM_{10} at current home address. Measures of forced expiratory volume in one second was negatively associated with copper concentrations in PM_{10} at the child's current home address and with copper concentrations in $PM_{2.5}$ at the birth home address (Gehring et al. 2015). These epidemiological studies examining the occurrence of adverse respiratory health effects from inhalation copper exposure are summarized in Table 2-4. For this table, note that the study quality varies; study limitations are included.

Copper is considered the etiologic agent in an occupational disorder referred to as "vineyard sprayer's lung." This condition is found in vineyard workers that used an anti-mildew agent known as the "Bordeaux mixture" that contains 1– 2.5% copper sulfate and the pH neutralized with hydrated lime (Pimentel and Marques 1969). Published information on this disorder is primarily from case reports (Pimentel and Marques 1969; Pimentel and Menezes 1975; Stark 1981; Villar 1974; Villar and Nogueira 1980). Through alveolar lavage and biopsy, case reports observed interalveolar desquamation of macrophages, formation of histiocytic and noncaseating granulomas containing inclusions of copper, and healing of lesions in the form of fibrohyaline nodules. These data are very similar to those found in

silicosis (Pimentel and Marques 1969; Plamenac et al. 1985). Higher incidences of abnormal columnar cells, squamous metaplasia without atypia, copper containing macrophages, eosinophilia, and respiratory spirals were found in the sputa of smoking and nonsmoking vineyard sprayers, as compared to rural workers from the same geographic region who did not work in the vineyards (Plamenac et al. 1985). The data are limited and there is no accompanying concentration-response information to correlate with the histological data.

The potential for copper to induce respiratory effects has been evaluated in mice, hamsters, and rabbits. Decreased cilia beating was observed in Syrian-Golden hamsters exposed to 3.3 mg Cu/m^3 as copper sulfate for 3 hours (Drummond et al. 1986). This effect was not observed in similarly exposed CD-1 mice. However, in mice repeatedly exposed to 0.12 mg Cu/m^3 as copper sulfate for 3 hours/day, 5 days/week for 1–2 weeks, mice showed increased alveolar wall thickening (Drummond et al. 1986). The severity of the effect increased with the duration of exposure. This alveolar wall thickening was not observed in similarly exposed hamsters (Drummond et al. 1986).

In rabbits (strain not reported) exposed to 0.6 mg Cu/m^3 as copper chloride for 6 hours/day, 5 days/week for 4–6 weeks, the only histological alteration in the lungs was a slight increase in alveolar type II cell volume density that was not considered adverse (Johansson et al. 1984). No functional or morphological alterations were observed in the alveolar macrophages of similarly exposed rabbits (Johansson et al. 1983). Combined, the mice, hamster, and rabbit studies indicate that there may be species differences in response to the inhalation of particulate copper. However, such a difference could also be influenced by the study (i.e., particle size and composition).

Oral

Several case studies reported respiratory effects in humans following both accidental and intentional ingestion of copper sulfate crystals, powder, or liquid, the most common effects being tachypnea (fast breathing) and dyspnea (labored breathing) (Sood and Verma 2011; Gupta et al. 2018; Franchitto et al. 2008; Higny et al. 2014; Sinkovic et al. 2008; Hassan et al. 2010; Cho et al. 2018; Gunay et al. 2006; Yang et al. 2004). Aspiration pneumonia was reported in two cases of intentional copper sulfate ingestion, one in a 45-year-old man and one in a 29-year-old man (Gamakaranage et al. 2011; Franchitto et al. 2008). Diffuse bilateral infiltration of the lungs was observed in a 44-year-old man who intentionally ingested >100 g copper sulfate (Cho et al. 2018).

Data on the potential of copper to induce respiratory effects after oral exposure in experimental animals are limited to a few studies. NTP (1993) found no histological alterations in the lungs of rats orally

exposed to 29 to 325 mg Cu/kg/day as copper sulfate in the diet for 15 or 90 days, respectively, or in mice exposed to 24 or 1,058 mg Cu/kg/day for 15 or 90 days. In pregnant rabbits, post-mortem evaluation of the lungs revealed no exposure-related changes following daily exposure to 7.5–30 mg Cu/kg/day, as copper hydroxide, from gestation days 7 to 28 (Munley 2003a). In a similar study by Munley (2003b), pregnant rabbits were given 18 mg Cu/kg/day as copper hydroxide via gavage. At necropsy, brown liquid was present in the chest cavity of 3 rabbits that died before the exposure period ended, including the one that experienced irregular respiration prior to death. Dark discoloration and/or mottling of lung tissue was observed among the three rabbits (Munley 2003b).

Dermal

Two case studies examined respiratory effects in humans following dermal exposure to copper. A 2-year-old female spilled a copper powder resulting in contact with her facial skin and some inhalation of the powder (Donoso et al. 2007). Her development of acute respiratory distress syndrome was attributed to inhalation of the powder. No effects were attributed to the dermal exposure. A 24-year-old man spilled molten copper on his face at work after his face shield was blown off. He developed a deviated septum with persistent sinus pressure and rhinorrhea (Gibson et al. 2011). This effect was likely caused by inhalation of the substance, but dermal exposure could not be conclusively ruled out as a contributing factor to the deviated septum and rhinorrhea.

No evidence of exposure-related gross lesions was observed in the lungs or trachea of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate applied to the shaved skin on their backs for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

Other Routes

A 40-year-old woman developed acute respiratory distress syndrome after intentionally inserting an unknown amount of copper sulfate into her rectum (Moussiegt et al. 2020). A 41-year-old woman developed respiratory failure with bi-basal pneumonia after intentionally injecting 2.5 g copper glycinate subcutaneously (Oon et al. 2006)

2.5 CARDIOVASCULAR

Inhalation

Human data on cardiovascular effects from inhalation exposure are limited to a few epidemiological studies. Suciu et al. (1981) compared the health outcomes of workers involved in the grinding and sieving

copper dust in 1970 when concentrations in air were high (up to 464 mg/m^3) to the outcomes of workers in 1972 when air concentrations were lower (≤111 mg/m^3). Among workers exposed in 1970, 16% showed arterial hypertension. In contrast 6% of workers in 1973 had arterial hypertension and palpitations. However, the findings from this study are limited because other factors that could have impacted the cardiovascular system were not reported (Suciu et al. 1981). There are a few epidemiologic studies that have looked at various cardiovascular-related mortality outcomes (Badaloni et al, 2017; Occelli et al. 2020; Ostro et al. 2008, 2015; Lavigne et al. 2019; Wang et al. 2020; Valdes et al. 2012). These studies have reported associations between copper air pollution, and cardiovascular mortality and heart disease. The details from these studies are presented in Section 2.1, Table 2-4.

No dose-response toxicity studies were located regarding cardiovascular effects in animals following inhalation exposure to copper.

Oral

Increased risk of death from coronary heart disease was associated with elevated serum copper levels among adult participants from NHANES (Ford 2000); for more information see Table 2-7. A number of case studies have reported cardiovascular effects following intentional or accidental ingestion of various copper compounds, including copper sulfate, copper oxychloride, and copper-8-hydroxyquinolate. The most common symptoms were elevated pulse rate, low blood pressure, and tachycardia (Sood and Verma 2011; Gupta et al. 2018; Franchitto et al. 2008; Sinkovic et al. 2008; Higny et al. 2014; Cho et al. 2018; Gunay et al. 2006; Griswold et al. 2017). Conversely, two case studies of patients who initially presented with blue-colored vomitus reported elevated blood pressure following accidental ingestion of copper sulfate, one in a 65-year-old man who accidentally ingested approximately 10 g copper sulfate diluted in water, and one in a 22-year-old man who accidentally ingested 1 cup of copper sulfate powder (Higny et al. 2014; Hassan et al. 2010). Ingestion of copper sulfate crystals resulted in fatal cardiac arrest in two cases, one in a 26-year-old man who intentionally ingested an unknown amount of crystals, and another in a 60-year-old man who accidentally ingested 15-18 mg of crystals (Gupta et al. 2018; Griswold et al. 2017). A 30-year-old female who intentionally ingested dehydrated copper sulfate developed swollen feet in addition to low blood pressure (Yadla et al. 2015). Thinned arteries, congested veins, and cardiac failure were reported in a 19-year-old woman who intentionally ingested an unknown amount of a liquid fungicide whose sole active ingredient was 50% copper oxychloride (Gunay et al. 2006).

Male Wistar rats exposed to 50.9 mg Cu/kg/day of copper sulfate for 30 days showed flabby, enlarged, congested hearts upon gross pathology (Khushboo et al. 2018). In another study, male Wistar rats

exposed to the same dose of 50.9 mg Cu/kg/day of copper sulfate pentahydrate for 90 days had a systolic blood pressure 33% higher than controls by the end of the exposure period (Arafa et al. 2019). No histological alterations were observed in the hearts of rats exposed to 29 to 325 mg Cu/kg/day for 15 to 90 days, or in mice exposed to 24 to 1,058 mg Cu/kg/day for 15 to 90 days (NTP 1993).

Dermal

No studies were located regarding cardiovascular effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the heart or aorta of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). The hearts were weighed and no exposure-related differences in relative heart weight were noted for exposed and non-exposed rats.

Other Routes

A 40-year-old woman developed toxic myocarditis followed by a 2-minute-long cardiac arrest after intentionally rectally inserting an unknown amount of copper sulfate (Moussiegt et al. 2020). A 29-year-old pregnant woman developed peripheral vasoconstriction after intentionally vaginally inserting an unknown amount of copper sulfate powder diluted in water (Motlhatlhedi et al. 2014). A 22-year-old man who was found dead had developed subpleural and sub-epicardial hemorrhage after intentionally injecting approximately 1 g copper sulfate into his arm (Behera et al. 2007). A 41-year-old woman developed low blood pressure and rapid atrial fibrillation after intentionally injecting 2.5 g copper glycinate subcutaneously into her arm at three sites (Oon et al. 2006).

2.6 GASTROINTESTINAL

Based on a systematic evaluation of the literature, gastrointestinal toxicity is presumed to be a consistent health effect of oral exposure to dissolved copper salts. A conclusion on gastrointestinal toxicity could not be determined because of the lack of data in human or animals following inhalation or dermal exposure to copper. The full results of the systematic review for the gastrointestinal endpoint are presented in Appendix C.

Inhalation

In workers involved in grinding and sieving copper dust, anorexia, nausea, and occasional diarrhea were reported (Suciu et al. 1981); exposure levels ranged from 111 to 464 mg Cu/m^3 over a 3-year period.

While initial exposure was via the inhalation route, it is possible that the observed gastrointestinal effects were due to oral exposure to copper. Ingestion probably resulted from mucociliary clearance of copper particles deposited in the nasopharyngeal and tracheobronchial regions of the respiratory tract. One case study reported vomiting in a 2-year-old female child following accidental inhalation of a copper powder (Donoso et al. 2007).

No studies were located regarding gastrointestinal effects in animals following inhalation exposure to copper.

Oral

There are numerous reports of acute gastrointestinal effects in humans after intentional or accidental ingestion of copper substances. The most common effects include abdominal pain, nausea, vomiting, diarrhea, and melena (black stool), which typically occur shortly after ingestion and are not persistent (Araya et al. 2001, 2003a, 2003b, 2003c; Gotteland et al. 2001; Knobeloch et al. 1994, 1998; Olivares et al. 2001; Pizarro et al. 1999, 2001). Gastrointestinal ulcerations and hemorrhaging were also observed following copper sulfate ingestion in several case studies (Gamakaranage et al. 2011; Du and Mou 2019; Franchitto et al. 2008; Griswold et al. 2017; Malik and Mansur 2011; Lubica et al. 2017). There have been several reports of upper gastrointestinal effects, including oral mucositis, pharyngeal edema, and odynophagia, following copper sulfate ingestion (Higny et al. 2014; Hassan et al. 2010). Inflammation of the gallbladder was observed in two cases: one in a 19-year-old woman who intentionally ingested an unknown amount of pesticide containing copper oxychloride, and another in a 40-year-old man who intentionally ingested 50 mL of a solution containing 33.5% weight by volume copper-8-hydroxyquinolate (Gunay et al. 2006; Yang et al. 2004).

Several epidemiological studies examined the occurrence of gastrointestinal symptoms in communities exposed to elevated levels of copper in drinking water; they are described in detail in Table 2-5. Gastrointestinal symptoms, including abdominal pain and diarrhea, were associated with copper exposure in several populations (Eife et al. 1999; Knobeloch et al. 1994, 1998; Pizarro et al. 2007). Conversely, two studies did not find significant associations of gastrointestinal symptoms with copper in drinking water (Buchanan et al. 1999; Pettersson et al. 2003). Specifically, Buchanan et al. (1999) observed no increased risk of gastrointestinal symptoms when comparing household waters with copper concentrations <1.3, 2 to 3, or >3 mg Cu/L (Buchanan et al. 1999). Pettersson et al. (2003) observed no increased hazard of diarrhea or vomiting among homes with children and copper concentrations in water ranging from 0.01 to 5 mg Cu/L.

Repeated exposure studies conducted in adults exposed to copper in drinking water have observed gastrointestinal symptoms (Araya et al. 2003b, 2004; Olivares et al. 2001; Pizarro et al. 1999, 2001). The doses calculated from these studies represent the exposure from copper in drinking water only. Several studies did survey participants on their diets; however, copper intake from normal diets was not considered in the dose. These studies are presented in Table 2-2 and Figure 2-3.

Symptoms of gastrointestinal upset following acute exposure to copper are suspected to be a direct contact effect, in that the symptoms result from the maximum serum concentration (C_{max}) of copper in the gastrointestinal system at a time point rather than the 24-hour intake (Donohue 1997). A study by Wang and Borison (1951) hypothesized a biphasic mechanism of copper sulfate-induced emesis. The effect of copper sulfate on the peripheral nervous system followed by a systemic effect on the central nervous system was associated with the absorbed copper intake (Horn et al. 2014; Wang and Borison 1951). Several studies in mammals have demonstrated that copper sulfate-induced emesis results from contact in the stomach mediated by the vagus nerve (Makale and King 1992) and shown that 5-HT_4 receptors and abdominal vagal afferents are closely associated and play a role in inducing vomiting (Bhandari and Andrews 1991; Fukui et al. 1994).

A study by Pizarro et al. (1999) demonstrated a dose-response relationship between copper sulfate and gastrointestinal symptoms (nausea, vomiting, and abdominal pain) in healthy adult women. Each study participant consumed either 0, 1, 3, or 5 mg/L of copper as copper sulfate in their drinking water daily for 2 weeks with a 1-week rest period before starting a new exposure; dosing for each exposure group was calculated as 0.0006, 0.0272, 0.0731, and 0.124 mg Cu/kg/day, respectively. The incidences of abdominal pain, nausea, diarrhea, and/or vomiting were reported, and no dose-response relationship for copper exposure and diarrhea was found (Pizarro et al. 1999). Abdominal pain, nausea, and vomiting were copper-related, and incidences for these symptoms were significantly higher in groups that consumed ≥0.0731 mg Cu/kg/day (≥3 mg Cu/L) than in groups consuming ≤0.0272 mg Cu/kg/day (≤1 mg Cu/L) (Pizarro et al. 1999). This study and its limitations are further described in Table 2-5. Abdominal pain, nausea, and/or vomiting have also been observed in women drinking water containing 5 mg Cu/L (0.096 mg Cu/kg) copper sulfate or copper oxide for 1 week (Pizarro et al. 2001).

The occurrence of gastrointestinal effects was not significantly different between subjects ingesting copper sulfate or copper oxide, with the same amount of copper in each (Pizarro et al. 2001). Both Pizarro et al. (1999, 2001) studies reported no associations between copper levels in drinking water and the incidence of diarrhea. In Pizarro et al. (1999), 8 of 12 cases of diarrhea presented within the first 2 weeks of exposure in all exposure groups, and then the number of cases declined, regardless of the copper

concentration. In Pizarro et al. (2001), 7 of 10 cases of diarrhea occurred during the first half of the exposure period, differing from other gastrointestinal effects, like nausea, which were uniformly distributed during the study period. Diarrhea appears to be an effect of copper exposure but does not appear to be concentration- or dose-dependent in these studies.

In a 2-month study by Araya et al. (2003b), 19.7% of male and female adults exposed to 0.138 mg Cu/kg/day (6 mg Cu/L) reported at least one gastrointestinal symptom, among nausea, vomiting, diarrhea, and abdominal pain, at some point during the exposure period. Incidence of symptoms was higher for this dose group compared to subjects exposed to ≤0.092 mg Cu/kg/day (2 or 4 mg Cu/L) (Araya et al. 2003b). Araya et al. (2004) observed that at the end of the first week of exposure, gastrointestinal symptoms (nausea, abdominal pain, vomiting, and diarrhea) were significant for females exposed to ≥0.106 mg Cu/kg/day (4 mg Cu/L) and males exposed to ≥0.169 mg Cu/kg/day (6 mg Cu/L). The study showed that the incidence of gastrointestinal symptoms increased with copper exposure (concentration in water and volume of water ingested) and females appeared to be at a higher risk for symptoms than males; however, results did not distinguish between gastrointestinal symptoms. In Araya et al. (2004) nausea and vomiting were the only symptoms that appeared to show a clear dose-response relationship with copper concentration in drinking water. As the duration of exposure increased, the concentration in water necessary to achieve a positive gastrointestinal response increased.

Multiple single-exposure studies have observed that nausea is adults' most reported symptom following ingestion of copper in drinking water (Araya et al. 2001, 2003a, 2003c; Gotteland et al. 2001; Olivares et al. 2001). Olivares et al. (2001) observed an increased incidence of nausea at 0.012 mg Cu/kg (4 mg Cu/L); no nausea was reported by subjects exposed to lower doses. Two studies by Araya et al. (2001; 2003c) reported a threshold of 6 mg Cu/L for increased incidence of nausea. In a multinational study by Araya et al. (2001), no nausea was reported following exposure at doses of ≤0.012 mg Cu/kg (4 mg Cu/L), while nausea occurred in 17/179 adults exposed to 0.018 mg Cu/kg (6 mg Cu/L). In this study, females appeared more sensitive to developing nausea following copper ingestion. In Araya et al. (2003c), a single exposure to 0.09 mg Cu/kg (6 mg Cu/L) resulted in nausea in 50/269 females, with a NOAEL of 0.06 mg Cu/kg for nausea. The study determined that both the copper concentration and the total copper dose are important variables in predicting a gastric response; as the concentration and dose increase, the probability of eliciting nausea increases (Araya et al. 2003c). Nausea was confirmed in adults exposed to 10 mg Cu/L of drinking water, as 9/30 adults reported nausea following a single dose of 0.046 mg Cu/kg (Araya et al. 2003a), and 6/31 adults reported nausea following a dose of 0.03 mg Cu/kg (Gotteland et al. 2001). Olivares et al. (2001) found that the threshold for nausea increased to 8 mg Cu/L when copper was given in an orange-flavored juice (instead of water). Increased incidence of vomiting

was observed in adults following exposure to single doses of 0.018 mg Cu/kg (6 mg Cu/L) and 0.03 mg Cu/kg (10 mg Cu/L) (Olivares et al. 2001; Gotteland et al. 2001).

A study of fifty-six healthy babies who received 2 mg/L of copper sulfate in water daily for 9 months did not report any significant difference in the incidence of gastrointestinal effects (Olivares et al. 1998). Two babies, who were formula-fed, had diarrhea, but this was not likely to be exposure-related, as none of the breast-fed babies had symptoms. Controls were exposed to copper doses ranging between 0.123 to 0.158 mg Cu/kg/day, and experimental infants to doses ranging from 0.248 to 0.319 mg Cu/kg/day (Olivares et al. 1998).

Two studies examined physiological alteration in the intestines of adults exposed to copper in drinking water (Araya et al. 2003a; Gotteland et al. 2001). Araya et al. (2003a) observed delayed gastric emptying time, as copper induced a significant delay in increasing the antral area of the stomach in adults exposed to a single dose of 0.046 mg Cu/kg (10 mg Cu/L of drinking water). The antral area is the lower part of the stomach that surrounds the portal entry to the small intestines. The effect occurred during the first hour after fasting adults (15 males, 15 females) were given a bolus of 10 mg Cu/L of copper sulfate solution. Three men and six women experienced nausea following ingestion. The delay in decreasing antral area indicated the continued presence of the ingested solution in the stomach and reflected an increase in the gastric emptying time for both males and females. There was no significant relationship between the delay in gastric emptying and the presence or absence of nausea (Araya et al. 2003a). A study by Gotteland et al. (2001) was designed to evaluate whether there was a change in the permeability of the gastric and intestinal mucosa following exposure to a bolus intake of a 10 mg/L copper sulfate solution. Twenty percent of subjects experienced nausea, and 5% reported vomiting. The copper sulfate solution contained either sucrose or lactulose/mannitol mixture to determine if copper sulfate influenced permeability of the stomach and intestinal membrane. There was a 36.5% increase in gastric permeability to sucrose following the bolus ingestion of 10 mg Cu/L copper solution, but a dose of 0.03 mg Cu/kg elicited no adverse effect. However, no alterations in intestinal permeability to lactulose/mannitol were found. The increased gastric permeability was independent of gastrointestinal nausea/vomiting response (Gotteland et al. 2001).

Gastrointestinal effects have been reported in multiple animal studies. Histological lesions and disruptions in intestinal microbiota homeostasis were reported in the cecum and rectum of mice exposed to 2.4 mg Cu/kg/day, as copper chloride, for 90 days (Cheng et al. 2020). The histological observations included increased thickness of outer muscularis and smooth muscle fiber, widened submucosa, decreased goblet cells, blunting of intestinal villi, and severe atrophy of the central lacteal. Changes in microbiota

homeostasis were reflected by changes in the microbial strain composition of the biota, with some strains increasing and others decreasing. The microbial changes were observed in the cecum and more pronounced in the rectum of the mice (Cheng et al. 2020). A different study showed focal intestinal ulceration in mice exposed to 4 mg Cu/kg/day for 7 days; however, no effects were seen at lower doses (Kadammattil et al. 2018). No gastrointestinal effects were seen in shrews exposed to a single dose of 4 mg Cu/kg; however, 15 episodes of emesis (vomiting) were reported in 4/4 shrews exposed to 48 mg Cu/kg by gavage (Yamamoto et al. 2004). In the same study, no clinical signs of gastrointestinal toxicity were reported in rats at 4–16 mg Cu/kg. At a high dose of 50.9 mg Cu/kg/day, signs of copper toxicity in rats included thickened stomach wall with corrugated mucosa (Khushboo et al. 2018).

Hyperplasia with hyperkeratosis of the squamous mucosa on the limiting ridge separating the forestomach from the glandular stomach was observed in male and female rats exposed to 44–46 mg Cu/kg/day for 15 days or 33–34 mg Cu/kg/day for 13 weeks in their diet, and in mice exposed to 197–216 mg Cu/kg/day for 15 days or 187–267 mg Cu/kg/day for 13 weeks in their diet (NTP 1993). No effects were seen at lower doses of copper in the diet of rats and mice. Animals exposed to copper in drinking water did not show any gastrointestinal effects, including in rats exposed to doses up to 31–36 mg Cu/kg/day and in mice exposed to doses up to 24–36 mg Cu/kg/day, both for 15 days (NTP 1993). Diarrhea was seen in pregnant rabbits exposed to 6–30 mg Cu/kg/day as copper hydroxide administered via gavage from gestation days 7–28 (Munley 2003a, 2003b). One rabbit who died prior to the end of the exposure period showed stomach hemorrhage at necropsy (Munley 2003a). Three rabbits exposed to 18 mg Cu/kg/day that died prior to the end of the exposure exhibited stomach hemorrhage, ulceration, or both (Munley 2003b). With the 18 mg Cu/kg/day diet, diarrhea preceded death in one rabbit, and fetal abortion occurred in two rats; red, discolored stomach lining was seen in one of the rats who aborted (Munley 2003b).

Dermal

No studies were located regarding gastrointestinal effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the stomach, esophagus, pancreas, small intestine, or large intestine of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper-8-quinolinolate applied to shaved areas on their backs for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

2.5 HEMATOLOGICAL

Inhalation

Decreased hemoglobin and erythrocyte levels were observed in workers exposed to airborne copper dust levels of 0.64–1.05 mg/m^3 (Finelli et al. 1981). The OSHA PEL for an 8-hour exposure to copper dusts and mist in general industry is 1 mg/m^3 (OSHA 2020a). Results of hair analysis reveal that the workers had also been exposed to iron, lead, and cadmium (Finelli et al. 1981). A 2-year-old female child who accidentally inhaled a copper powder developed hypoxemia and hemolytic anemia (Donoso et al. 2007).

No studies were located regarding hematological effects in animals following inhalation exposure to copper.

Oral

Numerous case studies have reported hematological effects in humans following ingestion of copper-containing substances. The most common effects are hemolytic anemia, hemoglobinemia, methemoglobinemia, leukocytosis, and reduced reticulocyte count (Cho et al. 2018; Du and Mou 2019; Franchitto et al. 2008; Gamakaranage et al. 2011; Griswold et al. 2017; Gunay et al. 2006; Gupta et al. 2018; Lubica et al. 2017; Malik and Mansur 2011; Mortazavi and Jafari-Javid 2009; Sinkovic et al. 2008; Sood and Verma 2011; Valsami et al. 2012; Yadla et al. 2015; Yang et al. 2004). Cyanosis, a blueish discoloration of the skin usually associated with methemoglobin accumulation, has also been reported in several case studies (Du and Mou 2019; Hassan et al. 2010; Malik and Mansur 2011; Sinkovic et al. 2008; Yang et al. 2004). In a study where 60 adult females were exposed to copper in drinking water daily for 2 weeks, no changes in hemoglobin were seen with doses as high as 0.1 mg Cu/kg/day (Pizarro et al. 1999).

Several studies examined the hematological effects of copper in rats, mice, pigs, and rabbits following intermediate-duration exposures. In rats exposed for an intermediate duration (20-90 days) to doses of 25.5-39.8 mg Cu/kg/day, effects observed included decreased hemoglobin (Kumar et al. 2015); anemia evidenced by reductions in hemoglobin and red blood cell levels (Kumar and Sharma 1987); and decreased erythrocyte, hemoglobin, and hematocrit levels (Rana and Kumar 1980). In female rats exposed to 196 mg Cu/kg/day for 13 weeks, depletion of hematopoietic cells in bone marrow was observed, and in female rats exposed to 66 mg Cu/kg/day for 15 days, effects included decreases in hematocrit, hemoglobin, reticulocyte, and mean cell volume levels and an increase in platelet levels (NTP 1993). In male rats exposed to high doses of 325 mg Cu/kg/day for 15 days, and in female rats exposed to 134 mg

Cu/kg/day for 13 weeks, no hematological effects were observed (NTP 1993). No hematological effects were observed in mice exposed to 10.7 mg Cu/kg/day ad libitum for 19 weeks or 1058 mg Cu/kg/day for 13 weeks (Kvietkauskaite et al. 2004; NTP 1993). One study in rabbits observed changes in blood composition including unspecified decreases in neutrophils, eosinophils, platelets, monocytes, and basophils at 16 mg Cu/kg/day for 50 days (Shen et al. 2005). In pigs, decreased hemoglobin levels and increased erythrocyte count was seen with 16.5–18.7 mg Cu/kg/day after 46-49 days of exposure (Suttle and Mills 1966). Kline et al. (1971) identified a NOAEL of 2.7 mg Cu/kg/day for hemoglobin levels in pigs following 88 days of exposure. Chronic-duration exposure studies in monkeys found no hematological effects after exposure to a daily dose of 0.77 mg Cu/kg/day for 3 years; however, lower hemoglobin levels were observed with a dose of 1.05 mg Cu/kg/day when compared to the controls (Araya et al. 2012).

Dermal

Hypoxemia and hemolytic anemia were observed in a 2-year-old female child who spilled a copper powder on her face and inhaled some of the powder (Donoso et al. 2007). Methemoglobinemia, leukocytosis, and hemolysis were observed in a 53-year-old man following dermal contact with a hot copper sulfate solution (Park et al. 2018). In a child who had been severely burned, copper sulfate crystals were applied to the burn area which resulted in hemolytic anemia, and increased serum and urine copper levels (Holtzman et al. 1966).

No evidence was observed of exposure-related differences in hematological parameters in rats exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved areas of their backs for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Hematological parameters measured included hemoglobin and hematocrit levels.

Other Routes

Intravascular hemolysis was observed in a 22-year-old man who intentionally injected approximately 1 g copper sulfate solution intravenously (Behera et al. 2007). Hemolytic anemia was observed in a 41-year-old female who intentionally subcutaneously injected a total of 2.5 g copper glycinate in solution via syringe among 3 different sites on the forearm (Oon et al. 2006). Methemoglobinemia, elevated blood glucose, and increased white blood cell counts were observed in a 29-year-old woman who intentionally vaginally inserted copper sulfate powder diluted in water (Motlhatlhedi et al. 2014).

2.8 MUSCULOSKELETAL

Inhalation

No studies were located regarding musculoskeletal effects in humans or animals following inhalation exposure to copper.

Oral

Data on musculoskeletal effects in humans following oral exposure to copper is limited but was observed in several case studies. Rhabdomyolysis (breakdown of skeletal muscle) was reported in two cases, one in a 25-year-old man who intentionally ingested an unknown amount of a substance thought to contain copper, and another in a 53-year-old man who intentionally ingested 120 g of copper sulfate (Lubica et al. 2017; Valsami et al. 2012).

Impaired muscle strength was observed in rats exposed to 4 mg Cu/kg/day for 16 weeks, measured by the rotarod test (Kumar et al. 2019). Depressed skeletal growth has been observed in rats administered 39.8 mg Cu/kg/day via gavage; tail length was used to assess skeletal growth (Rana and Kumar 1980).

Dermal

No studies were located regarding musculoskeletal effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the skeletal muscle, femur joint, and sternum of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

2.9 HEPATIC

Based on a systematic evaluation of the literature, hepatic toxicity is a suspected health effect of exposure to copper. The full results of the systematic review for the hepatic endpoint are presented in Appendix C.

Inhalation

Hepatomegaly was observed in workers involved in grinding and sieving copper dust (Suciu et al. 1981); the exposure levels ranged from 111 to 464 mg Cu/m^3 over a 3-year period during which exposure decreased over time. One case study reported elevated aspartate aminotransferase (AST) and bilirubin in a

2-year-old female child who accidentally inhaled a copper powder and got some on her face (Donoso et al. 2007).

No studies were located regarding hepatic effects in animals following inhalation exposure to copper.

Oral

Numerous case reports are available on the hepatic effects in humans following accidental or intentional ingestion of copper substances, including copper sulfate, copper oxychloride, and copper-8-hydroxyquinolate. The most common effect was altered liver enzyme activity, including AST, alanine aminotransferase (ALT), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) (Du and Mou 2019; Griswold et al. 2017; Gunay et al. 2006; Hassan et al. 2010; Malik and Mansur 2011; Mortazavi and Jafari-Javid 2009; Sinkovic et al. 2008; Yadla et al. 2015; Yang et al. 2004). Without providing many details, liver impairment was reported in two cases: one in a 26-year-old man who intentionally ingested approximately 30 g copper sulfate, and another in a 53-year-old woman who intentionally ingested 120 g copper sulfate (Gamakaranage et al. 2011; Lubica et al. 2017). A 17-year-old boy who ingested 10 g cupric sulfate developed hemolytic jaundice, and a 19-year-old woman who ingested an unknown amount of a copper oxychloride-containing pesticide developed jaundice of the conjunctivae (Du and Mou 2019; Gunay et al. 2006). In a compilation of case reports of individuals intentionally ingesting copper sulfate, jaundice was reported in 11 of 53 individuals (Chuttani et al. 1965). Centrilobular necrosis, biliary stasis, elevated serum bilirubin levels and AST activity, plus elevated bile salts in the urine were found in five of the individuals with jaundice. In case reports of lethal ingestion of copper sulfate, jaundice (Akintonwa et al. 1989), centrilobular congestion (Lamont and Duflou 1988), and acute hepatotoxicity (Ahasan et al. 1994) have been reported. O'Donohue et al. (1993) reported a case of an adult with jaundice and hepatomegaly following 3 years of exposure to copper in supplements. For 2 years, the individual ingested 30 mg Cu/day followed by 1 year of 60 mg Cu/day. Among six patients examined for chronic copper poisoning, five patients suffered from hepatopathy (Eife et al. 1999). Copper concentrations in tap water of the examined patients ranged from 0.1 to 16.9 mg/L (Eife et al. 1999). In a study of seven adults receiving capsules containing 0.15 mg Cu/kg/day as copper gluconate, no significant alterations in serum AST, ALP, serum gamma glutamyl transferase (GGT), or LDH activities were found (Pratt et al. 1985).

Several studies examined liver function in infants exposed to elevated levels of copper in drinking water. A NOAEL for liver effects was identified in a study of infants (3 months of age at study initiation) exposed to 0.3 mg Cu/kg/day as copper sulfate in drinking water for 9 months (Olivares et al. 1998). No alterations in total bilirubin levels or serum ALT, AST, or GGT activities were found. A higher

percentage of copper-exposed infants (30.4%) were dropped from the study, as compared to the control group (11.1%). The reasons for being withdrawn from the study were blood sampling refusal (8 infants in the copper group and 2 infants in the control group), protocol transgression (4 infants in the copper group and no infants in the control group), and change of address (5 infants in the copper group and 1 infant in the control group). Two studies of infants up to 12 months of age who were exposed to 0.8 mg Cu/L in household water did not find significant alterations in serum parameters of liver function or alterations in test results from liver ultrasound imaging (see Table 2-5) (Zietz et al. 2003a, 2003b).

A few studies examining hepatic effects in adults exposed to copper did not find significant changes resulting from copper exposure. Liver enzyme levels (ALT, GGT) were not significantly different from baseline levels in female adults following exposure to drinking water containing 0 to 5 mg Cu/L equivalent to doses of 0.0006 to 0.1 mg Cu/kg/day for 2 weeks (Pizarro et al. 1999) or exposure to 5 mg Cu/L equivalent to 0.1 mg Cu//kg/day for 1 week (Pizarro et al. 2001).. Similarly, no alterations in liver function were seen in adults exposed to doses of 0.046 to 0.138 mg Cu/kg/day in drinking water for 2 months (Araya et al. 2003b). No alterations in liver function, evidenced by no change in liver enzyme measurements of alanine aminotransferase and L-GGT, were seen in 11 males and 11 females whose diets were supplemented with 3 mg Cu/day for 6 weeks (O'Connor et al. 2003). In this study, participants served as their own controls, and normal dietary copper intake during the study period was 0.018 mg Cu/kg/day for males and 0.017 mg Cu/kg/day for females. Summing the normal dietary and supplemental exposure resulted in a dose of 0.058 mg Cu/kg/day and 0.067 mg Cu/kg/day for males and females, respectively (O'Connor et al. 2003).

Wilson's disease, Indian childhood cirrhosis (ICC), and idiopathic copper toxicosis (ICT) are diseases largely defined by accumulation of copper in the liver. While Wilson's disease is considered to be a genetic disorder, the etiologies of ICC and ICT are less clear.

Wilson's disease. Wilson's disease is a rare, autosomal, recessive genetic disorder with a prevalence of approximately 30 to 50 cases per million in most parts of the world, with a gene frequency of 0.56% and carrier frequency of 1 in 90 (Rodriguez-Castro et al. 2015). In Western countries, the gene frequency is generally lower at 0.36% (Liu et al. 2017). It is primarily characterized by low levels of serum ceruloplasmin, and by elevated urinary copper excretion, elevated copper levels in the liver, elevated serum free copper, or the presence of copperiedus (which refers to copper deposits in the cornea known as Kayser-Fleischer rings) (Rodriguez-Castro et al. 2015). The accumulation of copper in the liver is due to a genetic mutation in the ATP7B region on chromosome 13q14, resulting in impaired biliary excretion of copper (Liu et al. 2017). Clinical manifestation of the disease varies but is predominantly hepatic or

neurological. Liver effects can range from asymptomatic to liver failure and cirrhosis (Rodriguez-Castro et al. 2015), and three types of liver damage are seen: cirrhosis, chronic active hepatitis, and fulminant hepatic failure. In infants with Wilson's disease, the disease is first characterized by excess hepatic copper despite no histologic indications. Symptoms appear with age and include degenerative change in hepatocytes, fibrosis, and cirrhosis (Scheinberg and Sternlieb 1996). The manifestations of Wilson's disease are not considered to be related to exposure to high levels of copper, but rather the individual's impaired excretion of copper. Individuals with Wilson's disease have elevated levels of hepatic copper when consuming diets with average copper intakes (Taylor et al. 2020; Scheinberg and Sternlieb 1996).

Indian childhood cirrhosis. ICC is a type of liver cirrhosis that was previously considered endemic to India but has since been documented in children of non-Indian origin in multiple countries. It is typically seen in infants and young children 6 months to 3 years in age but has also been diagnosed in children up to 11 years of age (Nayak and Chitale 2013). Predisposition to ICC is suspected to be inherited due to its random occurrence among siblings (up to 22% of siblings affected) and mortality due to liver disease in second-degree relatives of affected children (Nayak and Chitale 2013; Pandit and Bhave 1996). Two widely recognized distinctive features of ICC are coarse, dark brown orcein hepatic staining (representing copper) and intralobular pericellular fibrosis (Pandit and Bhave 1996). Liver copper levels ranging from 790 to 6,654 μg/g dry weight (mean of 939 μg/g) were found in 53 children diagnosed with ICC, as compared to levels of 8–118 μg/g (mean 42–45 μg/g) in 12 controls aged 6 months to >1 year (Bhave et al. 1982). Interpretation of these study results is limited by the small number of controls and the lack of detail on the control group.

No specific genetic susceptibilities have been linked to ICC, and evidence is inconclusive on whether ICC is caused by external exposure to copper or endogenously through dysregulation of copper in the body (Nayak and Chitale 2013; Taylor et al. 2020). Several studies suggest that copper overload and liver injury in ICC-diagnosed children resulted from the use of brass vessels for milk storage (Bhave et al. 1987; Tanner et al. 1983; Tanner 1998). Other studies conversely conclude that excess dietary copper was not a likely cause of copper overload in ICC-diagnosed children, including in a 2006 multi-center study in India that compared 227 cases of confirmed ICC with 426 controls (Nayak and Chitale 2013; Sethi et al. 1993; Taylor et al. 2020). This conclusion is supported by several epidemiological studies of high copper-exposed populations that failed to reveal liver injury in children (Nayak and Chitale 2013).

Idiopathic copper toxicosis. While no precise mechanism is identified, ICT is believed to be caused by an autosomal-recessive genetic defect in copper metabolism combined with excess dietary copper (Müller et al. 1998; Nayak and Chitale 2013). In the literature, ICT is also referred to as ICC-like liver disease,

primary copper toxicosis, and Tyrolean infantile cirrhosis. In general, a few rare, sporadic cases of ICC-like diseases have been reported in 11 countries other than India (Nayak and Chitale 2013). With the exception of a study of ICT in 138 children living in Tyrol, Austria (Müller et al. 1996), most papers describe the clinical course for one to four children or at least one adult (Harada et al. 2020; Nayak and Chitale 2013). Compiling the data from these studies, Müller et al. (1998) found a number of consistent patterns: (1) the age of onset of clinical symptoms occurring before the age of 2 years (infantile onset) or before the age of 5 years (late onset), although onset as late as 10 years has also been observed; (2) rapid progression and death within 2 weeks to 11 months; (3) very high copper levels in the liver, 190–3,360 μg/g dry weight (normal is <50 μg/g); (4) abnormal biochemical markers of liver damage such as aminotransferases, ALP, bilirubin, albumin, and prothrombin time; and (5) marked panlobular and pericellular fibrosis associated with a usually mild inflammatory infiltrate, ballooning degeneration of hepatocytes, and an abundance of Mallory bodies. Previously, ICT was attributed to excess intake of exogenous forms of copper but is more likely attributable to a genetic defect along with abnormal copper metabolism (Harada et al. 2020; Nayak and Chitale 2013). A genealogic investigation conducted by Müller et al. (1996) provided suggestive evidence that the disease is transmitted in an autosomal recessive mode.

The hepatotoxicity of copper in animals is described and investigated in numerous acute- and intermediate-duration oral exposure studies. The majority of these studies used rats; a small number of studies used pigs, mice, rabbits, guinea pigs, and monkeys. Additionally, a number of studies examined animals with genetic defects similar to Wilson's disease, primarily in Long-Evans Cinnamon (LEC) rats and Bedlington terrier dogs. Studies examining animals with these genetic defects were not included in our database on copper toxicity for healthy humans. Additionally, it would be inappropriate to consider the results of these studies for MRL derivation, therefore they are not discussed in this chapter.

Seven-day oral exposure studies in rats and mice identified hepatic effects following exposure to varying doses of copper sulfate (Alhusaini et al. 2018a, 2018b; Kadammattil et al. 2018). Histological observation of the liver in rats showed unspecified reduced relative liver weights, and massive cellular degeneration and necrosis of hepatocytes after oral doses of 39.8 mg Cu/kg/day (Alhusaini et al. 2018b). These findings were supported by elevated serum biomarkers of ALT, AST, LDH, C-reactive protein (marker of inflammation), hepatic nitric oxide (NO), lipid peroxidation, protein expression of cyclooxygenase 2 (COX-2), and DNA fragmentation. Additionally, glutathione (GSH) and superoxide dismutase (SOD) levels were decreased (Alhusaini et al. 2018b). Findings for ALT, NO, malondialdehyde (MDA), GSH, and SOD indicating liver damage were validated at a dose of 119 mg Cu/kg/day and associated with elevated caspase-3, reduced interleukin-10, and hepatic CYP450 (Alhusaini et al. 2018a).

Intermediate-duration oral studies observed hepatic effects in various mammal species at doses ranging from 1.6 to 199 mg Cu/kg/day, primarily in rats (Epstein et al. 1982; Haddad et al. 1991; Hashish and Elgaml 2016; Khushboo et al. 2018; Kumar et al. 2015, 2016a, 2016b; NTP 1993; Rana and Kumar 1980; Sakhaee et al. 2012; Seven et al. 2018; Tian et al. 2019) and mice (Kvietkauskaite et al. 2004; Sakhaee et al. 2014; Wu et al. 2020). Hashish and Elgaml (2016) reported hepatic effects in rats at 1.6 mg Cu/kg/day. The study primarily tested the effects of curcumin as protective against copper sulfate exposure, so only one copper sulfate dose was tested. Liver histopathology showed acute swelling of hepatocytes, a few of which showed coagulative necrosis represented by karyolysis of nuclei, and mild hyperplasia of the epithelium lining of bile ducts with the presence of newly formed bile ductules. Further, hepatic damage due to membrane damage was indicated by elevated levels of hepatic marker enzymes of AST, ALT, ALP, and GGT. Lipid damage was indicated by increased MDA and decreased hepatic catalase, SOD, and GSH (Hashish and Elgaml 2016).

Changes in liver enzyme activity, an early sign of copper liver toxicity, appear to correlate with increased copper concentrations in the liver following oral exposures (Epstein et al. 1982; Kumar et al. 2016a, 2016b; Sakhaee et al. 2012; Tian et al. 2019). In Sprague-Dawley rats given 8.6 mg Cu/kg/day in water, liver copper concentrations and AST increased after 90 days, indicating liver cell damage (Epstein et al. 1982). Sakhaee et al. (2012) did not observe a clear toxicological effect of copper on serum AST and ALT over 56 days in rats exposed to 39.8 mg Cu/kg/day. However, at the higher dose of 79.6 mg Cu/kg/day, AST and ALT increased substantially with time, indicating a dose-response relationship (Sakhaee et al. 2012). In Wistar rats copper sulfate was administered via gavage daily for 30, 60, or 90 days, and a dose-response and duration-related increase of ALT was noted, along with duration-related increases in AST and bilirubin after administration of 25.5–50.9 mg Cu/kg/day (Kumar et al. 2015). Increased serum ALT and AST were reported in rats exposed to 39.8–79.6 mg Cu/kg/day for 30 days (Khushboo et al. 2018; Kumar and Sharma 1987; Tian et al. 2019) and 34 mg Cu/kg/day for 13 weeks (NTP 1993). Kumar et al. (2016b) found that increased ALT and AST correlated with decreased GSH and total antioxidant capacity, and with increased MDA and copper concentrations in liver tissue. These effects were found to be dose- and exposure duration-dependent.

Several studies reported histological findings in the liver at doses higher than the dose tested in Hashish and Elgaml (2016). The livers of male albino rats showed centrilobular necrosis and perilobular sclerosis with nuclear edema following a 20-day exposure to 39.8 mg Cu/kg/day as copper sulfate in the diet (Rana and Kumar 1980). At the same dose, an 8-week study in Wistar male rats reported hepatic lesions characterized by hepatocyte cell swelling, centrilobular necrosis, mild bile retention, and the presence of apoptotic bodies (Sakhaee et al. 2012). Wistar male rats showed a 30% decrease in relative liver weight

and enlarged liver with dark spots and swollen borders, friable and yellow in color, following 50.9 mg Cu/kg/day for 30 days (Khushboo et al. 2018). However, Abe et al. (2008) found no significant difference in liver weight in Fischer 344 rats receiving 62 mg Cu/kg/day for 6 weeks compared to controls. Pregnant Wistar rats exposed to 130 mg Cu/kg/day prior to mating and during gestation (up to 73 days total) showed histopathological changes in the liver including hypertrophy and degeneration of hepatocytes and areas of focal necrosis (Haddad et al. 1991). Seven et al. (2018) reported hepatocellular degeneration and necrosis, and karyolysis and karyomegaly in some hepatocytes with a dose of 199 mg Cu/kg/day in male Sprague-Dawley rats. Increased MDA and decreased GSH, SOD, and catalase were also noted.

NTP (1993) conducted a 13-week study in rats and noted that copper accumulation in the liver of males appeared dose-related, as did chronic active tissue inflammation in both sexes. In females, there were no effects at doses ≤34 mg Cu/kg/day. However, at 68 mg Cu/kg/day, chronic active liver inflammation was reported in 6/10 females, and it was reported in all females at the highest dose of 134 mg Cu/kg/day (NTP 1993). Chronic active inflammation with focal necrosis was first seen in 1 of 10 male rats at 33 mg Cu/kg/day and in all male rats at 66 mg Cu/kg/day. No effects were noted in males exposed to 8–16 mg Cu/kg/day (NTP 1993). In the 15-day studies, males showed no histological changes at 29–92 mg Cu/kg/day, but there was liver inflammation manifested as minimal to mild mononuclear inflammatory cell infiltrate at 198 mg Cu/kg/day. No histological changes were observed in any females in the 15-day studies, with no effects at doses from 31-285 mg Cu/kg/day (NTP 1993). The 15-day NTP animal studies tested lower doses in both sexes but did not evaluate serum chemistry changes. Kumar et al. (2016a) noted dose- and duration-related increases in the severity of histological findings in rats. Findings in the liver tissue included massive fatty liver change and centrilobular necrosis. Some rats showed occasional cell necrosis and petechial hemorrhage; mononuclear cell infiltration indicated hepatitis.

There is some evidence of a dose-response relationship between copper dose and hepatic effects in mice. No effect was seen in liver tissue following exposure to oral doses of 0.4, 1, or 2 mg Cu/kg/day; however, at 4 mg Cu/kg/day, mice showed lower hepatic cellularity and liver hemorrhage (Kadammattil et al. 2018). This finding is supported by observations of granular and vacuolar degeneration in hepatocyte and increased rate of hepatic apoptosis in mice exposed to 4 mg Cu/kg/day for 42 days (Wu et al. 2020). At 5.6 mg Cu/kg/day, mice showed decreased total liver protein, and mice exposed to 10.7 mg Cu/kg/day had significantly reduced liver weight along with increased liver copper levels (Kvietkauskaite et al. 2004). Similar to rats, there were significant increases in AST, ALT, and liver copper concentrations in mice at 79.6 mg Cu/kg/day (Sakhaee et al. 2014). In the NTP (1993) studies, no hepatic effects were seen in female mice exposed to 36 or 781 mg Cu/kg/day in either water or feed, respectively, for 15 days or

following 13-week exposures to doses ≤1058 mg Cu/kg/day. Similarly, no hepatic effects were seen in male mice exposed to 24 or 197 mg Cu/kg/day in either water or feed, respectively, for 15 days or following 13-week exposures to doses ≤815 mg Cu/kg/day (NTP 1993).

Two out of six pigs fed a diet containing 16.5 mg Cu/kg/day for 46 days displayed jaundice, while five out of six pigs given 18.7 mg Cu/kg/day for 49 days displayed jaundice and AST levels elevated by >100%, compared to controls (Suttle and Mills 1966). White Chinese big-ear rabbits showed increases in serum low-density lipoprotein and substantial decreases in triglycerides and very low-density lipoprotein; low-density lipoproteins are produced by the liver, while some triglycerides are from diet (Shen et al. 2005). In pregnant rabbits, post-mortem evaluations found no exposure-related changes in the liver or gallbladder following daily oral exposure to 7.5-30 mg Cu/kg/day, as copper hydroxide, from gestation days 7 to 28 (Munley 2003a). A similarly designed study observed pale liver in animals that died from exposure to 18 mg Cu/kg/day (Munley 2003b). In infant guinea pigs exposed to 9.6 mg Cu/kg/day in water after weaning, no liver histological or organ weight changes were observed (Seffner et al. 1997).

Dermal

Data regarding hepatic effects in humans following dermal exposure to copper are limited to one case study. Elevated serum AST and bilirubin and reduced serum albumin and total protein were observed in a 53-year-old man who slipped and landed on a hot copper sulfate solution on the floor of his workplace, resulting in burns primarily to his legs (Park et al. 2018).

No evidence of exposure-related gross lesions or organ weight difference were observed in the liver of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

Other Routes

Hepatic effects have been observed in humans following intentional injection of copper substances in two cases. A 22-year-old man intravenously injected approximately 1 g copper sulfate mixed with water into his arms and developed substantial hepatic necrosis (Behera et al. 2007). A 41-year-old woman subcutaneously injected 2.5 g copper glycinate (typically used in cattle) and then developed acute hepatic failure with changes in liver enzymes, including elevated AST and reduced ALT (Oon et al. 2006). Elevated AST and ALT were observed in a 29-year-old pregnant woman who intentionally vaginally inserted an unknown amount of copper sulfate powder dissolved in water (Motlhatlhedi et al. 2014).

2.10 RENAL

Inhalation

Data regarding renal toxicity of copper inhalation in humans is limited to a single case study. A two-year-old female child who inhaled an unknown amount of a copper powder and spilled some on her facial skin developed renal failure accompanied by oliguria (low urine output) (Donoso et al. 2007).

No studies were located regarding renal effects in animals following inhalation exposure to copper.

Oral

Renal toxicity was observed in a number of case studies following accidental and intentional ingestion of copper sulfate, the most common effects being elevated serum creatinine, oliguria, hemoglobinuria, and hematuria (blood in urine) (Du and Mou 2019; Franchitto et al. 2008; Gamakaranage et al. 2011; Gupta et al. 2018; Hassan 2010; Lubica et al. 2017; Malik and Mansur 2011; Mortazavi and Jafari-Javid 2009; Sinkovic et al. 2008; Sood and Verma 2011; Yadla et al. 2015; Yang et al. 2004). In some cases, renal failure was reported in conjunction with other manifestations of copper toxicity without providing further details on the nature of the renal effects (Valsami et al. 2012; Griswold et al. 2017; Gunay et al. 2006). In addition to oliguria and hemoglobinuria, a 40-year-old man also developed ketonuria and proteinuria following intentional ingestion of copper-8-hydroxyquinolate. A 19-year-old woman who intentionally ingested an unknown amount of a pesticide containing copper oxychloride developed chronic renal failure (Gunay et al. 2006).

Congestion of the glomeruli and denudation of tubular cells were observed in four individuals who consumed a single lethal dose of copper sulfate (Chuttani et al. 1965). Acute renal failure was reported in 5 of 125 individuals intentionally ingesting large doses of copper sulfate (Ahasan et al. 1994). Hematuria, glycosuria, cylindruria, and proteinuria, all indicative of renal tubular damage, were observed in a child who drank a solution containing approximately 3 g of copper sulfate (Walsh et al. 1977).

Several experimental rat studies confirm that the kidney is a target of copper toxicity in cases of copper overload. The mechanisms for copper renal toxicity are reported to be similar to those seen in the liver. Copper ions are a catalyst for initiating oxidative stress by generating reactive oxygen species or inhibiting enzyme activity (Abuja and Albertini 2001; Musacco-Sebio et al. 2017). Hashish and Elgaml (2016) reported indicators of renal toxicity that occurred with histological changes and biochemical changes in the kidneys of female rats given as low as 1.6 mg Cu/kg/day as copper sulfate in feed for 30 days. Kidney function was impaired as indicated by increased urea and creatinine levels in the serum,

indicating that the kidney was unable to filter and remove these reaction byproducts from the blood (Hashish and Elgaml 2016). Additionally, copper caused significant decreases in renal SOD, GSH, and catalase activity. Lipid peroxidation was further indicated by significantly increased MDA, which is an end-product of lipid peroxidation. Study authors suggest that lipid peroxidation may play a role in copper-induced renal toxicity (Hashish and Elgaml 2016). Histological examination of the kidneys noted degeneration of renal tissues and degeneration of the epithelial lining of some renal tubules.

No renal effects were observed in other species besides rats. An acute-duration study by Kadammattil et al. (2018) reported no significant cellular changes in the kidneys of mice dosed with 4 mg Cu/kg/day as copper sulfate. NTP (1993) reported no renal effects in male and female mice exposed to 24-36 mg Cu/kg/day and 717-781 mg Cu/kg/day for 15 days in water and feed, respectively, and in males and female exposed for 13 weeks to 815–1,058 mg Cu/kg/day in feed. No renal effects were reported in guinea pigs exposed to 6.6 mg Cu/kg/day for 4 weeks pre-weaning followed by 9.6 mg Cu/kg/day for up to 6 months (Seffner et al. 1997).

Several studies reported that copper concentration in the kidneys is proportional to the exposure dose and duration (Kumar et al. 2015, 2016a, 2016b). Kidney dysfunction indicated by significantly elevated blood urea nitrogen (BUN) was observed at 17 mg Cu/kg/day for 13 weeks (NTP 1993), 25.5 mg Cu/kg/day for 90 days (Kumar et al. 2015), and 39.8 mg Cu/kg/day for 8 weeks (Sakhaee et al. 2012). Kumar and Sharma (1987) reported significantly elevated urea levels at 39.8 mg Cu/kg/day for 30 days as an indicator of kidney damage. Increased BUN and serum creatinine correlated positively with free copper levels and hepatic MDA and inversely with GSH and total antioxidant capacity (TAC). The severity of effects increased with the duration of exposure (Kumar et al. 2016b). A second study by Kumar et al. (2016a) reported that the histopathological grading score (measure of severity) correlated positively with BUN and renal MDA levels and correlated inversely with GSH and TAC levels. Elevated MDA and reduced GSH are indicators of inflammatory renal injury.

Copper-induced histological changes in the kidneys of the male rats exposed to 39.8 mg Cu/kg/day as copper sulfate for 20–90 days included necrosis of the tubules, engorgement of uriniferous tubules, nuclear pyknosis, and cell proliferation in the medullary region, hemorrhage, and degeneration of Bowmen’s capsule in the cortex (Kumar et al. 2016a; Rana and Kumar 1980). A 15-day study in rats reported protein droplets in epithelial cells of the proximal tubule following exposure to 10 mg Cu/kg/day as copper sulfate (NTP 1993). The severity of histological findings increased with dose and exposure duration. Kumar et al. (2016a) observed increasing histological damage scores in rats treated for 30, 60, or 90 days, and in rats treated with a higher dose of copper sulfate. The grading criteria used a 1-5 scale to

grade vascular, inflammatory, and cellular degenerative changes in the kidney (Kumar et al. 2016a). Significantly increased urea and creatinine were noted in rats that showed bilateral enlargement of the kidney combined with a dark brown tissue color following 30 days of exposure to 50.9 mg Cu/kg/day as copper sulfate pentahydrate (Khushboo et al. 2018). Sakhaee et al. (2012) reported decreased BUN in rats treated with 39.8 mg Cu/kg/day and observed renal lesions with mild tubular necrosis and hyaline casts in renal tubules following 8 weeks of exposure. Similar histological observations included an increase in cortical tubule protein droplets, destroyed glomeruli corpuscles, plus damage to the epithelial lining of the proximal and distal convoluted tubules in rats exposed to doses ranging from 93–199 mg Cu/kg/day (Alharbi et al. 2019; NTP 1993; Seven et al. 2018).

Histopathological changes occurred in the kidneys of pregnant rats following exposure to 130 mg Cu/kg/day before and during gestation (Haddad et al. 1991). Changes included cloudy swelling of the proximal convoluted tubules due to hydropic degeneration and obliteration of the lumen. As noted with lower doses, similar changes in MDA, GSH, SOD, and catalase levels were observed at higher doses after 21 days of exposure (Seven et al. 2018). In Alharbi et al. (2019), there were similar effects on serum urea, creatinine, MDA, and GSH noted following a 7-day exposure. NTP (1993) reported no renal effects in female rats exposed to 31–285 mg Cu/kg/day for 15 days, and in male rats exposed to 46 or 140 mg Cu/kg/day in feed for either 15 days or 13 weeks, respectively. Alharbi et al. (2019) reported that high doses of copper (119 mg Cu/kg/day) in rats increased DNA fragmentation in kidney tissues, along with other previously noted histological changes. The authors suggested that copper induces apoptosis of renal tubules via activation of Bcl-2 associated X protein and suppression of Bcl-2. Alharbi et al. (2019) showed that copper as copper sulfate decreased expression levels of Bcl-2.

Of two pregnant rabbits that died during exposure to 30 mg Cu/kg/day from gestation days 7 to 28, one rabbit's death was attributed to a hemolytic event causing hemoglobin nephropathy and likely renal failure (Munley 2003a). The other dead rabbit had an autolyzed small liver. No effects were seen in kidneys of pregnant rabbits exposed to doses ≤18 mg Cu/kg/day (Munley 2003a, 2003b).

Dermal

No studies were located regarding renal effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the kidneys or urinary bladder of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Additionally, no differences in renal function parameters, including the concentrations of urea and creatinine in urine, were seen.

2.11 DERMAL

Inhalation

Information regarding dermal effects in humans following copper inhalation are limited to one occupational study and two case studies, both of which involved simultaneous inhalation and dermal exposures. Impregnation of the squamous nasal epithelium and nails with colored copper deposits was seen in 44 workers involved with grinding and sieving of copper dust. These workers made up more than half of the studied workers (Suciu et al. 1981). Forty-three workers had fissured palmo-plantar hyperkeratosis. The workers had been exposed to concentrations ranging from 111 to 464 mg Cu/m^3 over a 3-year period (Suciu et al. 1981). For reference, the PEL by OSHA for an 8-hour TWA exposure to copper dusts in general industry is 1 mg/m^3 (OSHA 2020a).

No studies were located regarding dermal effects in animals following inhalation exposure to copper.

Oral

Several case studies reported dermal effects in humans following intentional and accidental ingestion of copper substances, including copper sulfate, copper oxychloride, and copper-8-hydroxyquinolate. Reports of skin discoloration in human case studies following copper ingestion used descriptive terms such as mauve lavender (Sood and Verma 2011), yellow (Du and Mou 2019), and green (Yadla et al. 2015). Pallor was reported in conjunction with cyanosis or other skin discoloration in two cases (Malik and Mansur 2011; Yadla et al. 2015) and reported as the only observed dermal effect in two cases (Gunay et al. 2006; Mortazavi and Jafari-Javid 2009).

Data on dermal effects in animals following oral exposure to copper is limited to a single study in rats exposed daily to 50.9 mg Cu/kg/day for 30 days, where rough, dry skin with alopecia, most notably on the skin of the abdominal region, was reported (Khushboo et al. 2018).

Dermal

Second-degree chemical burns were reported in two cases following dermal exposure to copper. One case was a 53-year-old man who developed severe burns and cyanosis after spilling a hot copper sulfate solution on his leg; however, this may be due to the substance being hot (Park et al. 2018). Copper sulfate solutions are not usually regarded as caustic (causing chemical burns). Another case was an 11-year-old girl who developed burns on her hands with bilateral cellulitis after a blue substance, later identified as copper sulfate, was deliberately applied to her hands in a traditional healing ceremony (Lapid 2008).

New Zealand white rabbits exposed to 160 mg Cu/kg, as a copper herbicide, for 24 hours showed erythema, edema blanching, eschar, desquamation, and exfoliation at the site of application (Rush 1990a). However, no control was tested to determine if effects resulted from the copper. No evidence of exposure-related skin irritation at the application site was seen in rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

Other Routes

A 22-year-old man developed yellow skin discoloration after intentionally injecting approximately 1 g copper sulfate intravenously (Behera et al. 2007). A 41-year-old woman developed necrotic tissue surrounding injection sites after intentionally injecting 2.5 g copper glycinate subcutaneously (Oon et al. 2006). A 29-year-old pregnant woman developed cyanosis after intentionally vaginally inserting an unknown amount of copper sulfate powder diluted in water (Motlhatlhedi et al. 2014).

2.12 OCULAR

Inhalation

No studies were located regarding ocular effects in human or animals following inhalation exposure to copper.

Oral

Data regarding ocular effects in humans following ingestion of copper is limited to one case study where a 17-year-old boy developed yellowing of his sclera after ingesting 10 g copper sulfate (Du and Mou 2019). This case also presented with light yellowing of the skin and elevated total bilirubin.

Data in animals is limited to one study where histopathological observations in rats exposed to 50.9 mg Cu/kg/day for 30 days revealed paleness of mucous membranes of eyes (Khushboo et al. 2018).

Dermal

Eye irritation has been reported by factory workers exposed to copper dust (Askergren and Mellgren 1975). A 64-year-old man developed a corneal ulcer with gradual vision loss and pigment discoloration in his left eye three years after retiring from a job where he handled copper wire regularly (Cai et al. 2009). It was suspected that a small piece of copper wire was lodged into his eye, causing the ulcer and vision loss.

No evidence of exposure-related ocular effects was seen in rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). The eyes were examined for clinical signs throughout the exposure period.

2.13 ENDOCRINE

Inhalation

Seven cases of enlargement of the Sella turcica, and nonsecretive hypophyseal adenoma, accompanied by obesity, arterial hypertension, and "red facies" were observed in a group of 100 workers exposed to 111–464 mg Cu/m^3 as copper dust (Suciu et al. 1981). The study authors noted that there was a possibility that the clinical manifestations of hypophyseal adenoma or of Cushing's syndrome may have been the result of a disturbance of copper toxicokinetics (Suciu et al. 1981); however, both the significance of this effect and its relationship to copper exposure cannot be determined.

Oral

Three case studies reported endocrine effects in humans following intentional ingestion of copper sulfate. A 26-year-old man developed acute pancreatitis after intentionally ingesting approximately 30 g copper sulfate (Gamakaranage et al. 2011). A 33-year-old woman developed adrenal insufficiency with reduced cortisol after intentionally ingesting an unknown amount of copper sulfate (Sinkovic et al. 2008). A 53-year-old man developed acute pancreatitis after intentionally ingesting 120 g copper sulfate well above reported lethal doses; medical intervention prevented death (Lubica et al. 2017). In all cases, the endocrine effects were not permanent, and the patients made full recoveries within weeks.

Following histological analysis and weighing of the adrenal glands, no toxic effects were identified in young guinea pigs exposed to 9.6 mg Cu/kg/day starting on the first or second day of life and persisting for 6 months (Seffner et al. 1997). No histological differences were observed in the adrenal, mammary, parathyroid, pituitary, preputial, or clitoral glands of rats exposed to doses as high as 31–36 mg Cu/kg/day in water or 285–325 mg Cu/kg/day in feed for 15 days, or to dietary doses of up to 134-140 mg Cu/kg/day for 13 weeks (NTP 1993). Similarly, no difference in these measures were seen in mice exposed to doses as high as 24-62 mg Cu/kg/day for 15 days in water, or to doses as high as 815–1,058 mg Cu/kg/day for 13 weeks in their diet (NTP 1993). In pregnant rabbits, postmortem evaluations found no exposure-related changes in the pancreas following daily exposure to 7.5–30 mg Cu/kg/day as copper hydroxide from gestation days 7 to 28 (Munley 2003a)

Dermal

No studies were located regarding endocrine effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the adrenal, pituitary, or thyroid glands or in the thymus of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on their shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Additionally, there were no organ weight differences in the adrenals or thymus gland; the pituitary gland was not weighed.

2.14 IMMUNOLOGICAL

Inhalation

A single study in humans exposed to copper-containing welding fumes reported asymptomatic systemic inflammation evidenced by a significant increase in blood C-reactive protein (Markert et al. 2016). Men were exposed to a copper concentration of 0.53 mg/m^3 in welding fume for 6 hours per period, for 3 periods over 3 weeks. Welding fumes were generated in a separate room and were connected by a ventilation system to the room where subjects were exposed. The C-reactive protein increase was the highest during the period between the 6th and 24 hours after exposure ended (Markert et al. 2016).

An acute exposure study in mice reported an impaired immune response following inhalation exposure to copper sulfate after presentation of a bacterial challenge (Drummond et al. 1986). In CD-1 mice challenged by an aerosol of *Streptococcus zooepidemicus,* decreased pulmonary macrophage bactericidal activity was observed in those exposed to 0.56 mg Cu/m^3 for 3 hours and in those exposed to 0.12 mg Cu/m^3 for 3 hours/day, 5 days/week for 2 weeks. In the same study, bactericidal activity of alveolar macrophages decreased in mice exposed to 3.3 mg Cu/m^3 for 3 hours or 0.13 mg Cu/m^3 for 3 hours/day, 5 days/week for 2 weeks after exposure to an aerosol of *Klebsiella pneumonia* (Drummond et al. 1986). In a different study, there were no functional differences in macrophages in rabbits exposed to 0.28 mg Cu/m^3 for 6 hours/day, 5 days for 1 month (Johansson et al. 1983).

Oral

Three studies examined immunological effects in humans following copper ingestion. Nine men were studied for immune effects following copper exposure in a metabolic research unit during two separate 18-day study periods (Turnlund et al. 2004). During the first 18-day exposure to 0.02 mg Cu/kg/day in diet, no effects were seen on white blood cells, lymphocytes, immunoglobin, interleukin 2R, or interleukin 6. During the second 18-day period of exposure to 0.1 mg Cu/kg/day, the men had significant changes in lymphocyte levels and significantly less antibody to an influenza strain they were immunized

for, compared to the controls who were also immunized (Turnlund et al. 2004). Reduced ALB and GLB were observed in a 17-year-old boy who ingested 10 g copper sulfate (Du and Mou 2019). In the second study, among 29 patients with chronic copper poisoning from plumbing, one had a natural-killer cell deficiency (Eife et al. 1999). Copper levels in tap water measured in homes of these patients ranged from 0.1 to 16.9 mg/L.

Studies on immunotoxicity following oral exposure to copper in animals are limited to a few studies examining the spleen. In mice, no effects of immunotoxicity were seen following a daily 7-day exposure to 2 mg Cu/kg/day; however, follicular hyperplasia in the spleen was observed at 4 mg Cu/kg/day (Kadammattil et al. 2018). Rats exposed for 30 days to 50.9 mg Cu/kg/day showed congested and enlarged spleens (Khushboo et al. 2018). No histopathological changes were seen in the spleens of rats exposed to doses as high as 31–36 mg Cu/kg/day in drinking water or as high as 285–325 mg Cu/kg/day in feed for 15 days (NTP 1993). There were no changes in the spleen of young guinea pigs following exposure to 9.6 mg Cu/kg/day for 6 months (Seffner et al. 1997).

One study in mice exposed to 5.6 mg Cu/kg/day showed that exposure had a toxic effect on antioxidant defense system enzymes and phenotypic properties of immunocompetent cells of the mice as evidenced by decreases in percentage of suppressor, natural killer, and precursor cells and increases in immunoregulatory index (Kvietkauskaite et al. 2004). In rats exposed to 199 mg Cu/kg/day for 21 days, serum tumor necrosis factor-alpha (TNF) levels were increased 1.55 times greater than in controls (Seven et al. 2018). In pregnant rabbits, postmortem evaluation found no exposure-related changes in the spleen following daily exposure to 7.5–30 mg Cu/kg/day, as copper hydroxide, from gestation days 7 to 28 (Munley 2003a).

Dermal

In some individuals, exposure to copper metal produced pruritic dermatitis. Saltzer and Wilson (1968) reported a case of a woman who had recurrent pruritus on her ring finger and wrist caused by copper metal in her ring and wristwatch. Allergic contact dermatitis has been observed in individuals following a patch test using a copper penny and/or a copper sulfate solution (Barranco 1972; Saltzer and Wilson 1968). Axillary lymphadenopathy was reported in an 11-year-old boy who had copper sulfate crystals intentionally applied to his hands (Lapid 2008).

Five of five male rats exposed to 181 mg Cu/kg, as copper 8-quinolinolate, had higher incidence of necrotic thymic lymphocytes (Hagemann 1992). No evidence of exposure-related gross lesions was observed in the lymph nodes of male and female rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as

copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Additionally, there was no significant change in spleen weight.

2.15 NEUROLOGICAL

Inhalation

Three studies evaluating neurological effects in humans following copper inhalation were located. Headache, vertigo, and drowsiness were reported in factory workers exposed for 3 years, beginning with a max concentration of 464 mg/m^3 and declining over 3 years to 111 mg/m^3 copper dust (Suciu et al. 1981). A 2-year-old girl who accidentally inhaled copper dust that also got on her facial skin experienced sensory impairment (Donoso et al. 2007). An epidemiological study on children aged 8 to 12 years found that airborne copper exposure was significantly associated with poorer motor performance and detectable signs of brain damage (Pujol et al. 2016). Copper levels, primarily attributed to traffic pollution, were measured inside and outside of participant schools; 2,827 children participated in behavioral testing and a subset of 263 children participated in brain imaging. The reaction time in children with higher exposure was reduced, while imaging showed copper associated with higher gray matter concentration in the striatum. Copper also appeared to be related to changes in the architecture of neural tissue diffusion (Pujol et al. 2016). Additional details on this study are in Table 2-4.

Oral

Seven adult females exposed to 0.07 mg Cu/kg/day for 2 weeks resulted in clinical symptoms and increased salivation in six of the subjects and headaches in all subjects (Pizarro et al. 1999). Neurological effects following ingestion of copper substances, including copper sulfate, copper oxychloride, and copper-8-hydroxyquinolate, were also reported in several other cases. The most common effects were headache, dizziness, agitation, and drowsiness (Du and Mou 2019; Gunay et al. 2006; Malik and Mansur 2011; Yang et al. 2004).

No histological changes were observed in the brains of mice exposed to 4 mg Cu/kg/day for 7 days (Kadammattil et al. 2018). In mice exposed to trace levels of copper in drinking water (0.08 mg Cu/kg/day) for 21 days, there was no significant change in learning or memory performance as tested by the step-through tasks and Morris water maze task (Lu et al. 2009). At higher doses in mice, females had a 10% increase in relative brain weight following 13 weeks of exposure to 267 mg/Cu/kg/day, and males had a 13% increase in relative brain weight (NTP 1993). In the NTP (1993) study, no neurological effects were seen following exposure to doses of 44-97 mg Cu/kg/day or 52-267 mg/Cu/kg/day in the males and females, respectively (NTP 1993). Additionally, in the shorter duration 15-day mouse study, no

neurological effects were reported in the males receiving 10 to 58 mg Cu/kg/day/day or the females receiving 15 to 62 mg Cu/kg/day (NTP 1993). Although doses higher than 58 and 62 mg Cu/kg/day were tested, the mortality was high, reducing the ability to quantitatively evaluate statistical significance as compared to controls. Seffner et al. (1997) found no significant differences in brain weight of young guinea pigs exposed to 6.6 mg Cu/kg/day starting 1-2 days after birth and continuing through 4 weeks of preweaning followed by an increase to 9.6 mg Cu/mg/day for up to 6 months. Weakness preceded death in one pregnant rabbit that died from exposure to 18 mg Cu/kg/day from gestation days 7-18 (Munley 2003b).

Multiple neurological effects were observed in rats exposed to doses >4 mg Cu/kg/day for intermediate exposure durations. The lowest LOAEL for neurological effects was 4 mg Cu/kg/day in rats that showed neurobehavioral changes (Kumar et al. 2019). Changes include decreased passive avoidance response (refraining from an act or response that would produce an aversive stimulus), increased immobility time in a forced-swim test, decreased entries in an open-arm test, and decreased exploration time (Kumar et al. 2019). The rats also exhibited impaired muscle strength and coordination in the rotarod test. The severity of the neurotoxic effects increased with dose (Kumar et al. 2019). A feeding study in rats exposed to 250 ppm copper in the diet (daily dose could not be calculated) as copper sulfate reported no effects on spontaneous motor activity (assessed using an actophometer), learning ability (assessed using a pole climbing chamber), or relearning capacity and memory (assessed using a Y-maze) (Murthy et al. 1981). The same study observed 16% and 17% decreases in dopamine and norepinephrine neurotransmitters, respectively. De Vries et al. (1986) did not find significant alterations in corpus striatal dopamine levels in rats exposed to 46 mg Cu/kg/day as copper sulfate in drinking water for 11 months. However, a 25% decrease in the levels of a dopamine metabolite, 3,4-dihydroxyphenylacetic acid, in the corpus striatum was observed.

Serious neurotoxic effects observed in rats exposed to a dose of 25.5 mg Cu/kg/day included impaired motor coordination, cognitive function, and changes in locomotor activity (Kalita et al. 2020; Kumar et al. 2015). Toxicity was demonstrated by reductions in grip strength, fall time latency on a rotarod test, distance traveled, time moving, attention scores, plus an increase in resting time (Kalita et al. 2020; Kumar et al. 2015). Cell death through apoptosis and inflammatory pathways were indicated by increased expression of glial fibrillary acidic protein (GFAP) and caspase-3 (Kalita et al. 2020).

Multiple studies have demonstrated that copper, as copper sulfate, induces oxidative stress in the brain as evidenced by reduced SOD, catalase activity, TAC, and GSH, accompanied by increased brain MDA (Kumar et al. 2016b, 2019). In rats exposed to 39.8 mg Cu/kg/day for 30–90 days, changes in TAC, GSH,

and MDA correlated with functional neurological impairment (Kumar et al. 2016b). This was based on changes in grip strength, plus rotarod, and Y-maze tests results in rats exposed to 39.8 mg Cu/kg/day for 30–90 days; neurotoxicity increased with dose (Kumar et al. 2016b). In a similarly designed study by Kumar et al. (2016b), gliosis, pyknotic nuclei, and glial nodule formation in brain sections of rats were observed with doses of 39.8 mg Cu/kg/day for 60-90 days. More severe histological findings of neuronal loss and vacuolated spaces marked by depletion of myelin at 79.6 mg Cu/kg/day for 60–90 days were observed in a second study by the same authors (Kumar et al. 2016a). Histological findings graded on a 1 to 5 severity scale were positively correlated with MDA levels in the brain and inversely correlated with TAC and GSH levels (Kumar et al. 2016a).

In a separate study by Behzadfar et al. (2017), rats were exposed daily for 21 days starting at doses ≥39.8 mg Cu/kg/day. Dose-dependent reactive oxygen species (ROS) formation was demonstrated by increased hippocampal mitochondrial MDA formation (52%) and decreased mitochondrial GSH (29%). Mitochondrial swelling, an indicator of mitochondrial membrane permeability, was also dose dependent. Additionally, spatial memory of rats was tested using the Morris water maze task, which revealed that the impairment was dose dependent, as escape latency was significantly increased at 39.8 mg Cu/kg/day and distance traveled was increased at 79.6 mg Cu/kg/day. The study reported a NOAEL of 19.9 mg Cu/kg/day for neurotoxicity. The study authors suggested that ROS generation in hippocampal mitochondria due to copper exposure caused memory impairment (Behzadfar et al. 2017).

A study that only tested one dose (50.9 mg Cu/kg/day) in rats for 30 days reported that copper toxicity slowed brain activity and produced a swollen, congested, and edematous brain (Khushboo et al. 2018). Gliosis occurred in the brain of 10/10 female rats exposed to 134 mg Cu/kg/day in their diets for 13 weeks (NTP 1993). In the same study, males exposed to 140 mg Cu/kg/day had a 27% increase in relative brain weight at necropsy compared to controls. No neurological effects were seen at doses of 9–68 mg Cu/kg/day in females and 8–66 mg Cu/kg/day in males (NTP 1993). In the 15-day study, no neurological effects were seen in rats exposed to copper in water or feed at doses of 10–325 mg Cu/kg/day and 10–285 mg Cu/kg/day in males and females, respectively (NTP 1993).

Dermal

No studies were located regarding neurological effects in humans following dermal exposure to copper.

No evidence of exposure-related gross lesions was observed in the peripheral nerve, brain, or spinal cord of rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for

6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Additionally, no differences in brain weight were seen.

Other Routes

A 29-year-old pregnant woman who intentionally vaginally inserted an unknown amount of copper sulfate diluted in water was reported to be cyanotic with a decreased consciousness 12 hours after admission for copper poisoning (Motlhatlhedi et al. 2014). The patient was only responding to pain.

2.16 REPRODUCTIVE

Inhalation

Sexual impotence was reported in 16% of workers (75–100 workers examined) exposed to 111– 464 mg/m^3 copper dust during grinding and sieving operations (Suciu et al. 1981). The significance of this finding is difficult to assess because the study did not include a control group.

No studies were located regarding reproductive effects in animals following inhalation exposure to copper.

Oral

No studies were located regarding reproductive effects in humans following oral exposure to copper. Two cross-sectional studies (described in detail in Table 2-7) reported associations between serum copper levels, presumably resulting from oral ingestion, and reproductive outcomes (De Craemer et al. 2017; Kasperczyk et al. 2016). Due to usual limitations of cross-sectional studies, the results cannot be definitively attributed to copper exposure or blood copper levels.

Many animal studies have examined the reproductive toxicity of copper following oral exposures. Few of those studies examined female reproductive toxicity (Babaei et al. 2012; Munley 2003a, 2003b; NTP 1993). Female mice were exposed to 39.8 or 79.6 mg Cu/kg/day for either 14 or 35 days (Babaei et al. 2012). At both exposure durations serious effects occurred at 39.8 mg Cu/kg/day including a decrease in the number of antral follicles, ovarian cell damage, and a decrease in the corpus luteum. The severity of the antral follicle effects increased with dose. There were also decreases in the numbers of other follicles (i.e., primordial, primary, growing, and secondary), accompanied by severe changes to follicle structure. A 13-week NTP (1993) study in female rats reported chronic active inflammation of the clitoral gland and ovarian cysts in 10/10 rats exposed to 134 mg Cu/kg/day. However, no effects were seen in lower doses

of 9–68 mg Cu/kg/day. The 13-week study in female mice reported cysts in the clitoral glands of 8/10 female mice exposed to 1,058 mg Cu/kg/day (NTP 1993). No effects were seen in the female mice exposed to 52–536 mg Cu/kg/day. In 15-day studies, no histological or vaginal cytology alterations were reported in female rats exposed to 10–285 mg Cu/kg/day or in mice exposed to 15–36 mg Cu/kg/day (NTP 1993). In pregnant rabbits exposed daily to 30 mg Cu/kg/day as copper hydroxide from gestation days 7 to 28 there was an increase in fetal resorptions compare to controls (Munley 2003a). Total mean resorptions per litter were 1.3 for the 30 mg Cu/kg/day dose group compared to 0.3 resorptions for controls. At a dose of 15 mg Cu/kg/day no reproductive differences were observed in the rabbits (Munley 2003a). At the same exposure duration and frequency, two pregnant rabbits aborted pregnancies on gestation day 22 after exposure to 18 mg Cu/kg/day (Munley 2003b).

Multiple studies in male rats and mice exposed to copper for intermediate durations suggest that copper plays a role in spermatogenesis and male infertility. In a study exposing an unspecified number of male mice to 4 mg Cu/kg/day as copper sulfate, the frequency of folded sperm increased but other indicators of abnormal sperm morphology were not significantly altered (Kadammattil et al. 2018). Significant decreases in sperm concentration, count, motility, and viability were reported in rats exposed to ≥39.8 mg Cu/kg/day for 30 days–56 days (Liu et al. 2016; Sakhaee et al. 2012) and in mice exposed to ≥39.8 mg Cu/kg/day once every 2 days for 28-42 days or daily for 42 days (Sakhaee et al. 2016a, 2016b). In rats dosed at 39.8 mg Cu/kg/day, there were decreases in luteinizing hormone (LH) and follicle-stimulating hormone (FSH) (Liu et al. 2016). In mice, histological observations included depletion and vacuolation of seminiferous epithelium and degeneration of the seminiferous tubules at the same dose (Sakhaee et al. 2016a, 2016b). The severity of reproductive toxicity in male animals was found to be dose dependent (Liu et al. 2016; Sakhaee et al. 2012). The signs of reproductive toxicity reported at 39.8 mg Cu/kg/day were also present in two studies that only tested a dose of 50.9 mg Cu/kg/day in rats for 30 or 90 days (Arafa et al. 2019; Khushboo et al. 2018). These effects included significant reductions in testicular weight, testosterone levels, significant increases in sperm head and tail abnormalities, degeneration of epididymides, and testicular degeneration (Arafa et al. 2019; Khushboo et al. 2018). Khushboo et al. (2018) suggests an association between excess oral copper exposure and decreased synthesis of testosterone. Arafa et al. (2019) had similar findings, and study authors linked this to the downregulation of AT1R and ACE1 protein levels in testicular tissues. An in vitro study of rabbit spermatozoa exposed to copper sulfate found sperm abnormalities including altered anterior part of the sperm head and in the connection segment (Roychoudhury et al. 2010).

As previously mentioned, Liu et al. (2016) reported that several markers of male reproductive toxicity were dose dependent in rats. Additionally, at the highest dose tested (79.6 mg Cu/kg/day) in their study, a

significant increase in the sperm malformation rate and a decrease in testosterone were noted (Liu et al. 2016). In a separate study in rats by Babaei and Abshenas (2013), significantly decreased sperm count, percentage of live spermatozoa, sperm motility and testicular weight were seen after 56 days of exposure to 79.6 mg Cu/kg/day but not after 28 days of exposure. In two studies, male mice exposed to 79.6 mg Cu/kg/day had changes in sperm parameters similar to those seen in rats, in addition to histological changes including shrinkage and degeneration of seminiferous tubules, moderate to severe degeneration of germinal layers, significantly decreased Sertoli cells nuclei diameter and epithelial height, and significantly less meiotic index (Kheirandish et al. 2014; Sakhaee et al. 2014). Intermediate-duration studies by NTP (1993) found a significant 27% increase in relative right testis weight in male rats exposed to 140 mg Cu/kg/day for 13 weeks compared to controls; however, no histological or sperm morphology alterations were seen in male rats exposed to 66 mg Cu/kg/day. In male mice exposed to 187 mg Cu/kg/day, relative right testis weight was 12% greater than controls but no histological changes in the reproductive system or sperm morphology alterations were seen in mice exposed to doses up to 398 mg Cu/kg/day (NTP 1993).

Dermal

No studies were located regarding reproductive effects in humans following dermal exposure to copper.

There was no evidence of exposure-related gross lesions in the seminal vesicle, testis, or epididymis of male rats, or in the uterus, vagina, or ovaries of female rats. Dermal exposures in both the male and female rats were 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, applied to shaved area on their backs for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992). Additionally, there were no exposure-related differences in organ weights for the ovaries of females or the testes of males.

Other

A 29-year-old pregnant woman who intentionally vaginally inserted an unknown amount of copper sulfate powder diluted in water experienced a blue vaginal discharge, blue colored mucous on the cervix surface, and vaginal loss of amniotic fluid (Motlhatlhedi et al. 2014).

2.17 DEVELOPMENTAL

Inhalation

No toxicity studies were identified for developmental effects in humans and animals following inhalation exposure to copper. A retrospective birth cohort study, described in Table 2-4, did not report any associations of copper in the inhaled particulate matter with low birth weight (Pedersen et al. 2016).

Oral

No studies were located regarding developmental effects of humans following oral exposure to copper. A case-control study of mothers who had given birth to children with neural tube defects did not find any difference in the mean copper levels reported for their drinking water compared to mothers of children without neural tube defects (Longerich et al. 1991).

Data on the developmental toxicity of copper in experimental animals is limited. No significant difference was reported for the number of implantations, non-viable embryos, resorbed embryos, or mean embryo weight from pregnant mice exposed to 4 mg Cu/kg/day for 7 days from the 7^{th} to 12^{th} days of pregnancy as compared to controls (Kadammattil et al. 2018). After pregnant rats were exposed to 130 mg Cu/kg/day in diet before and during gestation, their offspring showed delayed growth and development (Haddad et al. 1991). In 11.5-day-old embryos, significant decreases in mean somite number, crown-rump length, and yolk sac diameter were observed. In 21.5-day-old fetuses and newborns, delayed ossification was observed in the cervical and cauda vertebrae, sternum, metacarpals, forelimb phalanges, metatarsals, and hindlimb phalanges (Haddad et al. 1991).

Pregnant rabbits were exposed to 0, 7.5, 15, or 30 mg Cu/kg/day, as copper hydroxide, from gestation days 7 to 28. The fetuses were removed and examined on gestation day 29 (Munley 2003a). Those exposed to the highest dose had 12% reduced mean weight compared to controls. Four of the fetuses were observed to have protrusion of intestines at the umbilicus (omphalocele) (Munley 2003a). In a similarly designed study, maternal exposure to 18 mg Cu/kg/day resulted in significantly increased incidence of hemivertebra, delayed ossification, and supernumerary ribs when compared to the controls (Munley 2003b).

Dermal

No studies were located regarding developmental effects in humans or animals following dermal exposure to copper.

2.18 OTHER NONCANCER

Inhalation

A few studies have reported metal fume fever, a 24–48-hour illness characterized by chills, fever, aching muscles, dryness in the mouth and throat, and headache, in workers exposed to copper dust or fumes (Armstrong et al. 1983; Gleason 1968). Gleason (1968) reported airborne copper dust concentrations of 0.075–0.12 mg/m^3. This range is lower than the OSHA PEL of 1 mg/m^3 for an 8-hour TWA exposure to copper dusts in general industry. It has been suggested that other metals present in the workplace could have been the causative agent for the metal fume fever, rather than copper. A review by Borak et al. (2000) supports this hypothesis as it reviewed occupational reports of metal fume fever and concluded that there is insufficient evidence to suggests these were caused by copper fumes or dusts as other agents appeared to contribute to reported symptoms.

Oral

Several experimental studies reported various noncancerous effects in rats or monkeys. Male rats ingested kaolin indicating pica behavior following exposure once to 10 mg Cu/kg as copper sulfate pentahydrate (Yamamoto et al. 2004). In the same study, copper sulfate induced emesis (vomiting) in shrews but not pica behavior. Pregnant female rabbits exposed to 9 or 15 mg Cu/kg/day from gestation days 7-28 had a significant 17% and 22% reduction, respectively, in mean food consumption compared to controls (Munley 2003a). In rats, decreases in food consumption and water intake were attributed to oral exposure to 50.9-199 mg Cu/kg/day (Khushboo et al. 2018; Seven et al. 2018). Two chronic studies in monkeys reported no differences in food intake following chronic 3-year oral intakes of 0.77–1.05 mg Cu/kg/day from diet and supplements (Araya et al. 2012).

Dermal

No evidence of exposure-related changes in food intake was recorded in rats dermally exposed to 0, 9, 36, or 181 mg Cu/kg, as copper 8-quinolinolate, on shaved back skin for 6 hours/day, 5 days/week for 4 weeks (Hagemann 1992).

2.19 CANCER

Inhalation

There are limited data for humans and no data for animals on the carcinogenicity of inhaled copper. Although a number of studies examined cancer risk among workers at copper smelters, these papers are

not discussed because the cancer risk was attributed to arsenic exposure rather than to copper. In a study of over 6,700 male workers at a Chinese copper mine, there was a significantly increased risk for cancer (all sites combined) (standardized mortality ratio [SMR] =123; 95% confidence interval [CI] =109–139), a significantly increased risk for stomach cancer (SMR=131; 95% CI=105–161), and a significantly increased risk for lung cancer (SMR=147; 95% CI=112–189) (Chen et al. 1993). The cancer risk increased with the duration of employment and time since first exposure (time between first exposure and cancer diagnosis). The risk was also higher in workers employed in the 1950s, when there was a dramatic increase in production, but poor underground ventilation and dry drilling methods were used which generated high levels of dust. Radon and radon daughters were measured in the underground mines; between 1960 and 1990, radioactivity levels of $1.29x10^{-11}$ Ci/L were recorded. To assess the relative contribution of radon and radon daughters to lung cancer risk, the workers were divided into two groups: underground miners and drilling miners. Increases in lung cancer risk were observed in both groups, and study authors suggested that exposure to radiation did not appear to be responsible for the risk of excess death from lung cancer.

The copper ore from the Chinese mine also contained silica, iron, manganese, arsenic, titanium, and sulfur (Chen et al. 1993). The study authors noted that the arsenic level in the copper was relatively low (0.061%) and did not likely contribute to the lung cancer risk; however, the lung cancer risk from exposure to silica and iron could not be ruled out. A significant increase in the risk of silicosis was observed in the miners. In a 7-year follow-up of this cohort, Chen et al. (1995) calculated the risks of cancer for: all sites (SMR=129; 95% CI=117–142), stomach cancer (SMR=141; 95% CI=116–169), and lung cancer (SMR=152; 95% CI=123–187). All risks were still significantly elevated. This study also conducted a worker smoking survey and found that a higher percentage of the miners were smokers (71.7%) than the control population of local residents (64.3%). The increased smoking rate, along with the exposures to radioactivity, silica, iron, and arsenic, could have contributed to the increased cancer risk. However, a prospective study of populations in Europe found that copper in $PM_{2.5}$ was associated with increased risk of lung cancer (Hazard Ratio (HR) = 1.25; 95% CI=1.01-1.53), while PM_{10} was not associated with increased risk of lung cancer (Hazard Ratio (HR) = 1.14; 95% CI=0.96-1.35) (Raaschou-Nielsen et al. 2016).

Oral

There is a case-control study that examined the relationship between copper loading in carpet dust, which can be ingested, inhaled, or can touch skin, and childhood acute lymphoblastic leukemia (Whitehead et al.

2015). There was no significant difference between the odds for cancer development between those exposed to high levels of carpet dust and those who were not.

Three oral studies examined the carcinogenicity of copper compounds in animals. These studies did not find increases in the occurrence of liver tumors in rats exposed to 130 mg Cu/kg/day as copper acetate for 24 weeks (Kamamoto et al. 1973) or large intestine tumors in rats exposed to weekly to 9 mg Cu/kg/day as an unspecified copper compound for 16 weeks (Greene et al. 1987). In a carcinogenicity study, rats were orally exposed to copper gluconate, 62 mg Cu/kg/day, for 6 weeks, and a significant increase in the number of glutathione *S*-transferase placental form (GST-P) positive single hepatocytes was seen compared to controls (Abe et al. 2008). However, the study authors remarked that the results are not necessarily indicative of carcinogenicity given that the induction of GST-P positive individual hepatocytes could act to protect the liver from exposure (Abe et al. 2008). The same study found evidence of carcinogenicity based on the increased number of GST-P positive lesions, given that injection with *N*-nitrosodiethylamine (DEN), a known carcinogen preceded the oral exposure to copper gluconate (Abe et al. 2008).

Dermal

No studies were located regarding cancer effects in humans or animals following dermal exposure to copper.

Other

Several studies examined the carcinogenicity of copper compounds following parenteral administration. No clear tumor incidence results were observed in a collection of studies that used non-oral exposures, including in: male Wistar rats receiving subcutaneous injections of 2 mg Cu/kg/day as copper acetate (Yamane et al. 1984); male and female F344 rats receiving intramuscular injections of 0.25 or 0.41 mg Cu/kg/day as finely ground copper (Furst 1971); Wistar rats receiving intramuscular injections of 150 mg Cu/kg as copper oxide, 150 mg Cu/kg as copper sulfide, or 70 mg Cu/kg as copper sulfate (Gilman 1962). An increase in the occurrence of renal cell carcinoma was observed in male Wistar rats receiving 3–5 mg Cu/kg as cupric nitrilotriacetate 5 days/week for 12 weeks (Toyokuni et al. 1996). Cupric nitrilotriacetate is a chelated compound of copper that is water soluble.

2.20 GENOTOXICITY

One study in humans examined the genotoxicity of copper following occupational exposure. Workers at a copper smelting plant showed significant increases in DNA damage of peripheral blood leukocytes

relative to controls. However, blood copper concentrations were not associated with the level of DNA damage (De Olivera et al. 2012). Shubber et al. (1998) analyzed blood lymphocytes of women using copper-containing contraceptive intrauterine devices (IUDs). Compared to age- and income-matched control women, those using IUDs had significantly higher frequencies of both chromosomal aberrations and sister chromatid exchanges. In a human study by O'Connor et al. (2003), healthy adults were provided with copper supplements for 6 weeks at doses up to 6 mg/day. There was no evidence of DNA damage to leukocytes. No studies were located regarding genotoxicity in humans after dermal exposure to copper or its compounds.

Several animal studies assessed the genotoxicity of copper sulfate following oral or parenteral exposures; the results of these *in vivo* genotoxicity studies are summarized in Table 2-8. Significant increases in the occurrence of micronuclei and chromosomal aberrations have been observed in chick bone marrow cells and erythrocytes (Bhunya and Jena 1996) and mouse bone marrow cells following exposure to copper sulfate (Agarwal et al. 1990; Bhunya and Pati 1987; Fahmy 2000; Kadammattil et al. 2018; Prá et al. 2008). Rabbits treated with copper sulfate in drinking water showed significant increases in sister chromatid exchanges and chromosomal aberrations (Georgieva et al. 2013). A study of copper sulfate by Tinwell and Ashby (1990) did not find increases in the number of micronuclei in mouse bone marrow cells. Several studies reported DNA damage in blood cells of mice exposed to copper sulfate at doses ranging from 1.25–12.5 mg/kg (Franke et al. 2006; Saleha Banu et al. 2004; Prá et al. 2008). DNA fragmentation was also observed in liver cells of rats after oral exposures to 100 or 300 mg/kg/day of copper sulfate for 7 days (Alhusaini et al. 2018a, 2018b). In Drosophila, exposure to copper sulfate resulted in significant increases in the occurrence of recessive lethals (Law 1938) and DNA damage (Shukla et al. 2011). Sperm abnormalities, including spermatocyte chromosome aberrations, double-headed, and double-tailed sperm, were observed in mice after intraperitoneal exposure (Bhunya and Pati 1987; Fahmy 2000) and oral exposure to copper sulfate (Kadammattil et al. 2018).

There were no significant increases in the occurrence of reverse mutations in *Salmonella typhimurium* (Marzin and Phi 1985; Tso and Fung 1981; Wong 1988) or *Saccharomyces cerevisiae* (Singh 1983). In contrast, Demerec et al. (1951) found an increased occurrence of reverse mutations in *Escherichia coli*. Positive results were found in studies testing for DNA damage including errors in DNA synthesis using viral DNA polymerase (Sirover and Loeb 1976), a reduction in DNA synthesis (Garrett and Lewtas 1983; Sirover and Loeb 1976), oxidative DNA damage (Schwerdtle et al. 2007), and an increase in the occurrence of DNA strand breaks (Anchordoquy et al. 2017; Grillo et al. 2010; Jing et al. 2016; Schwerdtle et al. 2007; Sideris et al. 1988; Sina et al. 1983). Using a comet assay approach, DNA damage was detected in mouse lymphocytes from copper alone and in the presence of curcumin (Urbina-Cano et

al. 2006). Oxidative DNA damage repair was inhibited in human HeLa S3 cells following induction with visible light (Schwerdtle et al. 2007). Unscheduled DNA repair synthesis occurred in rat hepatocytes at copper concentrations of 7.9-78.5 µM, in the presence or absence of hydroxyurea (Denizeau and Marion 1989). An increase in sister chromatid exchange in Chinese hamster cells (Sideris et al. 1988) is consistent with the clastogenic effects observed in *in vivo* assays. Caicedo et al. (2008) found that copper at concentrations of 5 mM (318 mg/L) did not induce DNA damage in human CD4+ T lymphocytes, whereas Husain and Mahmood (2019) found DNA damage to human lymphocytes at copper concentrations of 2.5 mM (159 mg/L). No DNA damage was observed in human blood cells exposed to copper alone (Prasad et al. 2006). The results of *in vitro* genotoxicity studies are summarized in Table 2-9.

Changes in DNA methylation and acetylation caused by exposure to copper can lead to modifications on the epigenome which could potentially have transgenerational effects. Recent evidence indicates that exposure to copper can influence gene expression by binding to metal response elements and also via epigenetic mechanisms (Cheng et al. 2012). Human cell line and animal studies have been used to demonstrate alterations to the epigenome. Melino et al. (2009) suggested that copper might also modulate histone deacetylase (HDAC) activity in *E. coli* cells, a crucial enzyme in the epigenetic machinery. In another study, rats were exposed to 6.5 mg/kg copper in their feed which increased DNA methylation (Ognik et al. 2019). No significant trends in global DNA methylation related to inhalation copper exposure in ICR mice were observed in Rossner et al. (2020). Human hepatocyte Hep3B cells treated with Cu^{2+} at 100–200 mM showed significant decreases in global histone acetylation (Kang et al. 2004). Hypoacetylation detected in histones demonstrates that copper is capable of altering the epigenome (Cheng et al. 2012).

Table 2-8. Genotoxicity of Copper and Copper Compounds In Vivo

Species (test system)	Endpoint	Results	Reference	Compound
Non-mammalian cells:				
Drosophila melanogaster (oral exposure)	DNA damage	+	Shukla et al. 2011	Copper sulfate
Drosophila melanogaster (injection into larvae)	Recessive lethals	+	Law 1938	Copper sulfate
Mammalian cells:				
Albino rat liver cells (oral exposure)	DNA damage	+	Alhusaini et al. 2018a	Copper sulfate
Albino rat liver cells (oral exposure)	DNA damage	+	Alhusaini et al. 2018b	Copper sulfate

Table 2-8. Genotoxicity of Copper and Copper Compounds In Vivo

Species (test system)	Endpoint	Results	Reference	Compound
CF1 mice blood cells (gavage exposure)	DNA damage	+	Prá et al. 2008	Copper sulfate
Human leukocytes (oral exposure)	DNA damage	–	O'Connor et al. 2013	Copper
Human peripheral blood leukocytes	DNA damage	–	De Olivera et al. 2011	Copper
Swiss Albino mice leukocytes (oral exposure)	DNA damage	+	Saleha Banu et al. 2004	Copper sulfate
Swiss Webster mice blood cells (oral exposure)	DNA damage	+	Franke et al. 2006	Copper sulfate
Human blood leukocytes	Chromosomal aberrations	+	Shubber et al. 1998	Copper
Inbred Swiss mice bone marrow cells (intraperitoneal and/or subcutaneous injection)	Chromosomal aberrations	+	Bhunya and Pati 1987	Copper sulfate
New Zealand rabbit blood cells (oral exposure)	Chromosomal aberrations	+	Georgieva et al. 2013	Copper sulfate
White Leghorn chicken bone marrow cells (intraperitoneal injection and oral exposure)	Chromosomal aberrations	+	Bhunya and Jena 1996	Copper sulfate
White Swiss mice spermatocytes (intraperitoneal injection)	Chromosomal aberrations	+	Fahmy 2000	Copper sulfate
White Swiss mice (intraperitoneal injection)	Chromosomal aberrations	+	Agarwal et al. 1990	Copper sulfate
CBA mice bone marrow cells (intraperitoneal injection)	Micronuclei	–	Tinwell and Ashby 1990	Copper sulfate
CF1 mice bone marrow cells (gavage exposure)	Micronuclei	+	Prá et al. 2008	Copper sulfate
Inbred Swiss mice bone marrow cells (intraperitoneal and/or subcutaneous injection)	Micronuclei	+	Bhunya and Pati 1987	Copper sulfate
Swiss Albino mice bone marrow cells (oral exposure)	Micronuclei	+	Kadammattil et al. 2018	Copper sulfate
White Leghorn chicken bone marrow cells (intraperitoneal injection and oral exposure)	Micronuclei	+	Bhunya and Jena 1996	Copper sulfate
White Leghorn chicken erythrocytes (intraperitoneal injection and oral exposure)	Micronuclei	+	Bhunya and Jena 1996	Copper sulfate
White Swiss mice bone marrow cells (intraperitoneal injection)	Micronuclei	+	Fahmy 2000	Copper sulfate
Human blood leukocytes	Sister chromatid exchanges	+	Shubber et al. 1998	Copper
New Zealand rabbit blood cells (oral exposure)	Sister chromatid exchanges	+	Georgieva et al. 2013	Copper sulfate

Table 2-8. Genotoxicity of Copper and Copper Compounds In Vivo

Species (test system)	Endpoint	Results	Reference	Compound
Inbred Swiss mice (intraperitoneal injection)	Sperm abnormalities	+	Bhunya and Pati 1987	Copper sulfate
Swiss Albino mice (oral exposure)	Sperm abnormalities	+	Kadammattil et al. 2018	Copper sulfate
White Swiss mice (intraperitoneal injection)	Sperm abnormalities	+	Fahmy 2000	Copper sulfate

+ = positive results; – = negative results; DNA = deoxyribonucleic acid

Table 2-9. Genotoxicity of Copper and Copper Compounds *In Vitro*

		Results			
Species (test system)	Endpoint	With activation	Without activation	Reference	Compound
Prokaryotic organisms:					
Avian myeloblasts virus, DNA polymerase	Errors in DNA synthesis	NT	+	Sirover and Loeb 1976	Copper chloride
Bacillus subtilis	Rec assay	NT	–	Nishioka 1975	Copper chloride
Salmonella typhimurium TA 102	Reverse mutation	NT	–	Marzin and Phi 1985	Copper sulfate
S. typhimurium TA98, TA102, TA1535, TA1537	Reverse mutation	–	–	Wong 1988	Copper chloride
S. typhimurium TA100	Reverse mutation	NT	–	Tso and Fung 1981	Copper chloride
Escherichia coli	Reverse mutation	NT	+	Demerec et al. 1951	Copper sulfate
Eukaryotic organisms:					
Fungi:					
S. cerevisiae	Recombination	NT	–	Sora et al. 1986	Copper sulfate
Saccharomyces cerevisiae	Reverse mutation	NT	–	Singh 1983	Copper sulfate
Mammalian cells:					
Human blood cells	DNA damage	NT	–	Prasad et al. 2006	Copper chloride
Human lymphocytes	DNA damage	NT	+	Husain and Mahmood 2019	Copper chloride
Human CD4+ T lymphocytes	DNA damage	NT	–	Caicedo et al. 2007	Copper
Human HeLa S3 cells	DNA strand breaks	NT	+	Schwerdtle et al. 2007	Copper sulfate
Human HeLa S3 cells	Impaired DNA damage repair	+	NT	Schwerdtle et al. 2007	Copper sulfate

Table 2-9. Genotoxicity of Copper and Copper Compounds *In Vitro*

Species (test system)	Endpoint	Results		Reference	Compound
		With activation	Without activation		
Human HeLa S3 cells	Oxidative DNA damage	NT	+	Schwerdtle et al. 2007	Copper sulfate
Chinese Hamster Ovary (CHO) cells	DNA strand breaks	NT	+	Grillo et al. 2010	Copper
Chinese Hamster Ovary (CHO) cells	DNA synthesis	NT	+	Garrett and Lewtas 1983	Copper chloride
Chinese hamster V79 cells	DNA strand breaks	NT	+	Sideris et al. 1988	Copper nitrate
Chinese hamster V79 cells	Sister chromatid exchange	NT	+	Sideris et al. 1988	Copper nitrate
Mouse Balb-C lymphocytes (comet assay)	DNA damage	+	+	Urbina-Cano et al. 2006	Copper
Mouse primary lymphocytes	DNA strand breaks	NT	+	Jing et al. 2016	Copper
Bovine ovary cells	DNA strand breaks	NT	+	Anchordoquy et al. 2017	Copper
Rat hepatocytes	DNA strand breaks	NT	+	Sina et al. 1983	Copper sulfate
Rat hepatocytes	Unscheduled DNA synthesis	+	+	Denizeau and Marion 1989	Copper sulfate

+ = positive results; – = negative results; NT = not tested

2.21 COPPER NANOPARTICLES

The following section provides a brief overview on toxicity of copper nanoparticles (CuNPs), including copper oxide nanoparticles (CuONPs) when indicated, and is focused on highlighting findings from experimental animal studies. Occupational populations are more likely to be exposed to CuNPs than the general population, and emissions may come from industrial facilities such as for asphalt and rubber production (Ameh and Sayes 2019). CuNPs are also found in pesticides, fertilizers, and personal care products which may result in its presence in wastewater and sewage (Ameh and Sayes 2019). Crops such as cucumbers or alfalfa can uptake CuNPs from applied agricultural products, and these plants can present another potential source of human exposure (Ameh and Sayes 2019). No epidemiology studies using CuNPs were identified. *In vitro* models using human cell lines have demonstrated that CuNPs induce dose- and time- dependent increases in cytotoxicity, reactive oxygen species and DNA damage (Alarifi et al. 2013; Karlsson et al. 2008). Research on the effects of CuNPs in animals are limited but suggest that CuNPs are toxic in laboratory animals. Several *in vivo* and *in vitro* studies have demonstrated that CuNPs increase the production of reactive oxygen species and reactive nitrogen species both associated in other

studies with serious adverse effects such as genotoxicity, inflammation, apoptosis, and fibrosis (Ameh and Sayes 2019).

The primary target organs for CuNP toxicity include the liver, kidneys, and spleen. Oral administration of copper oxide nanoparticles (CuONPs) can cause significant alterations in the activity of antioxidant enzymes including decreased activity for GSH, catalase, and SOD, plus increases in the lipid peroxidation product, malondialdehyde at doses as low as 5 mg/kg/day in rats (Anreddy et al. 2018). Hepatic effects in rats and mice resulting from acute oral exposure to CuONPs or copper carbonate NPs include an enlarged liver; histopathological changes in liver tissues including congestion, hepatocellular degeneration, and steatosis around the central veins of the hepatic tissue; inflammatory responses; increased mitosis; and significantly diminished CYP450 enzyme activities (El Bialy et al. 2020; Chen et al. 2006; De Jong et al. 2019; Tang et al. 2018). Oral exposure to CuONPs in mice resulted in increased levels of serum ALT, AST, BUN, ALP, and creatinine. Histopathological effects on the kidneys of rats and mice resulting from exposure to CuNPs include glomerular hypercellularity, severe coagulative necrosis, detached tubular epithelia, loss of brush border, and narrowing of tubular lumen (El Bialy et al. 2020; Chen et al. 2016; De Jong et al. 2019). In the spleen, CuNPs resulted in splenic, lymphatic and thymus atrophy and lymphoid depletion in rats and mice after acute oral exposure (El Bialy et al. 2020; Chen et al. 2016; De Jong et al. 2019).

Other adverse effects that were observed in animals exposed to CuNPs include evidence for neurological, gastrointestinal, and pulmonary toxicity. Neurotoxic findings following oral or intravenous CuNP injection in rodents include changes in motor activity, oxidative stress in various brain regions (thalamus, hypothalamus, and medulla) in addition to increasing levels of AChE in the hippocampus and striatum along with decreased exploratory behavior (Fahmy et al. 2020; Luo et al. 2020). In rats and mice, CuNPs altered the cecum microbiome; induced ulcerations in the cecum, colon, and rectum; and caused apoptosis in the duodenum, ileum, and cecum (Cholewińska et al. 2018; De Jong et al. 2019; Luo et al. 2020). A murine pulmonary infection model presents some evidence that CuNPs cause pulmonary inflammation and may reduce lung clearance, thus increasing the risks of pulmonary infections (Kim et al. 2011). No studies to date have directly linked CuNP exposure to carcinogenicity.

Hematological effects in rats and mice from CuNP exposure include decreased red blood cell counts, white blood cell counts, hematocrits, and hemoglobin levels (El Bialy et al. 2020; De Jong et al. 2019). CuNPs appear to affect reproduction in rats and mice as evidenced by decreased sperm count and testes weight in males and decreased FSH, LH, and progesterone in females. Exposure to CuNPs also resulted in ovarian atrophy, disturbance in follicular development, follicular atresia, and reduction in mature

follicles (Kadammattil et al. 2018; Yang et al. 2010). Kadammattil et al. (2018) reported that exposure to CuNPs was more toxic to the reproductive functioning of male mice than copper sulfate exposure. CuNPs resulted in fetal toxicity in rats, including a dose-dependent change in fetal weight, induction of oxidative stress in fetal liver, and increased expression of pro-inflammatory cytokines (Luo et al. 2020).

The toxicokinetics of CuNPs can vary widely depending on particle size, other physicochemical properties, and the preparation. Identified studies were limited to inhalation and ingestion of CuNPs. A higher rate of aggregation in the brain (direct translocation via the olfactory bulb) was observed than in the gastrointestinal system (as seen with copper) (Naz et al. 2020). Copper homeostasis in the brain is maintained by a coordinated system of copper transporters and chaperones which transport copper across the membranes as required (Haywood 2019). CuNPs can be distributed throughout the body. The primary target organs in animals tend to be the brain, liver, kidney, and spleen where the CuNPs induce pathological changes and organ injuries. It is hypothesized that the smaller particle size of CuNPs increases surface area which in turn increases its reactivity with hydrogen ions in gastric fluids. This in turn enables conversion to ionic copper resulting in increased systemic uptake of copper (Ameh and Sayes 2019). The ionic copper is distributed to the liver with some excreted in bile like other copper compounds. The unabsorbed CuNPs are primarily excreted in the feces of mammals with minimal excretion in urine.

Evidence to date suggests that CuNPs and soluble copper compounds share several target organs including the liver, kidney, and stomach. Specifically, since CuNPs are smaller, they can cross the cellular membrane and induce oxidative injury. In addition, the small particle size also assists them in evading phagocytosis and other immune response mechanisms allowing for translocation to other organs (Chen et al. 2006). The overall database for CuNP in mammals is limited to a few studies in rats and mice. Most of the copper nanoparticle toxicity studies use *in vivo* and *in vitro* approaches, and most of the toxicity studies thus far focus on aquatic organisms and/or microorganisms (Chang et al. 2012).

CHAPTER 3. TOXICOKINETICS, SUSCEPTIBLE POPULATIONS, BIOMARKERS, CHEMICAL INTERACTIONS

3.1 TOXICOKINETICS

- Absorption: Copper is absorbed in the gastrointestinal tract, primarily by the small intestine. Copper absorption in adult humans ranges from 12 to 71%, and 75-84% in infants. Dietary copper intake and copper absorption are tightly regulated by copper homeostasis maintenance.
- Distribution: Following absorption, copper is distributed by a 2-phase process. The first phase distributes copper by transport to portal venous circulation where copper is bound to serum protein and ultimately about 75% of this copper is taken up by the liver. In the second phase, copper is bound primarily to ceruloplasmin in the liver, is released to systemic blood circulation, and is redistributed to other organ tissues including the brain, kidneys, muscles, and connective tissues.
- Metabolism: Copper metabolism is largely regulated by copper-transporting P-Type ATPases: ATP7A and ATP7B. Cu(II) reduces to Cu(I) mediated by reductases for copper to transport through cellular membranes.
- Excretion: Bile excretion through feces is the major excretory pathway for copper.. Copper half-lives have been measured in various tissues and were 3.9-21 days in the liver, 5.4–35 days in the kidney, 23–662 days in the heart, and 457 days in the brain.

3.1.1 Absorption

No studies were located that provided data on the rate or extent of absorption following inhalation exposure of copper in humans or animals.

Oral copper absorption occurs in the gastrointestinal tract, primarily in the stomach and small intestine, mostly from the duodenum (Taylor et al. 2020; van den Berghe and Klomp 2009). Copper is absorbed from the gastrointestinal tract as ionic copper or bound to amino acids. Evidence indicates that oral copper absorption is dependent on transport proteins, particularly the high-affinity copper transport 1 (Ctr1) and ATP7A. Active mechanisms for copper absorption from the small intestine likely initially involve transport through Ctr1 into enterocytes. Prior to uptake across the apical membrane by Ctr1, the oxidized state Cu(II) is reduced to Cu(I) mediated by reductases activity at the apical membrane of the gastrointestinal enterocytes (Nishito and Kambe 2018; Ohgami et al. 2006). Cuprous, Cu(I), copper transported by Ctr1 concentrates in the apical membrane and early endosomes of the intestinal epithelial cells (Nishito and Kambe 2018). From the epithelial cells, copper is then transported by the copper

chaperone, antioxidant-1, to ATP7A that then readily exports copper into the blood of the portal venous system through which distribution occurs (Nishito and Kambe 2018). To maintain homeostasis and regulation of internal copper levels, copper absorption decreases with increased consumption of dietary copper (van den Berghe and Klomp 2009). In a study of adult men fed low-copper or high-copper diets, copper hemostasis was maintained, and absorption was similar between the groups (Harvey et al. 2003). Another study of 11 young men administered various copper doses in food over a period of 42–98 days found that absorption efficiencies of 55–56, 36, and 12% at doses of 0.785, 1.68, and 7.53 mg/day, respectively (Turnlund et al. 1989). In humans, the amount of stored copper does not appear to influence copper absorption (Strickland et al. 1972).

Multiple human studies examined the oral absorption of dietary copper and reported absorption rates ranging from 12 to 71% in presumably healthy adults (Harvey et al. 2003; Jacob et al. 1987; Johnson et al. 1992; Strickland et al. 1972; Turnlund et al. 1982, 1983, 1985, 1988, 1989, 2005; Weber et al. 1969). Peak copper absorption, estimated through a non-compartment analysis, occurred 1-2 hours after ingestion of a single oral dose of copper gluconate in a controlled study of obese males (Boullata et al. 2017). In infants, higher absorption rates are reported, ranging from 75-84% (Araya et al. 2003d; Domellof et al. 2009; Olivares et al. 2002).

As previously stated, infants appear to have higher absorption rates than those reported in adults (Araya et al. 2003d; Domellof et al. 2009; Olivares et al. 2002). Olivares et al. (2002) did not find significant differences in copper absorption between 1-month-old and 3-month-old infants. A linear relationship between copper intake and retention was found in a metabolic balance study of infants (aged 2–16 weeks) (Dörner et al. 1989). An animal study by Varada et al. (1993) reported age-related differences in copper absorption which was linear and nonsaturable in suckling (16 days of age) and weanling (21–22 days of age) rats, whereas in adolescent rats (6 weeks of age) copper absorption was saturable. The levels of copper retained in the intestine were greater in the suckling rats than in the weanling or adolescent rats (Varada et al. 1993).

Evidence showing sex and age differences in absorption rate are mixed. Several studies in adults did not find differences in copper absorption between older male and female adults aged 60-83 years (Johnson et al. 1992) or between older men (65-74 years) and young men (22-30 years) (Turnlund et al. 1982, 1988). Conversely, Johnson et al. (1992) did find that copper absorption was higher in women (71%) than in men (64%) aged 20-59 years. Obesity did not appear to impair copper absorption in adult males (Boullata et al. 2017). In addition, the composition of the diet can influence copper absorption including plant-based protein diets (Turnlund et al. 1983) and lacto-ovo-vegetarian diets through their impacts on the levels of

bioavailable copper ions (Hunt and Vanderpool 2001). One study of organic diets did not find an effect on copper absorption (Mark et al. 2013). Organic diets refer to eating crops grown without synthetic herbicides, pesticides, fertilizers, or without bioengineered genes.

Competition with other metals in the body can also affect copper absorption in humans and animals as iron and zinc are potential absorption inhibitors for copper uptake across cellular membranes. Increased levels of zinc in the diet result in decreased in copper absorption in humans and rats (Hall et al. 1979; Hoogenraad et al. 1979; Prasad et al. 1978). Turnlund et al. (1988) found that diets with zinc intake slightly above the Recommended Dietary Allowance did not interfere with copper absorption nor increase fecal copper loss. While absorption significantly varied between study groups (48.1% of radiolabeled copper was absorbed when the diet contained 1.3 mg Cu/day and 16.5 mg Zn/day; 37.2–38.5% of radiolabeled copper was absorbed when the diet contained 1.3 mg Cu/day and 5.5 mg Zn/day), both groups had positive copper balance at both levels. A decrease in copper absorption was observed in infants with high intakes of iron (Haschke et al. 1986). Conversely, iron supplements in healthy breastfed infants at 6-9 months of age had no effect on copper absorption (Domellof et al. 2009). Similarly, in adults with an ileostomy, oral iron therapy given as ferrous gluconate did not appear to impair copper absorption even with increasing doses (Troost et al. 2003).

In rats, the absorption of copper appears to be inversely related to the amount of cadmium in the diet (Davies and Campbell 1977). A significant decrease in copper absorption was observed when the copper: cadmium ratio was 1:4. The amount of copper retained in the intestinal mucosal cells was inversely related to cadmium dietary concentration. Conflicting results are reported on the effect of ascorbic acid on copper absorption in humans. Based on a decrease in serum ceruloplasmin levels, Finley and Cerklewski (1983) concluded that a diet high in ascorbic acid resulted in a decrease in copper bioavailability. However, in a study by Jacob et al. (1987), copper absorption was not affected by a high ascorbic acid intake. A decrease in serum ceruloplasmin activity was identified; however, the amount of ceruloplasmin protein was not affected.

The available *in vivo* data do not provide information on the rate and extent of absorption through intact skin following dermal exposure of humans or animals to copper. Following a copper azide explosion that yielded metallic copper and nitrogen fumes, a small increase in serum copper levels was found in the affected worker (Bentur et al. 1988). Animal studies demonstrate that copper can pass through dermal barriers when applied with an appropriate vehicle, (e.g., salicylic acid or phenylbutazone) (Beveridge et al. 1984; Walker et al. 1977). *Ex vivo* studies on human skin report mixed results. Less than 6% of copper deposited on *ex vivo* human skin samples was absorbed (Pirot et al. 1996a, 1996b); copper chloride was

absorbed to a higher extent than copper sulfate (Pirot et al. 1996b). Copper applied transdermally as a tripeptide on *ex vivo* human skin samples permeated the skin and was retained in the stratum corneum, total epidermis, and dermatomed skin (Hostynek et al. 2010). Retention was significant compared to baseline.

3.1.2 Distribution

One study examining respiratory toxicity in rats measured significantly elevated copper levels in the liver and plasma suggesting distribution into these organs (Romeu-Moreno et al. 1994). Elevated but non-statistically significant elevations of copper were measured in kidneys and lung following daily 1-hour inhalation chamber exposure to aerosol copper sulfate for up to 10 days. These results are consistent with more detailed findings of distribution following oral absorption of copper, which is largely similar between humans and animals.

Copper distribution in the body is considered biphasic where ATP7B, predominantly expressed in hepatocytes, is essential for normal distribution of copper. ATP7B has two primary functions: the transfer of copper to a ceruloplasmin that is secreted into the blood and then other organs, and excretion of copper from the body through bile (Guttmann et al. 2018). The first phase is the absorption of copper by enterocytes in the gut and subsequent absorption and distribution by active transport by way of the portal vein (van den Berghe and Klomp 2009). Subsequently, copper levels in the blood rapidly rise as the copper ions bind tightly to albumin and the transcuprin macroglobulin in blood plasma (Moriya et al. 2008). Albumin carries a large portion of the exchangeable copper in peripheral circulation, releasing it to other carriers for cell-specific uptake (Bost et al. 2016; Weiss and Linder 1985). Although passive cellular transport occurs with other metal ions, the absence of copper absorption in Menkes' disease patients and in mice lacking the copper uptake protein, hCTR1, suggest that under normal conditions passive paracellular transport likely does not occur for copper (van den Berghe and Klomp 2009). Prior to phase 1, some copper passes from the small intestines to the large intestines with indigested dietary materials and is then excreted with the feces.

Some dietary copper is transported to the liver where it is bound to ceruloplasmin (a copper-binding serum ferroxide) and released to circulation for distribution. This is the second phase for post-ingestion copper distribution (van den Berghe and Klomp 2009). In the liver, hepatocytes are responsible for the uptake, storage, and regulation of copper; about 75% of copper from the portal vein is taken up by the liver and the rest remains in circulation (Harvey et al. 2005). In an *in vitro* experiment using human hepatic (HepG2) and mammary epithelial (PMC42) cells lines, copper was shown to be transported to Ctr1 in hepatic cells by the plasma protein, α_2-macroglobulin (Moriya et al. 2008). Ceruloplasmin, which

tightly binds six or seven copper atoms (Musci et al. 1993; Saenko et al. 1994), is the most abundant copper protein in the plasma, binding 60–95% of serum copper. The remaining 10-18% is bound to albumin or carried as amino-acid bound copper and transported into other tissues (Harris 1993; Hellman and Gitlin 2002; Kodama et al. 2012; van den Berghe and Klomp 2009). Copper can also bind to α_2-macroglobulin and small peptides. Regulatory copper proteins ATP7A and ATP7B are responsible for the transport of copper out of cells (Taylor et al. 2020). Excessive hepatic copper is transferred from the liver with bile pigments via ATP7B and ultimately excreted with the feces. The brain is the second major site of copper distribution, and copper is also transported to the kidneys, muscle, and connective tissues (Kodama et al. 2012).

Copper crosses the placental barrier and is primarily found in fetal liver in mammals, as part of normal fetal development (Hardman et al. 2007). The fetus obtains copper from maternal serum, either from copper bound to ceruloplasmin, albumin, or anionic amino acids (McArdle 1995). Although copper is found in human breast milk, it is unclear if it is dependent on maternal plasma copper concentrations (Domellof et al. 2004; Khaghani et al. 2010; Kim et al. 2012). Pre-term infants appear to have lower copper stores than full-term infants (Kim et al. 2012). Intraperitoneal and intravenous exposure to ^{67}Copper or ^{64}Copper in non-pregnant and lactating rats showed that approximately 60% of copper in the lactating rats went directly to the mammary gland (Donley et al. 2002). Copper isotopes also rapidly appeared in milk. The ceruloplasmin in milk is attributed to copper in circulation that reaches the mammary gland.

No studies were located regarding the rate and extent of distribution of copper following dermal exposure of humans or animals to copper.

3.1.3 Metabolism

Full copper metabolism is presented in Figure 3-1. Copper metabolism is largely regulated by copper-transporting P-type ATPases ATP7A (also known as Menkes' protein) and ATP7B. Several specific other binding proteins for copper have been identified that are important in the uptake, storage, and release of copper from tissues, most notably ceruloplasmin, which is synthesized in the liver (van den Berghe and Klomp 2009).

In the liver and other tissues, copper is stored bound to metallothionein and amino acids and in association with copper-dependent enzymes. Metallothionein, a metal-binding protein, appears to play an important role in the storage of intracellular copper in a safe compartment and cell survival from both normal and excess copper levels (Tapia et al. 2004). Studies have shown that copper exposure induces

metallothionein synthesis which is important for copper homeostasis (Mercer et al. 1981; Wake and Mercer 1985).

STEAP4, a six-transmembrane epithelial antigen of prostate 4, acts as a metalloreductase and is involved in the reduction of Cu(II) to Cu(I), which is necessary for copper transport across the membrane (Scarl et al. 2017). This reduction reaction occurs at the apical membrane of intestinal epithelial cells (Ohgami et al. 2006).

3.1.4 Excretion

No studies were located regarding the rate and extent of excretion of copper following inhalation exposure of humans and animals. The half-time of copper sulfate in the lungs was estimated to be 7.5 hours after intratracheal instillation of 20 µg copper in rats (Hirano et al. 1990).

Bile is the major pathway for the excretion of copper, as illustrated in Figure 3-1, and primarily excreted in feces. Normally, approximately 2.5 mg Cu/day is excreted in bile (van den Berghe and Klomp 2009). Excessive copper in hepatocytes is excreted into bile from the liver via ATP7B; the reabsorption of biliary copper is negligible as copper binds to components that immobilize it (Farrer and Mistilis 1967; van den Berghe and Klomp 2009). Copper in bile is associated with low molecular weight copper binding components as well as macromolecular binding species (Gollan and Dellar 1973). After the oral administration of radioactive copper as copper acetate in healthy humans, 72% was excreted in the feces (Bush et al. 1955). In six adult men fed ^{63}Cu, 27–46% was excreted in the feces (Turnlund et al. 2005). In humans intravenously administered ^{64}Cu, measurements in feces and urine were negligible (Kjaergaard et al. 2020). In a study in 11 adult men, dietary copper intakes of 0.66, 0.38, and 2.49 mg Cu/day resulted in fecal elimination of 0.65, 0.33, and 2.17 mg Cu/day (Turnlund et al. 1998). A study in rats found an increase in fecal excretion of copper in rats fed a high fiber (potato fiber or sugar beet pulp) diet, likely as a result of reduced copper absorption (Gralak et al. 1996). Bile is also the major excretion pathway in children (Olivares et al. 2002).

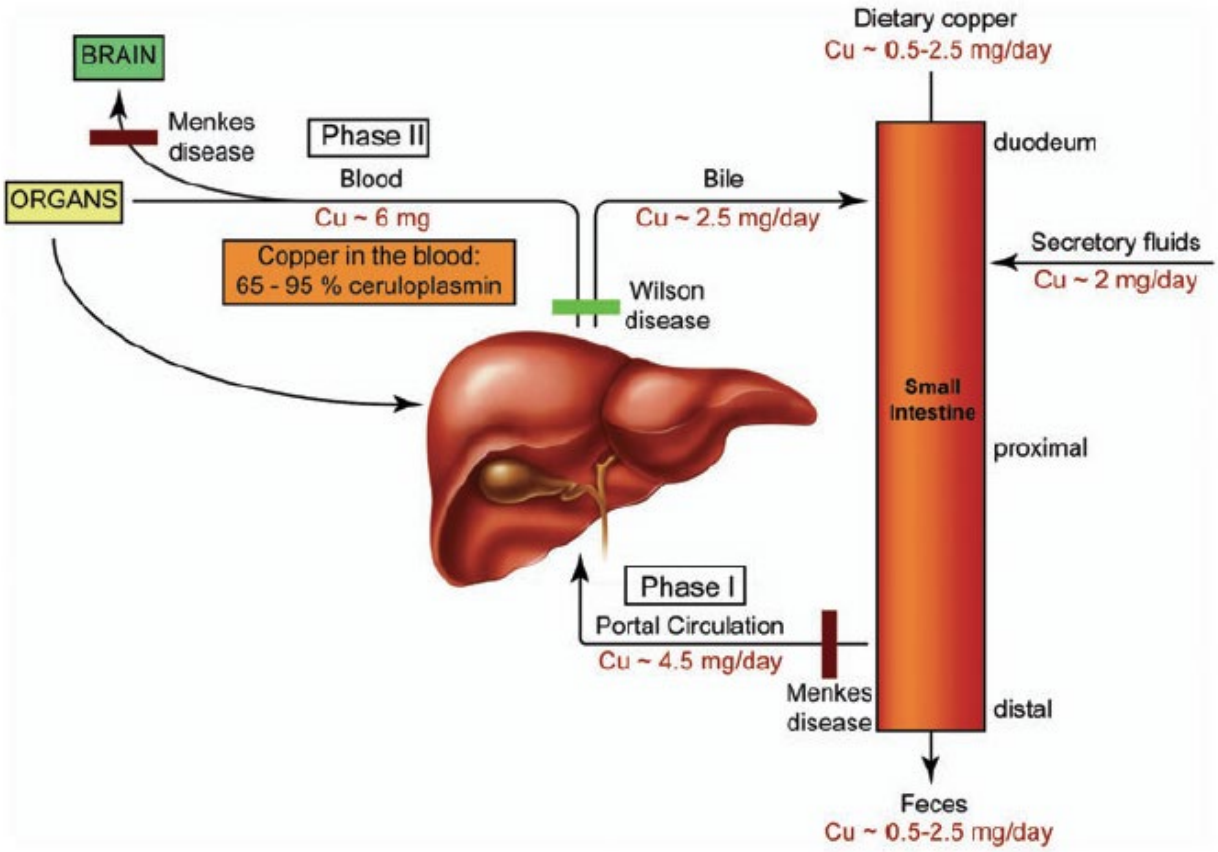
Copper excretion in urine is comparatively low relative to fecal excretion, and normal excretion is expected to be 0.01-0.025 mg Cu/day (Bost et al. 2016). In six adult men fed a diet with ^{63}Cu, 1.3–2.1% was excreted in the urine (Turnlund et al. 2005). One study in humans reported that urinary copper excretion in adult females (mean: 18.7 µg/24 hours) was lower than in adult males (mean: 26.2 µg/24 hours) (Vieira et al. 2012).

The half-life of copper in several tissues was calculated by Levenson and Janghorbani (1994). The study sought to understand the processes by which copper was excreted from several tissues. By restricting

copper in the diet of rats, the authors were able to model the competing processes by which the body tends to excrete copper, while concurrently attempting to retain copper for use in other metabolic processes. These were represented as components, where the first component had a relatively rapid half-life generally unaffected by copper dietary restrictions while the second component half-life was increased substantially by a copper restricted diet. The individual half-life component balance for each organ could not be calculated; however, they could be calculated for some organs. The half-lives for each tissue are presented as the component 1 then component 2 half-lives. The respective calculated copper half-lives were 3.9 and 21 days for the liver, 5.4 and 35 days for the kidney, and 23 and 662 days for the heart; copper turnover in the brain appeared to be monophasic with a half-life of 457 days.

No studies were located regarding the rate and extent of excretion of copper following dermal exposure of humans or animals to copper.

Figure 3-1. Overview of Copper Metabolism in Humans



Source: van de Berghe and Klomp (2009)

3.1.5 Physiologically Based Pharmacokinetic (PBPK)/Pharmacodynamic (PD) Models

PBPK models use mathematical descriptions of the uptake and disposition of chemical substances to quantitatively describe the relationships among critical biological processes (Krishnan et al. 1994). PBPK models are also called biologically based tissue dosimetry models. PBPK models are increasingly used in risk assessments, primarily to predict the concentration of potentially toxic moieties of a chemical that will be delivered to any given target tissue following various combinations of route, dose level, and test species (Clewell and Andersen 1985). Physiologically-based pharmacodynamic (PBPD) models use mathematical descriptions of the dose-response function to quantitatively describe the relationship between target tissue dose and toxic endpoints.

No chemical specific PBPK models have been developed for copper.

3.1.6 Animal-to-Human Extrapolations

NTP (1993) demonstrated that mice appeared less sensitive than rats to the hepatotoxicity of copper based on the observation that no hepatic effects occurred in mice given doses much higher than rats, who showed liver damage at much lower doses. The cause of this apparent difference in toxicity between the species has not been examined. Most of the experimental animal data on the toxicity of copper are from studies in rats.

The dietary requirement for copper in rats is 5–6 mg Cu/kg diet, commonly used in laboratory diets (NRC 1995). It is unlikely that humans would tolerate prolonged exposure to a copper dose that is about 6 times higher than the dietary requirement (0.9 mg Cu/day). Thus, the applicability of these animal data to humans is not known.

The Long-Evans Cinnamon rat is often used as a model for Wilson's disease. This rat strain shares many characteristics associated with Wilson's disease: accumulation of copper in the liver, decreased serum copper and ceruloplasmin levels, and impaired biliary excretion of copper (Sugawara et al. 1991, 1992, 1994; Suzuki et al. 1995).

3.2 CHILDREN AND OTHER POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

This section discusses potential health effects from exposures during the period from conception to maturity at 18 years of age in humans. Potential effects on offspring resulting from exposures of parental germ cells are considered, as well as any indirect effects on the fetus and neonate resulting from maternal exposure during gestation and lactation. Children may be more or less susceptible than adults to health effects from exposure to hazardous substances, and the relationship may change with developmental age.

This section also discusses unusually susceptible populations. A susceptible population may exhibit different or enhanced responses to certain chemicals than most persons exposed to the same level of these chemicals in the environment. Factors involved with increased susceptibility may include genetic makeup, age, health and nutritional status, and exposure to other toxic substances (e.g., cigarette smoke). These parameters can reduce detoxification or excretion or compromise organ function.

Populations at greater exposure risk to unusually high exposure levels to copper are discussed in Section 5.7, Populations with Potentially High Exposures.

Copper is an essential element required for normal growth and development and for a variety of metabolic functions including iron metabolism, cross-linking of connective tissue, and lipid metabolism. Copper deficiency in infants and children is rare, but signs include anemia that is unresponsive to iron supplementation, neutropenia, bone abnormalities, and hypopigmentation of the hair (Araya et al. 2003d; Cordano 1998; Danks 1988). Exposure to excess levels of copper is associated with adverse health effects in infants and children (Araya et al. 2003d; Eife et al. 2009). Wilson's disease, an autosomal recessive disorder which causes liver dysfunction, typically has a childhood onset. Affected individuals can develop toxic tissue accumulations of copper, even with low levels of dietary exposure (Taylor et al. 2020). They require lifelong medical treatment combined with a low-copper diet. Without medical treatment, Wilson's disease is fatal, usually early in life.

Another copper-related genetic disorder, idiopathic copper toxicosis (ICT), is largely believed to be caused by an autosomal recessive genetic susceptibility causing excess copper accumulation and subsequent liver damage; however, it is unclear whether exposure to excess copper plays a role in disease manifestation or if it merely exacerbates symptoms (Müller et al. 1998; Nayak and Chitale 2013). Another disorder, Indian childhood cirrhosis (ICC), is characterized by severe liver damage in infants and children (<5 years of age). It is suspected to be caused by a genetic predisposition due to its random occurrence in siblings and higher liver disease mortality in second-line family members (Nayak and Chitale 2013; Pandit and Bhave 1996). However, data is inconclusive on whether ICC is caused by external exposure to copper, such as the consumption of milk stored in copper or brass vessels, or endogenously through copper dysregulation in the body (Nayak and Chitale 2013; Tanner 1998; Taylor et al. 2020). ICC was previously considered endemic to India, but it has been documented in children of non-Indian origin in other countries (Nayak and Chitale 2013). ICT and ICC lead to a loss of copper homeostasis in diagnosed children and may make them more susceptible to excess copper accumulation especially early in life. In early stages of post-natal development, the mechanisms of copper intestinal

absorption and excretion through bile are not fully developed causing children to be susceptible to even small excesses of copper in water (Puchkova et al. 2018).

Gastrointestinal upset, the most commonly reported adverse health effect in adults, has also been reported in infants and children. It is manifested as nausea, vomiting, abdominal pain, and/or diarrhea. Symptoms usually occur shortly after ingesting a copper-contaminated beverage or drinking water containing a high level of copper. In most of the reports of gastrointestinal upset in children, no reliable information on copper concentration or dose was reported (Gill and Bhagat 1999; Karlsson and Noren 1965; Knobeloch et al. 1994; Spitalny et al. 1984; Walsh et al. 1977). In one report where school-age children ingested a beverage stored in an old urn, the concentration of copper in the beverage was estimated to be 300 mg/L (Gill and Bhagat 1999). Another study reported vomiting in infants ingesting a single dose of 7.5 mg/L copper sulfate (Karlsson and Noren 1965). Knobeloch et al. (1994) noted that children appear to be more sensitive to the gastrointestinal effects of copper than adults. This statement was based on two surveys of residents with elevated copper levels in the drinking water. In the first survey, it appears that children who were categorized as having gastrointestinal upsets were described as “unusually irritable” or had recurrent headaches. In a second survey, mothers were asked to recall the frequency of gastrointestinal effects for all family members (Knobeloch et al. 1994). A significantly higher percentage of children, as compared to adults, were reported to have gastrointestinal effects. Recall bias can be affected by self-reporting or adult reporting of symptoms in children in the household. The available data are inadequate to assess accurately whether there is an age-related difference in the gastrointestinal toxicity of copper.

Copper accumulation in fetal tissues primarily occurs in the second half of pregnancy (Chernenkov et al. 2018). Approximately half of the copper in the fetus is stored in the liver, mostly bound to metallothionein. During that phase of a pregnancy, the rate of transfer of copper from the liver to the bile or blood is decreased due to the immaturity of the fetal liver. The magnitude of the amount of copper in the fetal liver is similar to levels observed in Wilson’s disease; however, the fetal and neonatal liver can tolerate these high concentrations (Olivares et al. 2000). Copper levels are imbalanced in early stages of post-natal development for all infants, as the mechanisms for excreting copper through bile and controlling copper absorption in the small intestine have not developed fully (Puchkova et al. 2018). After birth, copper levels decrease to normal levels in infants lacking a genetic defect (Olivares et al. 2000; Chernenkov et al. 2018). A cross-sectional study (see Table 2-7) in mother-infant pairs measured serum copper concentrations in the umbilical cord blood collected at birth, and no association between copper in cord serum and low infant birth weight was observed (Yang et al. 2020).

Copper, likely bound to albumin, is found in human breast milk and is necessary for infant development.

Copper was measured in breast milk at concentrations of 0.12-0.69 mg/L (Choi et al. 2016; Domellof et al. 2004; Khaghani et al. 2010; Yalcin et al. 2015). Maternal dietary copper intake is not likely to affect copper concentrations in breast milk (Choi et al. 2016); thus, excess dietary maternal copper intake may not impact infant copper intake from breast milk. A study in lactating rats suggests that transport of copper to the mammary gland is about 60% following intraperitoneal or intravenous injection of ionic copper (Donley et al. 2002). Subsequently, the labeled isotopes rapidly appeared in milk and milk ceruloplasmin. These results were not found in non-pregnant rats, where transport was primarily to the liver and kidney.

The potential age-related differences in the toxicity of copper have been assessed in rats exposed to 120 mg Cu/kg/day as copper sulfate in the diet for 12 weeks (Fuentealba et al. 2000). The observed liver effects of enzyme activity alterations and hepatitis were more severe in young rats (exposed *in utero*, during lactation, and for 12 weeks post weaning) as compared to the effects observed in adult rats. Copper levels in the liver of young rats, 1,553–1,635 μg/g, were higher than in adult rats, 472–534 μg/g. It is uncertain if these data in rats would be suggestive of sensitivity in human infants and children.

Several studies investigated the potential developmental toxicity of excess dietary copper sulfate and copper hydroxide; some results suggest that *in utero* exposure to copper can result in delays in growth and development in the offspring of rats (Haddad et al. 1991), mice (Lecyk 1980), and rabbits (Munley 2003a, 2003b). However, some studies testing similar or lower doses in mice and mike observed no developmental effects in offspring (Aulerich et al. 1982; Kadammattil et al. 2018).

A number of populations were identified as unusually susceptible to copper toxicity. The increased susceptibility to copper toxicity is associated with genetic defects that impair copper homeostatic mechanisms. Wilson's disease, also referred to as hepatolenticular degeneration, is an autosomal recessive disorder with an estimated prevalence of 1 case per 30,000 live births among most populations (Schilsky 2019). The primary genetic defect in Wilson's disease is in the ATP7B gene that encodes a P-type ATPase (Wilson protein), that delivers copper to ceruloplasmin. The genetic defect results in impaired biliary excretion of copper and an accumulation of copper in the liver. The progression of the disorder begins with an accumulation of copper in the liver, structural damage to the liver, and subclinical liver cirrhosis (Rodriguez-Castro et al. 2015). Over time, the individual will develop hepatic, neurological, and psychiatric symptoms. The hepatic effects are characterized by jaundice, hypoalbuminemia, ascites, coagulation defects, hyperammonemia, hepatic encephalopathy, and/or liver failure. In the cases with massive liver failure, large amounts of copper are released from the liver, impacting red blood cells and leading to hemolytic anemia. Neurological symptoms include tremors,

other movement disorders, and speech abnormalities. Psychiatric and behavioral symptoms are often found in individuals who also manifest neurological symptoms. The psychiatric symptoms include reduced performance in school or work, inability to cope, depression, very labile moods ranging from mania to depression, sexual exhibitionism, and frank psychosis. Individuals with Wilson's disease have low serum ceruloplasmin levels, elevated urinary copper levels, and elevated liver copper levels. Kayser-Fleischer rings, which result from corneal copper deposits, are also present in some individuals with Wilson's disease. Individuals who are heterozygotes for Wilson's disease may also be more susceptible to the toxicity of copper. Increases in urinary copper and hepatic concentrations and decreased copper incorporation into ceruloplasmin have been observed in heterozygotes. These findings suggest that long-term exposure to elevated levels of copper could result in copper overload.

Individuals with a common deficiency of the enzyme glucose-6-phosphate dehydrogenase (G6PD) could be more susceptible to the toxic effects of oxidative stressors such as copper (Calabrese and Moore 1979; Chugh and Sakhuja 1979; Sansinanea et al. 1996). Red blood cell models were used to analyze the effects of copper chloride on oxidative markers while measuring G6PD activity (Swastika et al. 2020). There was a negative correlation between G6PD activity and copper chloride dose. In the blood, most of the copper is bound to ceruloplasmin. With the exception of ingestion of a very large dose of a copper salt, the levels of non-ceruloplasmin-bound copper remain low. Thus, it is unlikely that this relatively small change in free copper would alter the survival of glucose-6-phosphate dehydrogenase deficient red blood cells.

3.3 BIOMARKERS OF EXPOSURE AND EFFECT

Biomarkers are broadly defined as indicators signaling events in biologic systems or samples. They have been classified as biomarkers of exposure, biomarkers of effect, and biomarkers of susceptibility (NAS/NRC 1989).

The National Report on Human Exposure to Environmental Chemicals provides an ongoing assessment of the exposure of a generalizable sample of the U.S. population to environmental chemicals using biomonitoring (see http://www.cdc.gov/exposurereport/). If available, biomonitoring data for copper and copper compounds from this report are discussed in Section 5.6, General Population Exposure.

A biomarker of exposure is a xenobiotic substance or its metabolite(s) or the product of an interaction between a xenobiotic agent and some target molecule(s) or cell(s) that is measured within a compartment of an organism (NAS/NRC 1989). The preferred biomarkers of exposure are generally the substance itself, substance-specific metabolites in readily obtainable body fluid(s), or excreta. Biomarkers of exposure to copper are discussed in Section 3.3.1.

Biomarkers of effect are defined as any measurable biochemical, physiologic, or other alteration within an organism that (depending on magnitude) can be recognized as an established or potential health impairment or disease (NAS/NRC 1989). This definition encompasses biochemical or cellular signals of tissue dysfunction (e.g., increased liver enzyme activity or pathologic changes in female genital epithelial cells), as well as physiologic signs of dysfunction such as increased blood pressure or decreased lung capacity. Note that these markers are not often substance specific. They also may not be directly adverse, but can indicate potential health impairment (e.g., DNA adducts). Biomarkers of effect caused by copper are discussed in Section 3.3.2.

A biomarker of susceptibility is an indicator of an inherent or acquired limitation of an organism's ability to respond to the challenge of exposure to a specific xenobiotic substance. It can be an intrinsic genetic or other characteristic or a preexisting disease that results in an increase in absorbed dose, a decrease in the biologically effective dose, or a target tissue response. If biomarkers of susceptibility exist, they are discussed in 3.2, Children and Other Populations that are Unusually Susceptible.

3.3.1 Biomarkers of Exposure

Copper levels can be readily measured in tissues, body fluids, and excreta. Depending on the dose and exposure duration, inhalation and/or oral exposure to copper can result in increased levels of copper in serum, urine, hair, and nails.

The normal serum copper level in human adults is 11–22 μmol/L (70-140 μg/dL) (LaGow et al. 2007). It can be used to evaluate copper toxicity, deficiency, or the possibility of copper metabolism disorders. Increased serum copper levels (>22 μmol/L) were reported in several human case studies following intentional ingestion of copper compounds, such as copper sulfate biocides (Chuttani et al. 1965; Du and Mou 2019; Franchitto et al. 2018; Higny et al. 2014; Yang et al. 2004). Serum copper levels reported in these studies ranged from 22.7 to 140 μmol/L. Elevated plasma copper was also measured in a 23-month-old child who had accidentally ingested an unknown amount of a disinfectant agent containing an unknown concentration of copper sulfate (Mortazavi and Jafari-Javid 2009). Fifteen days after admission, the patient’s plasma copper level was 216 μg/dL (33.9 μmol/L) (normal range in 6-month to 6-year-old children: 14-30 μg/dL). Whole blood copper levels measured in humans following intentional ingestion of copper sulfate ranged from 60.3 to 107.6 μmol/L, while in non-exposed individuals the whole blood copper was 34.1 μmol/L (Chuttani et al. 1965). Following chronic inhalation exposure to 111-464 mg Cu/m^3 copper in dust, serum copper levels greater than 31.8 μmol/L were observed in 16% of exposed factory workers (Suciu et al. 1981). However, increased serum copper levels may only be reflective of recent exposure. Chuttani et al. (1965) observed that serum ionic copper rapidly diminished within a few

days to normal levels following ingestion of an acute bolus dose. Mortazavi and Jafari-Javid (2009) observed that in a 23-month-old child, copper levels took about 2 months to fall to within normal range, even after treatment with a chelating agent. A relationship between blood copper levels and the severity of symptoms has not been established. Among individuals intentionally ingesting a single dose of copper sulfate (1–30 g) there did not appear to be a correlation between serum copper levels and symptom severity (Chuttani et al. 1965). In contrast, whole blood copper levels did have a significant relationship with the severity of symptoms.

Serum ceruloplasmin, a copper-related carrier protein is a biomarker for copper exposure. Based on a significant correlation of serum copper with serum ceruloplasmin levels, it has been suggested that serum ceruloplasmin is a reliable biomarker for chronic occupational exposure to copper (Saha et al. 2008). A human dietary study by Turnlund et al. (2004) reported that a high copper intake resulted in an increase of ceruloplasmin in subjects given supplements, when compared to controls. Nine men had been exposed to 0.02 and 0.1 mg Cu/kg/day during separate 18-day period in a metabolic research unit (Turnlund et al. 2004). A metabolism study in rats observed increases in ceruloplasmin with copper exposure, as over 90% of the copper dose was found primarily in ceruloplasmin as opposed to other serum proteins (Weiss and Linder 1985).

Several epidemiological studies in human suggest associations between increased serum copper levels and various health effects; however, it is not known if increases in serum copper are a result of copper exposure. Age-adjusted serum copper was 5% higher in subjects who died from cardiovascular disease compared to those who did not, and odds of mortality from cardiovascular disease increased by 6% per 1 μmol/L increase in serum copper (Ford 2000). A cross-sectional study in adolescents found a negative association between increased blood copper levels and production of sex hormones, odds of male genital development, and age menarche (De Craemer et al. 2017). A separate cross-sectional study found an association between blood copper levels in the umbilical cord and decreased birthweight (Yang et al. 2020).

Similar to serum, copper can be measured in urine, but this is primarily used to test for diseases affecting copper homeostasis and the liver. According to the Mayo Clinic, a normal 24-hour urine copper level is between 9-71 μg/24 hours (Mayo Clinic 2020). In one patient who intentionally ingested a copper sulfate containing fungicide, the urine copper level three days after admission was 112 μg/dL and decreased to 16 μg/dL in follow-up 11 days after admission (Yang et al. 2004).

Copper levels in hair and nails can also be used to assess copper exposure; however, the reliability of these biomarkers has not been established. In a study of preschool children, the levels of copper in hair and toenail samples were log-normally distributed (Wilhelm et al. 1991). The geometric mean concentrations of copper in hair and toenails were 10.6 µg/g (range of 5.4–20.7 µg/g) and 7.5 µg/g (range of 3.0–18.6 µg/g), respectively. A study by Hopps (1977) calculated that for a hair growth rate of 10 mm per month, the copper levels in the first 2 cm proximal to the scalp would represent copper intake over 2 months. In an occupational study of workers exposed to unspecified levels of copper from fossil fuel combustion, oil distribution workers had a mean hair copper level of 69.6 ug/g, which was significantly higher than in controls (defined in the study as non-exposed "healthy individuals living far from hazardous exposure with age and weight matching the test group") who had a mean hair copper level of 36.8 ug/g (Jaccob 2020). The study author suggested that hair levels may be a useful biomarker for copper and heavy metal exposure. Increased hair copper levels have been reported in workers exposed to 0.64–1.05 mg/m^3 of an unspecified copper compound; the concentration of copper in their hair was 705.7 µg/g, as compared to a concentration of 8.9 µg/g in non-exposed workers (Finelli et al. 1981).

Based on a toenail growth rate of 1 mm/month, toenail samples would represent copper intakes over 12–18 months (Fleckman 1985). Increased hair and fingernail copper levels were observed in children with ICC (Sharda and Bhandari 1984). An epidemiological study found that mean toenail copper concentrations were significantly higher among residents who lived in copper-mining towns than those who did not (Ndilila et al. 2014). Among adults in the copper-mining town, the mean copper concentration in toenails was 132 mg/kg. Study authors suggest that copper levels in toenails may be an indicator of exposure.

3.3.2 Biomarkers of Effect

No copper-specific biomarkers of effects resulting from copper toxicity have yet been identified. The most notable sign of toxicity in humans ingesting a beverage or water containing copper is gastrointestinal distress. Symptoms (typically nausea, vomiting, and abdominal pain) usually occur shortly after ingesting the contaminated beverage. The liver is another sensitive target of copper toxicity. Alterations in several serum enzymes, including serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), and ALP, were reported in a number of human case studies of copper-induced liver damage (Chuttani et al. 1965; Donoso et al. 2007; Gamakaranage et al. 2011; Hassan et al. 2010; Lubica et al. 2017; Malik and Mansur 2011; Sinkovic et al. 2008). Increases in serum bilirubin levels have also been observed in humans. Animal studies demonstrate that the rise in serum enzyme activities

is evidence of liver damage (see Section 2.9). However, alterations in serum enzyme levels are not unique to copper-induced liver damage.

3.4 INTERACTIONS WITH OTHER CHEMICALS

Numerous studies demonstrate the interaction between copper and metals such as cadmium, iron, and tin. Dietary zinc strongly affects copper absorption, and a diet high in zinc can result in copper deficiency (Igic et al. 2002; Myint et al. 2018). Uptake of copper from the small intestine is susceptible to competition from other transition metals including zinc. Increased dietary zinc results in induction of metallothionein synthesis in the intestine. Since metallothionein has a greater binding capacity for copper than for zinc, dietary copper is sequestered in the intestinal mucosal cell metallothionein and is eventually excreted in the feces when the mucosal cell is sloughed off (Hall et al. 1979; Whanger and Weswig 1971). Because exposure to excess dietary zinc results in both decreased copper absorption and decreased serum levels, it is considered an effective therapy for Wilson's disease (Ranucci et al. 2014).

Animal studies demonstrate that ingestion of copper and zinc ions simultaneously results in reduction of systemic copper toxicity because it decreases systemic uptake (Babaei and Abshenas 2013; Kheirandish et al. 2014). Mice given both zinc sulfate and copper sulfate had less histological damage in the testis compared to mice give copper sulfate only (Kheirandish et al. 2014). Similar results were observed in rats, as improvements in sperm counts, viability, and motility were observed in rats given copper sulfate and zinc sulfate, while no such recovery was seen over the same time period of rats only given copper sulfate (Babaei and Abshenas 2013).

A study in rats found that exposure to sodium arsenate resulted in increased copper concentration in the kidney (Cui and Okayasu 2008). Rats were orally exposed to varying doses of sodium arsenate daily for 4 and 16 weeks. Exposure to manganese in rats resulted also increased copper as demonstrated by a 7-day exposure to manganese in diet, water, or gavage resulted in increased copper levels in the liver (Mercadante et al. 2016). Exposure to manganese by diet and gavage resulted in decreased copper levels in bile; both effects suggest a relationship between manganese and copper hepatobiliary excretion. Several other divalent cations compete with copper for intestinal absorption. Exposure to dietary cadmium (Evans et al. 1970), iron (Ha et al. 2016), and stannous tin (Pekelharing et al. 1994; Wapnir et al. 1993) can result in decreased copper absorption. In the case of cadmium, the copper ion decrease is related to cadmium's induction of metallothionein synthesis and the binding of copper to it. Tetrathiomolybdate is used for the treatment of Wilson's disease (Brewer et al. 2006), thus excessive dietary molybdenum can also result in decreased copper uptakes and, therefore, alterations in copper utilization and toxicity. Two mechanisms of action of tetrathiomolybdate have been proposed: (1) it reacts

with copper-metallothionein to form a soluble complex that is excreted (Ogra et al. 1996), and (2) it can complex with non-ceruloplasmin-bound plasma copper, impeding its cellular absorption (Brewer et al. 2006). Interactions with copper sulfate may differ, as molybdenum may lower the activity of sulfide oxidase, resulting in the accumulation of copper sulfide (Vyskocil and Viau 1999)

Vitamin C, also known as ascorbic acid, interferes with intestinal copper absorption resulting in reduced copper concentration in various tissue (Van Den Berg and Beynen 2007). This suggests that a diet high in vitamin C can result in copper deficiency.

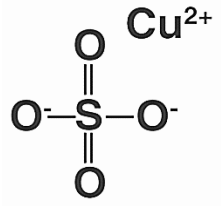
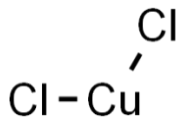
Several other natural substances have been tested in animals, and studies suggest that they may protect against copper toxicity. In mice, copper-induced toxicity changes in the liver, kidneys, and stomach were less pronounced in mice treated with copper sulfate and coriander, or copper sulfate, coriander, and zinc, compared to mice treated only with copper sulfate (Hashimyousif et al. 2019). Curcumin, the main active ingredient in turmeric and a natural inflammatory agent, appeared to alleviate the hepatic and renal toxicity of copper sulfate (Hashish and Elgaml 2016). This was based on a comparison of hepatic enzyme levels, and liver and kidney antioxidant levels, between rats orally exposed to copper sulfate only and rats exposed to copper sulfate and curcumin at the same time or in succession. Resveratrol, an antioxidative compound, was observed to possibly attenuate copper sulfate-induced liver injury by decreasing oxidative stress and the concentrations of liver transaminases (Tian et al. 2019). An *in vivo* genotoxicity study using mouse blood cells reported that orange juice appeared to have a modulating effect on the action of metallic sulfate salts and was both restorative and protective of the copper-induced genotoxic effects (Franke et al. 2006). Study authors hypothesized that the genotoxic effects could be mediated by the interaction of unspecified orange juice components or that the juice's antioxidant byproducts can interact with transition metals.

CHAPTER 4. CHEMICAL AND PHYSICAL INFORMATION

4.1 CHEMICAL IDENTITY

Copper is a transition metal and Group 11 essential element on the periodic table, atomic number 29, that can occur naturally in elemental form. Copper displays four oxidation states: Cu(0), Cu(I), Cu(II), and Cu(III). Its industrial uses include electrical products and equipment, wiring, piping, sheet metal, building material, machinery, and motors. Copper is essential to human health and is found in many foods. Copper sulfate ($CuSO_4$) is an inorganic compound that can occur in nature. It is the most common compound used in commercial applications. It is the most widely used copper salt and is an ingredient in pesticide formulations. Copper chloride is another important copper salt. It is used as a catalyst in chemical reactions and in dyeing, printing, and fungicides (Budavari et al. 1996). Copper nanoparticles are formed through natural processes or can be manmade. They are primarily used as antimicrobial, antibacterial and antifungal agents. A summary of copper nanoparticle toxicity is in Section 2.21. Table 4-1 lists common synonyms, trade names, and other pertinent identification information for copper, copper sulfate, and copper chloride.

Table 4-1. Chemical Identity of Copper, Copper Sulfate and Copper Chloride

Characteristic	Copper	Copper Sulfate	Copper Chloride
Chemical Name	Copper	Copper Sulfate	Copper Chloride
Synonym(s) and Registered trade name(s)	M1; M2; M3; M4; Cuprum; Gold Bronze; 1721 Gold; Bronze powder; Cobre; Cuivre; Rame; Allbri Natural Copper; M3R; M3S; E 115; OFHC CU	Cupric Sulfate; Copper (II) sulfate; cupric sulfate anhydrous; copper sulphate; Blue stone; copper monosulfate; Hylinec; Trinagle; Delcup, cupric sulphate; sulfuric acid copper (2+) salt (1:1); monocopper sulfate	copper(II) chloride; cupric chloride; cupric chloride anhydrous; cupric chloride dihydrate
Chemical formula	Cu	$CuSO_4$	$CuCl_2$
Chemical structure	Cu		
CAS registry number	7440-50-8	7758-98-7	7447-39-4

Source: PubChem 2020

4.2 PHYSICAL AND CHEMICAL PROPERTIES

Copper is a metallic solid that is malleable and has high thermal conductivity, high electrical conductivity, low corrosivity, and alloying ability. Its malleability is attributed to its relatively low number of electrons on its outer shell. The properties of copper typically vary with purity. Metallic copper is naturally a reddish color, and when exposed to oxygen in the air, it forms copper oxide which is black (Haynes 2015). As copper reacts with carbon dioxide in the air, copper carbonates, which are usually green, form. Copper is positioned below hydrogen in the electromotive-force series (lower reactivity); therefore, it will not displace hydrogen ions in water, and thus has no single displacement interaction with water. It is soluble in dilute acid and in ammonia with the presence of an oxidizing agent. Copper will undergo galvanic corrosion when in contact with other metals. Copper sulfate is typically produced by treating hot copper with sulfuric acid. The resulting material is a white-green solid when anhydrous and blue crystals when hydrated ($CuSO_4.5H_2O$), and it readily absorbs water (Haynes 2015). Copper chloride is produced by reaction of metallic copper with chlorine. It is a yellow-brown powder in the anhydrous form**.** Table 4-2 lists important physical and chemical properties of copper, copper sulfate, and copper chloride.

Table 4-2. Physical and Chemical Properties of Metallic Copper, Copper Sulfate, and Copper Chloride

Property	Copper	Copper (II) Sulfate	Copper (II) Chloride
Molecular weight	63.55 g/mol	159.61 g/mol	134.45 g/mol
Color	Reddish, lustrous	White, off-white when dehydrated; blue crystals when hydrated	Yellow to brown
Physical state	Solid	Solid	Solid
Melting point	1083°C (1981°F)	590°C	630°C
Boiling point	2595°C (4703°F)	650°C	993°C
Density: At 20°C/4°C	8.94	3.6	3.39
Odor	Odorless	Pleasant Odor	Odorless
Odor threshold: Water Air	 No data No data	 No data No data	 No data No data
Taste threshold	No data	No data	No data
Solubility: Water Organic solvent(s)	 Insoluble Slightly soluble in dilute acid and ammonia water	 Soluble Soluble in methanol Insoluble in ethanol	 Soluble in acetone, ethanol
Partition coefficients:			

Table 4-2. Physical and Chemical Properties of Metallic Copper, Copper Sulfate, and Copper Chloride

Property	Copper	Copper (II) Sulfate	Copper (II) Chloride
Log K_{OW} Log K_{OC}	No data No data	No data No data	No data No data
Vapor pressure At 20°C	1 mm Hg at 1628°C	No data	No data
Henry's law constant	No data	No data	No data
Autoignition temperature	No data	No data	No data
Flashpoint	No data	No data	No data
Flammability limits	No data	No data	No data
Conversion factors	Since these substances exist in the atmosphere in the particulate state, the concentration is expressed as mg/m^3.		
Explosive limits	No data	No data	No data

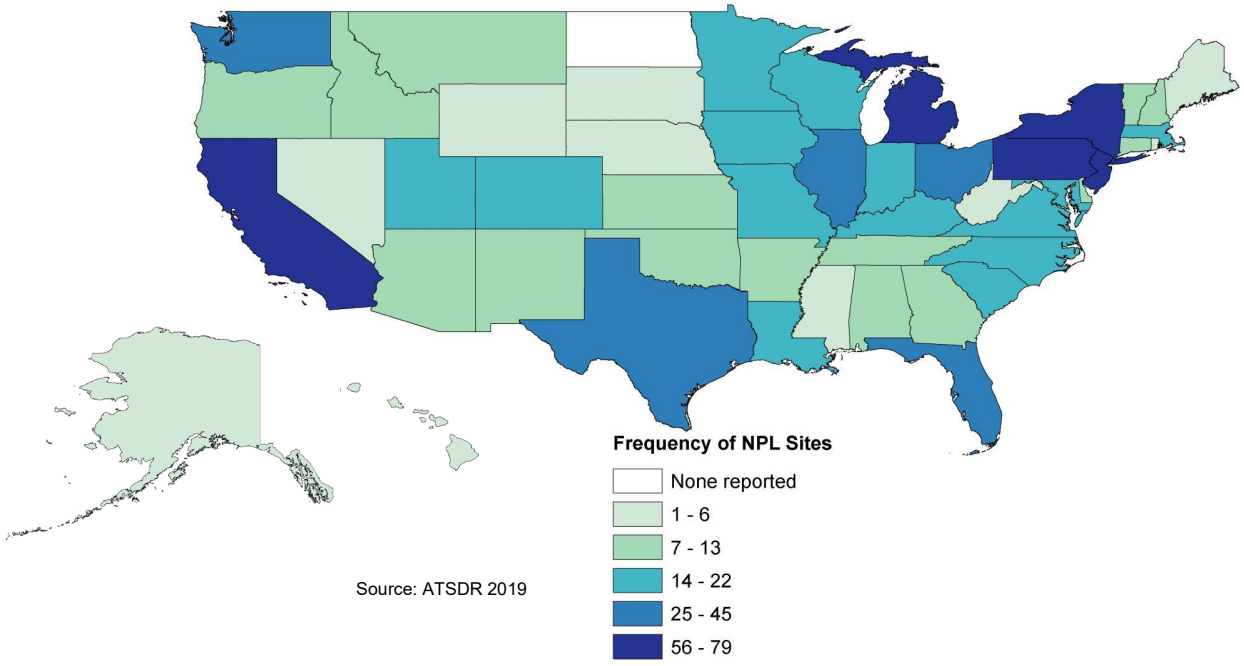
Source: Haynes 2015; PubChem 2020

CHAPTER 5. POTENTIAL FOR HUMAN EXPOSURE

5.1 OVERVIEW

Copper and copper compounds have been identified in at least 929 of the 1,867 hazardous waste sites that have been proposed for inclusion on the EPA National Priorities List (NPL) (ATSDR 2019). However, the number of sites evaluated for copper and copper compounds is not known. The number of sites in each state is shown in Figure 5-1. Of these sites, 920 are located within the United States, 1 is located in Guam, 1 is located in the Virgin Islands, and 7 are located in Puerto Rico (not shown). Of the sites in the United States, 2 sites did not report a location and are not reflected in Figure 5-1.

Figure 5-1. Number of NPL Sites with Copper and Copper Compound Contamination



- Copper occurs naturally both in many minerals and in the metallic state. The top 10 applications for copper in the United States, in order of percentage of total use, are building wire, plumbing, and heating, automotive, air conditioning, refrigeration and natural gas, power utilities, telecommunications, in-plant equipment, ordnance, business electronics, and lighting and wiring devices.
- Industrial effluents, mining and production of copper and other metals, municipal solid waste management, and fossil fuel combustion account for a large portion of the total environmental

releases of copper and copper compounds. Natural sources of copper releases include windblown dust, volcanoes, decaying vegetation, forest fires, and sea spray.

- Copper is an essential micronutrient present in many foods. Copper gluconate and copper sulfate are direct food additives generally recognized as safe by the FDA.
- The general population is expected to be exposed to copper daily via inhalation and ingestion of foods, and dermally to a lesser extent.
- People living near copper smelters and refineries and workers in these and other industries may be exposed to high levels of dust-borne copper by both inhalation and ingestion.

Copper and its compounds are naturally present in the earth's crust and can be discharged naturally to air and water during weathering. Mean copper concentrations in the atmosphere measured at multiple U.S. locations ranged between 0.013 to 0.0792 µg/m^3 from 2016 to 2019 (EPA 2020a). Airborne copper is associated with particulates that are derived from suspended soils, combustion sources, the manufacture or processing of copper-containing materials, or mine tailings. Copper associated with particulate matter is emitted into the air naturally from windblown dust, volcanoes, and anthropogenic sources, the largest of which are primary copper smelters and ore processing facilities. The major sources of releases to water are mining operations, agriculture, sludge from publicly owned treatment works (POTWs), and municipal and industrial solid waste. Mining and milling contribute the most waste. Copper is released to water as a result of natural weathering of soil and discharges from industries and sewage treatment plants. Copper compounds may also be intentionally applied to water to kill algae. Copper concentrations in groundwater vary widely from 0.2 to 98.4 µg/L (USGS 2020b). Copper is predominantly found in the Cu(II) state. Most of it is complexed or tightly bound to organic matter. Little is present in the free (hydrated) or readily exchangeable form. The combined processes of complexation, adsorption, and precipitation control the level of free Cu(II). The chemical conditions in most natural water is such that, even at relatively high copper concentrations, these processes will reduce the free Cu(II) concentration to extremely low values. The USGS reports the median level of copper in soil and sediment as 30 ppm (USGS 2016). Copper concentrations will be higher in soils that are close to sources of copper emissions.

In the general population, the highest exposures to copper come from drinking water and food. Copper can leach into drinking water from contact surfaces within the water distribution systems, the water treatment plant, and the in-home plumbing system. When a system has not been flushed after a period of disuse, the concentration of copper in tap water can exceed 1.3 mg/L, the EPA drinking water Action Level.

Many workers are exposed to copper in agriculture, industries connected with copper production, metal plating, and other industries. Based on the available data, people living close to NPL sites contaminated with copper may be at greater risk for exposure to copper than the general population with respect to inhalation of airborne particulates from the NPL sites, ingestion of contaminated water or soil, and/or uptake of copper by fruits and vegetables raised in gardens of residents living near NPL sites. People living near copper smelters and refineries and workers in these and other industries may be exposed to high levels of dust-borne copper by both inhalation and ingestion routes.

5.2 PRODUCTION, IMPORT/EXPORT, USE, AND DISPOSAL

5.2.1 Production

Copper occurs naturally in many minerals, such as cuprite (Cu_2O), tenorite (CuO), malachite ($CuCO_3 \cdot Cu(OH)_2$), azurite ($2CuCO_3 \cdot Cu(OH)_2$), antlerite ($CuSO_4 \cdot 2Cu(OH)_2$), brochantite ($CuSO_4 \cdot 3Cu(OH)_2$), chrysocolla ($CuO \cdot SiO_2 \cdot 2H_2O$), chalcopyrite ($CuFeS_2$), chalcocite ($Cu_2S$), covellite (CuS), and bornite (Cu_5FeS_4). It also occurs uncombined as solely copper metal (Davenport 2001). Copper is most commonly present as copper-iron-sulfide and copper sulfide minerals (Schlesinger et al. 2011a). The copper content of ores ranges from 0.5 to 1 or 2% copper (Schlesinger et al. 2011a). Most copper is obtained from Cu-Fe-S ores, such as chalcopyrite and chalcocite, and the principal copper ore mineral is chalcopyrite, which yields a matte of approximately 50% copper (Morris and Wadsley 2001; Schlesinger et al. 2011a).

Mine production of recoverable copper in the United States totaled 1.3 million tons in 2019 (USGS 2020a). In 2015, the recoverable copper content per unit of ore mined was 0.47% (USGS 2017b). The United States is the world's fourth leading copper producer, along with Congo and following Chile, China, and Peru (USGS 2020a). In 2019, copper was actively mined in seven states with Arizona accounting for 68% of U.S. copper production, followed by Utah, New Mexico, Nevada, Montana, Michigan, and Missouri (USGS 2020a). There were 24 copper-producing U.S. mines in 2019, and 15 mines accounted for 99% of production in the United States. Production, processing and use of copper and copper compounds in the United States reported to the EPA’s Toxics Release Inventory (TRI), listed by state, are displayed in Table 5-1 and Table 5-2, respectively.

Table 5-1. Facilities that Produce, Process, or Use Copper

State[a]	Number of facilities	Minimum amount on-site in pounds[b]	Maximum amount on-site in pounds[b]	Activities and uses[c]
AK	4	40,000	399,996	1, 10, 12, 13, 14
AL	61	808,228,700	2,282,286,941	1, 2, 3, 5, 7, 8, 9, 10, 11, 12, 13, 14
AR	48	25,378,100	153,781,055	1, 2, 3, 5, 6, 7, 8, 9, 10, 11, 12, 14
AZ	22	2,596,000	25,959,978	1, 3, 5, 6, 7, 8, 9, 11, 12
CA	102	4,811,600	48,116,295	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
CO	11	550,323,000	1,103,229,990	1, 2, 3, 4, 6, 7, 8, 11, 12
CT	40	3,477,000	34,769,961	2, 3, 4, 7, 8, 9, 11, 12, 14
DE	2	1,000	9,999	8
FL	33	50,777,000	107,769,969	1, 2, 3, 5, 6, 7, 8, 9, 10, 11, 12
GA	67	8,364,200	83,642,238	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 14
HI	3	200,000	2,000,097	1, 5, 9, 12
IA	46	4,406,300	44,062,956	1, 2, 3, 5, 7, 8, 9, 11, 12, 14
ID	7	241,000	2,409,993	8, 9, 12
IL	139	44,863,100	298,631,064	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
IN	134	18,664,500	186,644,970	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
KS	36	1,599,000	15,990,066	1, 2, 3, 4, 5, 6, 7, 8, 11, 12, 14
KY	60	7,032,200	70,321,941	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
LA	19	761,200	7,612,083	1, 2, 5, 7, 8, 9, 10, 12, 13, 14
MA	54	14,511,100	95,110,951	1, 3, 6, 7, 8, 9, 11, 12, 14
MD	7	222,000	2,219,994	1, 2, 3, 4, 5, 8, 9, 12
ME	7	700,000	6,999,993	8, 9, 12
MI	103	6,541,500	65,415,105	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
MN	63	102,889,300	528,893,041	1, 2, 3, 4, 5, 7, 8, 9, 11, 12, 14
MO	63	6,661,100	66,610,944	1, 5, 6, 7, 8, 9, 10, 11, 12, 14
MS	28	25,991,000	159,909,974	1, 2, 3, 4, 5, 7, 8, 12, 13, 14
MT	1	1,000	9,999	8
NC	83	62,153,300	221,533,020	1, 2, 3, 4, 5, 6, 7, 8, 9, 11, 12, 14
ND	4	111,000	1,109,997	8
NE	20	2,322,000	23,219,982	2, 3, 7, 8, 9, 11, 12, 14
NH	22	4,836,100	48,360,978	1, 2, 3, 5, 7, 8, 9, 11, 12
NJ	30	13,897,000	88,970,071	1, 2, 3, 4, 5, 7, 8, 9, 12
NM	3	20,000	200,097	8, 12
NV	8	52,000	519,993	1, 5, 8, 9, 11, 12
NY	93	13,982,500	139,824,909	1, 2, 3, 4, 5, 7, 8, 9, 10, 11, 12, 14
OH	198	23,512,400	185,124,010	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
OK	61	126,709,100	367,091,045	2, 3, 7, 8, 9, 11, 12, 14
OR	16	481,100	4,811,184	1, 5, 6, 7, 8, 9, 10, 12, 14
PA	196	46,671,200	316,712,019	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14

Table 5-1. Facilities that Produce, Process, or Use Copper

State[a]	Number of facilities	Minimum amount on-site in pounds[b]	Maximum amount on-site in pounds[b]	Activities and uses[c]
PR	14	452,000	4,520,088	7, 8, 9, 11, 14
RI	15	2,361,000	23,609,988	7, 8, 9, 12, 14
SC	63	10,739,400	107,394,039	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
SD	13	353,200	3,531,987	1, 2, 3, 4, 5, 7, 8, 9, 14
TN	64	13,852,000	138,519,936	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
TX	128	85,804,000	358,039,880	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
UT	11	1,452,000	14,519,988	8, 12
VA	32	2,836,100	28,361,070	1, 4, 7, 8, 9, 11, 12, 14
VT	4	121,000	1,209,996	2, 3, 4, 8, 11, 12
WA	25	417,100	4,171,275	1, 2, 3, 5, 6, 7, 8, 9, 11, 12, 13, 14
WI	155	10,012,300	100,122,849	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
WV	4	3,101,000	31,009,995	2, 3, 7, 8
WY	3	120,000	1,199,997	1, 2, 4, 9, 12, 13

[a]Post office state abbreviation used.
[b]Amounts on site reported by facilities in each state,
[c]Activities/Uses:

1. Produce
2. Import
3. Used Processing
4. Sale/Distribution
5. Byproduct
6. Reactant
7. Formulation Component
8. Article Component
9. Repackaging
10. Chemical Processing Aid
11. Manufacture Aid
12. Ancillary
13. Manufacture Impurity
14. Process Impurity

Source: TRI18 2019; Data are from 2018

Table 5-2. Facilities that Produce, Process, or Use Copper Compounds

State	Number of facilities	Minimum amount on-site in pounds	Maximum amount on-site in pounds	Activities and uses
AK	4	2,110,000	21,099,996	1, 2, 3, 5, 6, 10, 12, 13, 14
AL	65	6,335,200	63,352,062	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
AR	49	2,103,200	21,031,974	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
AZ	28	1,262,064,300	11,020,643,075	1, 2, 3, 4, 5, 6, 8, 9, 10, 11, 12, 13, 14
CA	68	488,300	4,883,148	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 14
CO	17	93,100	930,987	1, 2, 4, 5, 7, 8, 10, 12, 13, 14
CT	17	12,464,000	74,639,983	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12
DC	1	10,000	99,999	12
DE	6	100	999	8
FL	39	50,424,200	104,241,978	1, 3, 4, 5, 6, 7, 8, 9, 10, 12, 13, 14

Table 5-2. Facilities that Produce, Process, or Use Copper Compounds

State	Number of facilities	Minimum amount on-site in pounds	Maximum amount on-site in pounds	Activities and uses
GA	59	11,815,100	68,150,974	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 13, 14
IA	38	10,664,000	56,640,280	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
ID	14	562,000	5,619,987	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
IL	83	5,291,500	52,915,428	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
IN	64	26,688,600	166,886,144	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
KS	21	285,000	2,849,985	1, 3, 4, 5, 6, 7, 8, 9, 10, 12, 13, 14
KY	40	3,136,100	31,360,968	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
LA	36	2,801,200	28,011,969	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
MA	8	230,000	2,299,995	1, 2, 3, 4, 5, 7, 8, 11, 14
MD	10	1,040,100	10,400,994	1, 3, 4, 5, 7, 8, 9, 12, 13
ME	1	1,000	9,999	1, 5, 7
MI	59	15,924,200	109,241,950	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
MN	45	1,406,100	14,060,979	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13
MO	48	15,807,100	108,070,969	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 13, 14
MS	30	642,000	6,419,988	1, 2, 3, 5, 8, 10, 13, 14
MT	11	11,411,100	64,110,991	1, 2, 3, 4, 5, 6, 8, 9, 10, 12, 13, 14
NC	65	15,285,200	102,852,067	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
ND	6	42,000	419,994	1, 5, 9, 12, 13, 14
NE	18	3,142,100	31,420,989	1, 3, 4, 5, 6, 7, 8, 9, 12, 13, 14
NH	5	1,121,000	11,209,995	1, 3, 5, 6, 7, 8, 9, 11, 14
NJ	15	1,552,100	15,520,986	1, 2, 3, 4, 5, 7, 8, 9, 12, 14
NM	8	11,031,000	60,309,994	1, 3, 4, 5, 8, 11, 12, 13, 14
NV	15	11,462,000	64,620,184	1, 2, 3, 4, 5, 7, 8, 10, 11, 12, 13, 14
NY	26	10,211,200	52,112,083	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 13, 14
OH	88	28,510,300	185,103,125	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
OK	31	687,300	6,872,976	1, 2, 3, 4, 5, 7, 8, 9, 10, 11, 12, 13, 14
OR	23	383,100	3,830,985	1, 2, 3, 4, 5, 6, 7, 8, 10, 11, 12, 14
PA	88	56,782,200	167,822,325	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
PR	7	110,000	1,099,998	11, 12
RI	8	412,100	4,120,992	7, 8, 11, 12, 14
SC	36	2656100	26,560,971	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13
SD	8	10,000	99,999	1, 5, 9, 13, 14
TN	51	3,170,200	31,702,257	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
TX	117	165,435,500	704,355,812	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
UT	20	1105373000	10,553,729,981	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
VA	38	1,034,000	10,340,073	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14
VT	1	0	0	
WA	21	2,483,100	24,831,180	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14

Table 5-2. Facilities that Produce, Process, or Use Copper Compounds

State	Number of facilities	Minimum amount on-site in pounds	Maximum amount on-site in pounds	Activities and uses
WI	50	3,446,200	34,461,963	1, 2, 3, 4, 5, 6, 7, 8, 10, 11, 12
WV	20	293,100	2,931,084	1, 3, 4, 5, 7, 8, 9, 11, 12, 13, 14
WY	6	211,2000	21,119,994	1, 3, 4, 5, 12, 13, 14

[a]Post office state abbreviation used.
[b]Amounts on site reported by facilities in each state,
[c]Activities/Uses:

1. Produce
2. Import
3. Used Processing
4. Sale/Distribution
5. Byproduct
6. Reactant
7. Formulation Component
8. Article Component
9. Repackaging
10. Chemical Processing Aid
11. Manufacture Aid
12. Ancillary
13. Manufacture Impurity
14. Process Impurity

Source: TRI18 2019; Data are from 2018

Copper from oxidized minerals is usually produced by leaching, solvent extraction, and electrowinning (Schlesinger et al. 2011b). Since most copper comes from Cu-Fe-S ores that are not easily dissolved by aqueous solutions, most extraction occurs by concentration, smelting, and refining (Schlesinger et al. 2011b). This extraction occurs by crushing and grinding the ore and then isolating mineral particles to a concentrate by froth flotation, smelting the concentrate to a matte, oxidizing the matte to impure molten copper, and then fire- and electrorefining the copper (Schlesinger et al. 2011b).

Production of copper in the United States includes not only the processing of both domestic and foreign ores, but also the recovery of scrap. Scrap is a significant part of the U.S. copper supply. There are three types of scrap: home scrap (copper that primary producers cannot further process or sell), old scrap (metal that has been used in products), and new scrap (generated during manufacturing) (Schlesinger et al. 2011c). In 2015, smelting was performed in the United States by three smelters with a combined production of 527,000 metric tons per year (USGS 2017b). During 2015, three refineries produced 1,090,000 metric tons of copper from primary sources and 48,800 from secondary materials (scrap) for a combined total refinery production in the United States of 1,140,000 tons (USGS 2017b). Production of secondary copper amounted to 805,000 metric tons in 2015 (USGS 2017b). In 2019, three smelters, three electrolytic refineries, four fire refineries, and 14 electrowinning facilities operated in the United States (USGS 2020a). Refineries produced 1,000,000 metric tons from ore and 45,000 metric tons from scrap, for a total refinery production of 1,045,000 metric tons.

Copper sulfate is also produced as a byproduct of copper production by ore-leaching with sulfuric acid as the solvent. Production of copper sulfate in the United States increased from 22,800 metric tons in 2011

to 23,000 metric tons in 2013 but decreased to 18,497 metric tons in 2015 (USGS 2017b). Production figures for other copper compounds are not reported by the USGS.

5.2.2 Import/Export

In 2019, 35,000 metric tons of unmanufactured copper and 650,000 metric tons of refined copper were imported into the United States. (USGS 2020a). Chile, Canada, and Mexico were the principal sources of imported refined copper. Imports of copper sulfate amounted to 43,900 metric tons in 2015 and were primarily obtained from Mexico (USGS 2017b).

In 2019, the United States exported 330,000 metric tons of unmanufactured copper and 140,000 metric tons of refined copper (USGS 2020a). In 2015, copper scrap was the leading U.S. copper export at 426,000 metric tons (USGS 2017b). Exports of copper sulfate amounted to 6,170 metric tons in 2015 (USGS 2017b).

5.2.3 Use

Copper is one of the most important metals used in industries because of its resistance to corrosion, antimicrobial properties, durability, ductility, malleability, and electrical and thermal conductivity. It is used primarily as the metal or in alloys. Its alloys, including brass and bronze, are important commodities (USGS 2009a). Currently American coins are copper alloys (USDT 2018). A small percentage of copper production goes into the manufacture of copper compounds, primarily copper sulfate.

After accounting for production, imports, and exports, 1,800,000 metric tons of copper were available for use in 2019 (USGS 2020a). The Copper Development Association estimates that the end-use distribution of copper and copper alloy products in 2019 were: building construction, 43%; electrical and electrical products, 20%; transportation equipment, 20%; consumer and general products, 10%; and industrial machinery and equipment, 7% (USGS 2020a). The top 10 applications for copper in the United States, in order of percentage of total use, are building wire, plumbing and heating, automotive, air conditioning, refrigeration and natural gas, power utilities, telecommunications, in-plant equipment, ordnance, business electronics, and lighting and wiring devices (Schlesinger et al. 2011b). Copper plumbing is used in water distribution systems (Edwards et al. 2001; EPA 1995; Grace et al. 2012; Knobeloch et al. 1998; Lagos et al. 2001; Rajaratnam et al. 2002; Schock and Sandvig 2009; Turek et al. 2011). Copper and its salts are also used in cookware, kitchen utensils, and mugs; marine antifouling paints; animal feed supplements; fertilizers, fireworks; brake pads; water pipes; roofs; gutters; shingles; wood preservatives; and tires (Banavi et al. 2020; Koo et al. 2020; Lifset et al. 2012; Ni and Li 2008).

The EPA has registered about 300 copper compounds and alloys as antimicrobial agents (Vincent et al. 2016). Copper-silver ionization filters have been used in hospital water systems to control waterborne pathogens (Huang et al. 2008; Rohr et al. 1999), and copper sulfate is used as an algaecide and bactericide in drinking water in the United States (NSF 2021). Since copper's antimicrobial properties make it useful for drinking water treatment and distribution, it also has potential uses for reducing microbial contamination and health care-associated infections by controlling microorganisms in heating ventilation and air-conditioning systems (Arendsen et al. 2019; Vincent et al. 2016). Aside from possible use for controlling contamination and infections, copper has some other uses in medicine and health care. Copper-containing ointments are used in anthroposophical medicine (Gorter et al. 2004). Copper intrauterine devices (IUDs) are commonly used forms of birth control (Gu et al. 2012; Wildemeersch et al. 2014). Copper is also available in multivitamins, dietary supplements, and fortified foods.

Copper and copper compounds have many applications in agriculture, food processing, and production. Copper and copper compounds are registered as fungicides, bactericides, algaecides, herbicides, insecticides, and molluscicides for use on almost all food and feed crops (EPA 2009b). Copper can be present in growth stimulants and fertilizers for plants. Copper sulfate is used in land-applied pesticides in United States agriculture, primarily as a fungicide and bactericide for fruits and vegetables, and as a algaecide n reservoirs and waterways (Lifset et al. 2012). Industrial applications of copper sulfate include use as an activator in froth flotation of sulfide ores, production of chromated copper arsenate wood preservatives, electroplating, azo dye manufacturing, mordant for textile dyes, petroleum refining and in the manufacture of other copper salts such as copper hydroxide and copper carbonate (Mannsville Chemical Products 1984).

USGS estimates annual agricultural pesticide use in U.S. counties as part of the Pesticide National Synthesis Project. Estimated use for copper and copper compounds pesticides is presented in Table 5-3.

Table 5-3. Estimated Pesticide Use (kg) in the United States from 2013-2017

	Range				
Compound	2013	2014	2015	2016	2017
Copper	345,176–391,165	393,407–435,968	416,161–478,510	528,201–540,102	435,373–531,312
Copper hydroxide	2,218,149–2,378,077	1,867,194–1,951,160	1,952,598–2,129,077	1,989,599–2,101,067	2,063,632–2,204,163
Copper sulfate	924,911–1,017,101	1,014,369–1,077,501	969,716–1,026,607	931,604–958,736	1,135,793–1,270,874
Copper sulfate tribasic	465,364	456,608	601,444	603,385	638,114
Copper oxychloride	142,874–144,116	116,065–120,032	214,475–247,904	206,268–217,275	257,866–290,005

Table 5-3. Estimated Pesticide Use (kg) in the United States from 2013-2017

	Range				
Compound	2013	2014	2015	2016	2017
Copper oxychloride	50,723 – 136,649	67,750 – 114,027	67,025-107,164	61,052-100,496	10,535 – 16,516
Copper octanoate	4,439 – 4,463	7,730 – 7,938	10,056 – 10,179	11,184 – 11,230	12,117 – 12,448

Source: USGS 2017a

Copper is widely used in many applications, and demand is projected to increase. However, as ore grades and natural deposits are depleted, more emphasis may be put on a circular economy of copper and secondary production (Ciacci et al. 2020; Shipper et al. 2018). Under different models to explore the impacts of different futures on global copper supply/demand, demand is estimated to increase by 300-2100% through 2100, depending on population, welfare, and renewable energy development (Schipper et al. 2018). All scenarios result in increased demand that would deplete copper resources (Schipper et al. 2018). While increasing secondary flows and recycling could meet increasing demands and result in a circular economy, most scenarios analyzed by Ciacci et al. (2020) for Europe would not meet greenhouse gas reduction targets unless green technology and equitable lifestyles are emphasized.

5.2.4 Disposal

Based on a review of several papers, it is estimated that 40%-84% of copper in waste materials is recovered, depending on the country (Schlesinger et al. 2011c). The recycling rate in the United States is estimated to be between 29 and 49% (Lifset et al. 2002; Lifset et al. 2012). In 2019, copper in scrap was estimated to contribute about 35% of the U.S. copper supply (USGS 2020a). There are several recycling processes depending on the copper content of scrap material, other metals present in the scrap, and size. Clean, high-grade copper scrap can be re-melted and recovered without further refining, while scrap of lower grade must be refined, often through electrorefining (Samuelsson and Bjorkman 2014). Copper is removed from industrial wastewaters using a variety of processes, including chemical precipitation, ion exchange, membrane filtration, flotation, electrochemical treatment, coagulation/flocculation, and adsorption (Bilal et al. 2013). Copper and copper compounds that are not recycled are disposed of in landfills (Cui and Zhang 2008).

In case of a solid copper sulfate spill on land, the solids should be protected from rain and fire-fighting water by covering the material with plastic sheeting (AAR 1994). In the event of a water spill, the copper sulfate should be neutralized with crushed limestone, slaked lime, or sodium bicarbonate, and the solidified masses should be removed (AAR 1994).

5.3 RELEASES TO THE ENVIRONMENT

The Toxics Release Inventory (TRI) data should be used with caution because only certain types of facilities are required to report (EPA 2005). This is not an exhaustive list. Manufacturing and processing facilities are required to report information to the TRI only if they employ ≥10 full-time employees; if their facility is included in Standard Industrial Classification (SIC) Codes 10 (except 1011, 1081, and 1094), 12 (except 1241), 20–39, 4911 (limited to facilities that combust coal and/or oil for the purpose of generating electricity for distribution in commerce), 4931 (limited to facilities that combust coal and/or oil for the purpose of generating electricity for distribution in commerce), 4939 (limited to facilities that combust coal and/or oil for the purpose of generating electricity for distribution in commerce), 4953 (limited to facilities regulated under RCRA Subtitle C, 42 U.S.C. section 6921 et seq.), 5169, 5171, and 7389 (limited S.C. section 6921 et seq.), 5169, 5171, and 7389 (limited to facilities primarily engaged in solvents recovery services on a contract or fee basis); and if their facility produces, imports, or processes ≥25,000 pounds of any TRI chemical or otherwise uses >10,000 pounds of a TRI chemical in a calendar year (EPA 2005).

Industrial releases such as industrial effluents, mining and production of copper and other metals, municipal solid waste management, and fossil fuel combustion account for a portion of the total environmental releases of copper and copper compounds. Other sources of copper released into the environment include pesticides, marine paints, animal feeds, fertilizers, fireworks, brake pad wear, copper pipe corrosion, leaching from architectural surfaces, releases from treated wood, vehicle fluid leaks, tire wear, wood combustion, biomass burning, and sewage sludge (Lifset et al. 2012; Rauch and Graedel 2007). Natural sources of copper releases include windblown dust, volcanoes, decaying vegetation, forest fires, and sea spray (Georgopoulos et al. 2001; Rauch and Graedel 2007).

5.3.1 Air

Estimated releases of 329,895 pounds (~150 metric tons) of copper to the atmosphere from 2,428 domestic manufacturing and processing facilities in 2018 accounted for about 1.7% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). These releases are summarized in Table 5-4. Estimated releases of 567,357 pounds (~257 metric tons) of copper compounds to the atmosphere from 1,635 domestic manufacturing and processing facilities in 2018 accounted for about 0.3% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). These releases are summarized in Table 5-5.

Table 5-4. Releases to the Environment from Facilities that Produce, Process, or Use Copper

		Reported amounts released in pounds per year[b]							
							Total Release		
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site
IN	135	19,923	5,283	2,800	1,388,811	3,059,426	85,881	4,390,361	4,476,242
CA	105	4,573	1,552	0	2,581,572	4,995	2,412,938	179,754	2,592,693
TX	126	7,633	2,194	30,324	563,540	1,017,217	536,814	1,084,095	1,620,908
IL	140	25,748	18,172	250	813,374	188,624	39,065	1,007,103	1,046,169
NV	60	43,801	761	0	654,467	113,970	525,538	287,461	812,999
OK	8	49	5	0	770,369	51	769,932	542	770,474
TN	64	3,654	25,500	0	716,475	2,199	363,072	384,755	747,827
WA	198	28,180	3,328	0	297,307	370,411	242,461	456,765	699,226
GA	60	14,216	110	0	647,820	1,496	658,404	5,238	663,642
OH	68	5,329	11,468	0	584,639	23,290	448,491	176,234	624,726
KY	25	2,461	1,755	0	173,940	352,453	163,768	366,842	530,609
NC	152	8,728	1,078	0	290,422	128,664	77,837	351,054	428,891
ID	82	2,677	1,909	0	373,708	4,405	375,817	6,881	382,699
MN	63	3,430	2,722	0	258,960	37,857	60,324	242,644	302,969
NY	63	3,768	3,664	0	249,080	34,993	177,769	113,736	291,505
SC	30	14,420	9,908	0	251,880	3,833	254,090	25,951	280,041
NJ	31	10,663	253	0	128,450	139,768	63,276	215,858	279,134
AR	48	3,916	396	0	231,077	36,074	206,549	64,914	271,463
CO	196	55,002	8,575	1,034	150,460	36,260	100,711	150,620	251,331
VA	61	5,680	34,450	0	149,015	38,223	22,230	205,138	227,369
WI	94	2,498	10,569	0	188,105	23,891	162,896	62,166	225,062
MO	33	6,466	152	52,772	115,277	33,060	172,449	35,278	207,727
LA	7	35	2	0	204,648	1,202	203,628	2,259	205,887
WY	19	1,629	794	1,400	187,299	0	89,556	101,566	191,122
AL	62	3,127	536	0	121,628	24,944	93,710	56,525	150,235
PA	4	14	0	0	139,757	0	139,771	0	139,771

Table 5-4. Releases to the Environment from Facilities that Produce, Process, or Use Copper

	Reported amounts released in pounds per year[b]								
							Total Release		
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site
FL	13	549	123	0	125,913	12,412	125,887	13,111	138,997
NM	104	8,593	1,066	6,589	59,997	31,700	41,865	66,079	107,944
WV	3	77	0	0	103,692	0	103,769	0	103,769
AK	28	18,977	3,393	0	46,036	10,651	63,439	15,618	79,056
IA	46	3,017	371	0	55,189	7,899	3,102	63,374	66,476
MS	3	0	0	0	64,641	0	64,641	0	64,641
OR	4	1,018	339	0	56,972	275	1,050	57,554	58,603
UT	3	0	0	0	58,482	0	58,482	0	58,482
HI	11	8	13	0	58,305	40	58,012	354	58,366
KS	17	1,704	9	0	44,408	6,412	37,577	14,957	52,534
CT	54	1,195	574	0	33,636	16,731	13,901	38,235	52,136
MI	36	389	74	0	33,064	28	396	33,159	33,555
VT	41	44	448	0	304	21,407	86	22,117	22,203
AZ	20	1,433	44	0	17,332	2,798	15,880	5,727	21,607
MA	4	0	5	0	21,054	2	20,353	708	21,061
PR	22	525	95	19	18,096	348	18,438	645	19,083
NE	14	11,935	3	0	8	4,006	11,935	4,017	15,952
RI	15	260	78	0	1,449	8,500	269	10,018	10,287
NH	22	31	238	0	153	7,140	37	7,525	7,562
ME	7	6	82	0	3,852	69	10	3,998	4,008
SD	13	2,485	11	0	159	46	2,571	130	2,701
MD	7	0	273	0	2	61	0	336	336
DE	2	10	18	0	5	0	10	23	33
ND	4	20	11	0	1	0	28	5	33
MT	1	0	0	0	0	0	0	0	0
Total	2,428	329,895	152,405	95,188	13,034,828	5,807,829	9,088,718	10,331,429	19,420,147

Table 5-4. Releases to the Environment from Facilities that Produce, Process, or Use Copper

	Reported amounts released in pounds per year[b]							
							Total Release	
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site

Source: TRI18 2020; Data are from 2018

RF = Reporting Facilities; UI = Underground Injection

[a] The TRI data should be used with caution since only certain types of facilities are required to report. This is not an exhaustive list. Data are rounded to nearest whole number.
[b] Data in TRI are maximum amounts released by each facility.
[c] Post office state abbreviations are used.
[d] Number of reporting facilities.
[e] The sum of fugitive and point source releases by a given facility.
[f] The sum of on-site surface water discharges, and off-site transfers to wastewater treatment-(metals only), and publicly owned treatment works (POTWs) (metal and metal compounds).
[g] The sum of on-site and off-site disposal to underground injection wells (Class I wells and Class II-V).
[h] The sum of on-site and off-site disposal to: Resource Conservation and Recovery Act (RCRA) subtitle C landfills, other landfills, RCRA subtitle C surface impoundments, other surface impoundments, land treatment, other land disposal.
[i] Includes the sum of off-site transfers to storage only, solidification/stabilization (metals only) disposal, other off-site management, waste broker for disposal, unknown.
[j] Total on-site disposal or other releases of the chemical including emissions to air, surface water discharges, land, and underground injection wells.
[k] Total amount of chemical transferred off-site for disposal or other releases, including to POTWs

Table 5-5. Releases to the Environment from Facilities that Produce, Process, or Use Copper Compounds

		Reported amounts released in pounds per year[b]							
							Total Release		
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site
UT	20	40,536	1,351	0	69,016,437	3,206	69,045,478	16,052	69,061,530
AZ	28	123,811	17,201	0	45,689,957	17,474	45,596,152	252,292	45,848,444
MT	11	97,590	0	0	12,633,619	362	12,676,390	55,181	12,731,571
NV	15	1,447	0	0	8,576,085	22	8,577,532	22	8,577,555
AK	4	308	81	0	7,940,479	0	7,940,868	0	7,940,868
AL	65	2,239	4,955	0	5,248,165	32,979	4,909,339	379,000	5,288,339
MO	48	16,388	5,944	0	4,994,977	22,733	4,895,396	144,646	5,040,042
MI	59	8,268	13,963	0	1,899,392	59,217	1,651,526	329,314	1,980,840
OH	88	22,166	20,320	487,459	1,095,825	197,821	981,489	842,102	1,823,591
TX	117	28,722	43,278	95,834	1,216,687	245,388	991,798	638,111	1,629,909
IN	67	27,096	17,031	964	1,453,457	117,998	939,425	677,121	1,616,547
CA	68	1,141	3,901	0	1,601,273	7,163	1,087,899	525,579	1,613,478
TN	51	3,089	16,463	0	1,544,928	5,782	1,332,230	238,032	1,570,263
LA	36	9,620	5,820	2,806	1,409,159	93,215	1,207,486	313,134	1,520,620
PA	85	20,420	3,839	250	1,167,455	92,396	588,139	696,221	1,284,360
IL	83	5,027	21,694	0	849,895	216,132	440,133	652,616	1,092,749
Y	40	3,534	21,247	1	1,011,104	42,012	850,399	227,499	1,077,898
ID	14	1,696	411	0	978,906	3,894	750,660	234,248	984,908
WV	19	2,985	379	0	782,083	4,304	540,062	249,690	789,752
AR	49	6,320	2,503	0	466,070	273,365	381,434	366,823	748,257
WA	21	1,474	410	0	177,795	425,057	90,199	514,537	604,736
NC	66	17,453	1,642	0	539,083	45,820	369,936	234,063	603,999
MS	30	3,754	548	55,212	374,355	9,925	79,235	364,559	443,794
OK	31	1,891	2,248	0	423,369	446	361,074	66,880	427,954
IA	38	7,119	7,514	5	390,669	15,172	129,887	290,591	420,478
OR	24	12,642	1,238	0	329,651	11,690	313,562	41,658	355,221
MN	45	1,164	1,166	0	286,030	3,268	230,781	60,848	291,628

Table 5-5. Releases to the Environment from Facilities that Produce, Process, or Use Copper Compounds

		Reported amounts released in pounds per year[b]							
							Total Release		
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site
NY	26	24,424	101,111	0	37,035	121,362	52,962	230,970	283,932
CO	17	5,493	305	0	260,595	0	242,850	23,542	266,393
SC	36	4,628	6,366	1	217,902	35,532	62,427	202,003	264,430
NJ	15	6,006	10,877	0	41,698	194,852	6,007	247,426	253,432
VA	39	4,757	6,149	0	180,582	27,490	126,098	92,880	218,978
GA	60	9,889	29,438	0	123,181	38,475	92,111	108,871	200,983
NE	19	2,994	436	0	188,229	2,065	176,504	17,220	193,724
WY	6	1,842	24	0	189,625	0	138,896	52,595	191,491
ND	6	716	86	3	177,405	2,477	116,263	64,424	180,687
KS	21	1,120	10,184	281	137,951	19,920	107,912	61,543	169,455
WI	49	2,673	6,467	0	145,454	8,457	21,101	141,949	163,050
NM	8	5,864	1,231	0	109,924	40,550	116,798	40,770	157,568
FL	39	643	9,399	0	84,187	12,892	74,270	32,852	107,122
CT	17	3,028	9,150	3,167	51,958	38,748	3,395	102,657	106,052
RI	8	34	428	0	0	44,792	418	44,836	45,254
ME	1	106	160	0	42,985	0	266	42,985	43,251
MD	10	11	78	0	26,451	13,949	19	40,470	40,489
MA	8	1,110	1,516	14,381	7,806	4,633	1,413	28,034	29,447
PR	7	23,970	8	0	703	0	23,978	703	24,681
SD	8	87	0	0	18,444	0	18,392	139	18,531
DC	1	0	0	0	10,103	0	10,103	0	10,103
NH	5	60	434	0	10	41	315	231	545
DE	6	0	0	0	0	0	0	0	0
VT	1	0	0	0	0	0	0	0	0
Total	1,635	567,357	408,995	660,365	174,149,134	2,553,076	168,351,009	9,987,919	178,338,928

Table 5-5. Releases to the Environment from Facilities that Produce, Process, or Use Copper Compounds

		Reported amounts released in pounds per year[b]							
							Total Release		
State[c]	RF[d]	Air[e]	Water[f]	UI[g]	Land[h]	Other[i]	On-site[j]	Off-site[k]	On and off-site

Source: TRI18 2020; Data are from 2018

RF = Reporting Facilities; UI = Underground Injection

[a] The TRI data should be used with caution since only certain types of facilities are required to report. This is not an exhaustive list. Data are rounded to nearest whole number.
[b] Data in TRI are maximum amounts released by each facility.
[c] Post office state abbreviations are used.
[d] Number of reporting facilities.
[e] The sum of fugitive and point source releases by a given facility.
[f] The sum of on-site surface water discharges, and off-site transfers to wastewater treatment-(metals only), and publicly owned treatment works (POTWs) (metal and metal compounds).
[g] The sum of on-site and off-site disposal to underground injection wells (Class I wells and Class II-V).
[h] The sum of on-site and off-site disposal to: Resource Conservation and Recovery Act (RCRA) subtitle C landfills, other landfills, RCRA subtitle C surface impoundments, other surface impoundments, land treatment, other land disposal.
[i] Includes the sum of off-site transfers to: storage only, solidification/stabilization (metals only) disposal, other off-site management, waste broker for disposal, unknown.
[j] Total on-site disposal or other releases of the chemical including emissions to air, surface water discharges, land, and underground injection wells.
[k] Total amount of chemical transferred off-site for disposal or other releases, including to POTWs.

Copper is emitted into the air from both natural and anthropogenic sources. Global atmospheric concentrations and releases of copper from manmade and natural sources have been estimated (Rauch and Graedel 2007) Estimates for the natural and anthropogenic emissions copper from various sources are shown in Table 5-6 and Table 5-7. Based on these data, $6.9x10^7$ kg/year of copper from natural sources is estimated to be emitted to the atmosphere.

Table 5-6. Global Emissions of Copper from Natural Sources in the mid-1990s

Source	Emissions (Gg[a] Cu/year)
Windblown dust	50
Sea salt spray	13
Biomass burning	3.3
Agricultural burning	0.14
Volcanic outgassing	2.7

[a]One Gg is one billion (10^9) grams. It is the same as one million (10^6) kilograms.
Source: Rauch and Graedel 2007

Table 5-7. Global Emissions of Copper from Anthropogenic Sources in the mid-1990s

Source	Emissions (Gg[a] Cu/year)
Metal production	18
Nonferrous metal production	18
Iron and steel production	0.14
Fossil fuel combustion	7.1
Metal fabrication	1.4
Metal discard management	0.62

[a]One Gg is one billion (10^9) grams. It is the same as one million (10^6) kilograms.
Source: Rauch and Graedel 2007

Windblown dusts account for an estimated global emission of $5.0x10^7$ kg/year of copper into the atmosphere (Rauch and Graedal 2007). Other natural sources of copper emitted into air (in order of highest to lowest worldwide emissions) are sea salt spray, biomass burning, and volcanoes. Anthropogenic emission sources include nonferrous metal production, fabrication, and use; fossil fuel combustion; metal production; and mining. Lifset et al. (2012) estimates the following emissions to the atmosphere in the United States: fireworks ($2.2x10^5$ kg/year), copper primary production ($4.7x10^5$ kg/year), copper waste management ($1.9x10^5$ kg/year), coal combustion ($1.36x10^6$ kg/year), oil combustion ($4.5x10^5$ kg/year), metals production ($2.0x10^4$ kg/year), and wood combustion ($4.0x10^4$ kg/year).

The EPA conducted a detailed study of the total amount of copper emitted into the atmosphere (Weant

1985). The sources of emissions and the estimated quantities of copper emitted in 10^6 kg/year are presented in Table 5-8.

Table 5-8. Copper Emissions into the Atmosphere in 1984

Source	Emissions (10^6 kg/year)
Primary copper smelter	0.042 – 6
Copper and iron ore processing	0.48 – 0.66
Combustion sources	0.045 – 0.36
Primary zinc smelting	0.024 – 0.34
Municipal incinerators	0.0033 – 0.27
Iron and steel production	0.112 – 0.24
Secondary copper smelters	0.16
Gray iron foundries	0.079
Primary lead smelting	0.0055 – 0.065
Copper sulfate production	0.045
Brass and bronze production	0.0018 – 0.036
Carbon black production	0.013
Ferroalloy production	0.0019 – 0.0032

[a]One Gg is one billion (10^9) grams. It is the same as one million (10^6) kilograms.
Source: Weant 1985

Using the ranges of copper emitted from these sources, it is estimated that annual U.S. copper emissions into air are 0.94 to 79.74(x10^5) kg. No recent reports updating these estimates have been found but due to changes in these industries over time, emissions are likely different now.

Daily stack emission rates have been reported for three coal-burning power plants on a kg/day/1,000 megawatt basis (Que Hee et al. 1982); they were 0.3–0.7 and 2.00 kg/day/1,000 megawatt for those using low-sulfur western coal and high-sulfur eastern coal, respectively. This amounted to annual emission rates of 110–260 megawatt for the low-sulfur western coal and 730 kg/1,000 megawatt for the high-sulfur eastern coal.

Emission factors in grams of copper released to the atmosphere per ton of product have been estimated for various industries (Nriagu and Pacyna 1988). These factors would enable estimation of an industry's copper emissions from its production volume. Missing from these emission estimates is fugitive dust arising from drilling, blasting, loading, and transporting operations associated with copper mining. The most common control for reducing fugitive dust is the manual use of water sprays (EPA 1980). The highest concentrations of copper in atmospheric particulate matter were obtained from mining activities, primary and secondary production, and industrial manufacturing (Table 5-8).

Romo-Kröger et al. (1994) were able to show, through the use of radioactive tracers and cluster analysis of inter-elemental correlations, that copper, arsenic, sulfur, and zinc measured near a copper smelter in

Chile were derived from the plant and not from the surrounding soil. The concentration of copper in air near the plant decreased from 66 to 22 ng/m^3 of fine particles and from 131 to 50 ng/m^3 of coarse particles during a period of inactivity at the plant, demonstrating the contribution of plant emissions to copper levels in the surrounding area.

Table 5-9. Concentrations of Copper in Particulate Matter (<10 µm) Generated from Various Sources

Source[a]	Median
Metal mining	6.17*
Secondary metal production	4.60*
Primary metal production	3.50*
Industrial manufacturing	2.16*
Steel production	0.55*
Gray iron foundries	0.19*
Steel foundry, general	0.17*
Solid waste	0.09*
Food and agriculture	0.05*
Chemical manufacturing	0.03*
Petroleum industry	0.03*
Gasoline vehicle exhaust	0.05†
Paved road dust	0.0162†
Construction dust	0.0102†
Landfill dust	0.0102†
Unpaved road dust	0.0087†
Agricultural lands, dust	0.0067†
Diesel vehicle exhaust	0.003†

[a] Values obtained from CEIDARS 2000
* Data obtained from USEPA Speciate 3.0; Shareef, G.S; Radian, September 1987
† Data obtained from KVB Literature Search

Copper and other pollutants are present in fugitive dust originating from copper production sites or from waste sites. In one study, the amount of airborne copper and other heavy metals deposited near a large refuse dump that received municipal and industrial waste and sewage sludge was determined by first measuring the amount of the metal accumulated in moss bags suspended 1–3 meters above the ground. The deposition rate was then determined from the amount of copper in the moss bags accumulated over the summer of 1985 and compared with that for an agricultural control area. The mean copper deposition rates in the two areas were about the same: 0.55 mg/kg-month (range of 0.04–1.6 mg/kg-month) over the refuse dump and 0.51 mg/kg-month (range of 0.26–0.76 mg/kg-month) in the control area (Lodenius and Braunschweiler 1986). Lodenius and Braunschweiler (1986) concluded that the refuse dump did not contribute to copper concentrations in urban air above normal values.

A study of automobile exhaust emitted from light-duty vehicles conducted in Denver, Colorado showed that this source of copper emission makes a small local contribution to copper in air. The amount of

copper emitted in exhaust from automobiles powered by regular gasoline has been measured to be 0.001–0.003 mg/mile driven using the Urban Dynamometer Driving Schedule (UDDS) of the Federal Test Schedule (FTS) during the summer of 1996 and the winter of 1997 (Cadle et al. 1999). Diesel-powered vehicles were also studied and found to emit 0.005–0.039 mg of copper per mile driven for vehicles using #2 diesel fuel.

Only in a few cases has the form of copper released into the air been determined. Copper released into the atmosphere can be in particulate matter in the elemental form or in the form of an oxide, sulfate, or carbonate. Because copper smelters co-emit sulfur oxides gases, copper is expected to be released largely as the sulfate in particulate matter from these facilities. Combustion processes are reported to release copper into the atmosphere as the oxide, elemental copper, and adsorbed copper. Cupric oxide has been identified in emissions from steel manufacturing and in fly ash from oil-fired power plants and open-hearth steel mills (Perwak et al. 1980). Copper associated with particles (≤10 μm) has been suggested to originate from windblown soil and dust (Schroeder et al. 1987). Generally, aerosols from sea spray, dust, and volcanic mineral emissions tend to be larger than particles formed by condensation of gases in the troposphere (Buseck and Posfai 1999).

Copper and copper compounds were detected in air at 19 of the 929 NPL hazardous waste sites where copper had been detected in other environmental media (ATSDR 2019).

In a study of particulate matter emitted by fireworks, Hickey et al. (2020) sampled 10 types of fireworks and found that four of the 12 samples contained copper at concentrations of 12,000 to 53,000 ppm in the PM_{10} size range. Using an emission factor of 3,000 ppm developed by the European Copper Institute, Lifset et al. (2012) estimated that releases from fireworks in the United States increased from 40 metric tons in 1975 to 220 metric tons in 2000.

5.3.2 Water

Estimated releases of 152,405 pounds (~69 metric tons) of copper to surface water from 2,428 domestic manufacturing and processing facilities in 2018 accounted for about 0.8% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). An additional 58,472 pounds (~24 metric tons) were released to publicly owned treatment works (POTWs) (TRI18 2020). These releases are summarized in Table 5-4.

Estimated releases of 408,995 pounds (~186 metric tons) of copper compounds to surface water from 1,635 domestic manufacturing and processing facilities in 2018 accounted for about 0.2% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). An additional

51,907 pounds (~23 metric tons) were released to publicly owned treatment works (POTWs) (TRI18 2020). These releases are summarized in Table 5-5.

Sources of copper releases to water include algaecides, marine paints, corrosion of metallic copper, architectural uses, chromated copper arsenate (CCA) wood management, industrial effluent, and copper mining leachate (Lifset et al. 2012). Copper and copper compounds were detected in water at 195 of the 929 NPL hazardous waste sites where copper has been detected in environmental media (ATSDR 2019).

Copper is a natural constituent of soil and will be transported into streams and waterways in runoff either due to natural weathering or anthropogenic soil disturbances (Rader et al. 2018). Sixty-eight percent of releases of copper to water is estimated to derive from soil runoff and weathering, while copper sulfate use represents 13% of releases to water and urban runoff contributes 2% (Perwak et al. 1980). In the absence of specific industrial sources, runoff is the major factor contributing to elevated copper levels in river water (Nolte 1988). In the previous EPA National Urban Runoff Program, 86 samples of runoff from 19 cities throughout the United States were analyzed, and copper was found in 96% of samples, at concentrations of 1–100 µg/L (equivalent to ppb) with a geometric mean of 18.7 µg/L (Cole et al. 1984).

Giusti et al. (1993) provided estimates of global anthropogenic and natural copper inputs into oceans that are derived from two sources: atmospheric deposition and riverine input. Atmospheric input has been estimated at 14–45x10^6 kg/year for copper in a dissolved form (e.g., rainwater) and 2–7x10^6 kg/year for copper in a particulate form (e.g., aerosols). Riverine input is estimated to be 10x10^6 kg/year as dissolved copper and 1,500x10^6 kg/year as copper bound to particulates.

Domestic wastewater is the major anthropogenic source of copper in waterways (Isaac et al. 1997; Nriagu and Pacyna 1988). Studies in Cincinnati and St. Louis showed discharges of copper into sewer systems from residential areas to be significant, with an average loading of 42 mg/person/day (Perwak et al. 1980). In a more comprehensive review, Jenkins and Russell (1994) reported a range of average copper loadings derived from residential and some small industrial contributions of 2.8–83 mg/person/day. Concentrations of copper in influents to 239 wastewater treatment plants (12,351 observations) were 0.0001–36.5 ppm, and the median value was ~0.4 ppm (Minear et al. 1981). Copper is not entirely removed in POTWs, and releases from these facilities contribute ~8% of all copper released to water (Perwak et al. 1980). Inputs into the Narraganset Bay, Rhode Island, in decreasing order of importance, are sewage effluent, rivers, urban runoff, and atmospheric fallout (Mills and Quinn 1984; Santschi et al. 1984). Ninety percent of both dissolved and particulate copper was from the effluent of sewage treatment plants that discharged into the Providence River.

While some copper is removed from the waste stream by sewage treatment facilities, considerable copper remains in the effluent and is released into receiving waters (EPA 1981; Perwak et al. 1980). Because removal efficiencies for copper from waste streams tend to remain constant rather than proportional to influent copper concentrations, increases in copper concentrations in POTW influent streams will also result in increased copper concentrations in the effluent streams (Isaac et al. 1997). The copper in domestic wastewater has been found to make up a substantial fraction of the copper found in POTW influent in the wastewater systems of four Massachusetts municipalities. The range of removal efficiencies reported for pilot and full-scale plants suggests that removal depends strongly on plant operation or influent characteristics.

A source of copper released into waterways is from urban stormwater runoff. Copper in stormwater runoff originates from the sidings and roofs of buildings, various emissions from automobiles, and wet and dry depositional processes (Davis et al. 2001). Concentrations of between 1 and 100 µg/L of copper in stormwater runoff have been measured (Georgopoulos et al. 2001). Stormwater runoff normally contributes approximately 2% to the total copper released to waterways. In contrast, copper in runoff that is obtained from the natural weathering of soil or is released from disturbed soils contributes 68% of the copper released to waterways (Georgopoulos et al. 2001).

The best data on typical POTWs using secondary treatment show that 55–90% of copper is removed in these plants with a median and mean removal efficiency of 82% (Perwak et al. 1980). By contrast, those plants using only primary treatment had a 37% median removal efficiency. A more recent study focused on heavy metal removal in three POTWs that received primarily municipal sewage and used activated sludge as a secondary treatment. The study looked at removals in both the primary and secondary treatment stage. The mean removal of soluble copper and total copper after secondary treatment were 49–82 and 83–90%, respectively. The average copper concentration in the final effluent was 17–102 ppb, which would amount to an output of between 0.58 and 3.47 kg of copper into receiving waters per day, based on an effluent volume of 34,000 cubic meters (9 million gallons) per day (Aulenbach et al. 1987; Stephenson and Lester 1987).

Overflow outfalls within combined sewer systems (e.g., combination of domestic and industrial wastewater plus stormwater) are the primary sources of copper pollutants entering estuaries and other coastal areas of the United States (Crawford et al. 1995; Georgopoulos et al. 2001; Huh 1996; Iannuzzi et al. 1997). For example, Crawford et al. (1995) compiled a summary of the sources of various metals and other contaminants into the Newark Bay estuary. The mass loadings of copper into the estuary as a function of source are (in kg/day): discharges from the Passaic Valley Commission and Middlesex

County Sewerage Authority, 126.5; municipal treatment systems, 103.4; stormwater runoff, 62.2; combined sewer overflows, 48.0; tributary flow, 39.1; and industry direct discharge, 8.82.

Discharges to water from active mining and milling are small, and most of the western U.S. operations do not release any water because water is a scarce resource and is recycled (Perwak et al. 1980). Discharges from electroplating operations are either made directly to the water environment or indirectly via POTWs. Runoff from abandoned mines is estimated to contribute 314 metric tons annually to surface water (Perwak et al. 1980). These discharges are primarily insoluble silicates and sulfides and readily settle out into stream, river, or lake beds. Releases from manufactured products containing copper may be substantial but are difficult to predict. Corrosion of copper in plumbing or construction may result in direct discharges or runoff into waterways. Copper and brass production releases relatively little copper to water.

Wastewater generated from copper mining operations comes from seepage, runoff from tailing piles, or utility water used for mine operation. The amount of wastewater generated ranges from 0–300 L water/metric ton of ore mined for open pit copper mines and 8–4,000 L water/metric ton of ore mined underground (EPA 1980). Copper concentrations in wastewater from a selected open pit and underground copper mine were 1.05 and 0.87 ppm, respectively. Data regarding copper concentrations in wastewater associated with selected concentrating, smelting, and refining operations can be found in EPA (1980). Drainage from mining operations and abandoned mines has been shown to have an effect on copper content in local surface waters with concentrations as high as 69,000 ppb being measured (Rösner 1998).

Results of an EPA industrial effluent survey show that mean and maximum levels of copper in treated wastewater from six industries exceeded 1 and 10 ppm, respectively (EPA 1981). These industries and their mean and maximum discharges in ppm are inorganic chemicals manufacturing (<1.6, 18); aluminum forming (<160, 2,200); porcelain enameling (1.3, 8.8); gum and wood chemicals (1.4, 3.0); nonferrous metals manufacturing (1.4, 27.0); and paint and ink formulation (<1.0, 60.0). Emission factors in nanograms of copper released per L of water outflow have been estimated for various industries. These factors would enable estimation of an industry's copper releases if the discharge volumes were known (Nriagu and Pacyna 1988).

Effluents from power plants that use copper alloys in the heat exchangers of their cooling systems discharge copper into receiving waters (Harrison and Bishop 1984). The largest discharges occur after startup and decrease rapidly thereafter. At the Diablo Canyon Nuclear Power Station, a very high startup discharge containing 7,700 ppb of copper fell to 67 ppb after 24 hours (Harrison et al. 1980). During

normal operation at two nuclear power stations $6.5x10^6$ cubic meters (1,700 million gallons) of seawater per day is used as cooling water for these facilities and discharged into the ocean with copper levels in the effluent ranging between 0.6 and 3.3 ppb (Harrison et al. 1980). This amounts to a total output of copper in the discharged seawater of 3.9–42 kg per day or 1,400–15,000 kg/annum from these two power plants. Except for after start-up of the cooling system, most of the soluble copper (that which passes through a 0.45 µm filter) discharged was in bound forms (Harrison et al. 1980). During normal operation, <20% of the copper released was in the <1,000 molecular weight fraction, which contains the more available copper species.

Copper sulfate is added directly to lakes, reservoirs, and ponds for controlling algae. However, the copper concentration in the water column generally returns to pretreatment levels within a few days (Effler et al. 1980; Perwak et al. 1980). The reduction in dissolved copper during this period was accompanied by an increase in particulate copper (e.g., sorption to algae or other organic matter, which settles into the sediments of these bodies of water). The copper in the settled particulates is in equilibrium with the water column, which greatly favors copper in a bound state.

A potential source of copper release into waterways is leachate from municipal landfills. Copper concentrations in leachate obtained from waste sites have been found to vary widely. For example, copper concentrations in leachate from municipal landfills have been found to range from 0.005 to 1,110 ppm (Christensen et al. 1994; Perwak et al. 1980; Roy 1994). Although copper was measured in these leachates, its origin may not be from copper contained within the waste site, but from the surrounding soils. Cyr et al. (1987) reported that leachate from three municipal landfills in New Brunswick, Canada, did not contain copper concentrations significantly above those in control samples representing the surrounding soil types. Therefore, the emissions of copper from landfills into leachates should be made relative to the contribution of copper from surrounding soils, as determined from appropriately selected control samples.

Copper can enter surface waters as a result of agricultural runoff. For example, estimated loading rates of copper into surface water from irrigation water runoff near the Stillwater National Wildlife Refuge ranged from 0.307 to 8.34 mg/hour, depending on what period of the irrigation season samples were taken (Kilbride et al. 1998). The highest loading rates were obtained during the middle period (August through mid-September) of the irrigation season. The copper in the runoff water was found to be predominantly bound to drift material in the water (e.g., algae, vascular plants, invertebrates, vertebrates, and detrital material).

5.3.3 Soil

Estimated releases of 13,034,828 pounds (~5,912 metric tons) of copper to soils from 2,428 domestic manufacturing and processing facilities in 2018 accounted for about 67% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). An additional 95,188 pounds (~43 metric tons), constituting about 0.5% of the total environmental emissions, were released via underground injection (TRI18 2020). These releases are summarized in Table 5-4.

Estimated releases of 174,149,134 pounds (~78,993 metric tons) of copper compounds (excluding elemental copper) to soils from 1,635 domestic manufacturing and processing facilities in 2018 accounted for about 98% of the estimated total environmental releases from facilities required to report to the TRI (TRI18 2020). An additional 660,365 pounds (~300 metric tons), constituting about 0.4% of the total environmental emissions, were released via underground injection (TRI18 2020). These releases are summarized in Table 5-5.

Copper and copper compounds were detected in soils from 245 of the 929 NPL hazardous waste sites where copper had been detected in environmental media (ATSDR 2019). An estimated 97% of copper released from all sources into the environment is primarily released to land (Perwak et al. 1980). These include primarily tailings and overburdens from copper mines and tailings from mills. The copper in tailings represents the portion of copper that could not be recovered from the ore and is generally in the form of insoluble sulfides or silicates (Perwak et al. 1980). These wastes accumulate in mining states. Other releases to land include sludge from POTWs, municipal refuse, waste from electroplating, iron, and steel producers, discarded copper products (e.g., plumbing, wiring) that are not recycled, fungicides, animal feed, fertilizers, brake pads, vehicle leaks, and tire wear (Lifset et al. 2012; Perwak et al. 1980). The copper content of municipal solid waste is ~0.16%. Much of this waste is landfilled directly or is in the form of residues following incineration. Emission factors in milligrams of copper released per gram of solid waste have been estimated for various industries. These factors would enable estimation of an industry's copper releases in terms of total quantity of solid waste discharged. Sludge from sewage treatment plants is a major source of copper released to land (Nriagu and Pacyna 1988). Agricultural products are believed to constitute 2% of the copper released to soil (Perwak et al. 1980).

5.4 ENVIRONMENTAL FATE

5.4.1 Transport and Partitioning

Air. Copper is released to the atmosphere in the form of particulate matter or adsorbed to particulate matter. It is removed by gravitational settling (bulk deposition), dry deposition (inertial impaction

characterized by a deposition velocity), in-cloud scavenging (attachment of particles to rain droplets within clouds), and washout (collision and capture of particles by falling raindrops below clouds) (Schroeder et al. 1987). The removal rate and distance traveled from the source depend on a number of factors, including source characteristics, particle size, turbulence, and wind velocity.

Gravitational settling governs the removal of large particles with mass median aerodynamic (MMA) diameters of >5 µm, whereas smaller particles are removed by the other forms of dry and wet deposition. The importance of wet to dry deposition generally increases with decreasing particle size. The scavenging ratio (ratio of the copper concentration in precipitation [ppm] to its air concentration [µg/m^3]) for large particles displays a seasonal dependence that reflects more effective scavenging by snow than by rain (Chan et al. 1986). Copper from combustion sources is often adsorbed to sub-micron particulate matter. Thermal process may also release copper oxide or elemental copper as a vapor or copper adsorbed to larger particulates (Perwak et al. 1980). Copper adsorbed to sub-micron particles remain in the troposphere for an estimated 7–30 days. In that time, some copper may be carried far from its source (Perwak et al. 1980).

Rates of metal deposition (e.g., depositional fluxes) vary between dry and wet depositional processes and show spatial variability. Dry depositional fluxes of copper tend to be higher in highly urbanized areas and lower in less urbanized areas or areas with minimal anthropogenic activity. For example, average depositional rates were 0.06 mg/m^2/day in Chicago, Illinois, 0.007 mg/m^2/day in South Haven, Michigan, and 0.01 mg/m^2/day 6 to 10 km offshore of Lake Michigan (Paode et al. 1998). Estimated copper deposition rates in urban areas are 0.119 and 0.164 kg per hectare per year (kg/ha/year) or 0.0326 and 0.0449 mg/m^2/day for dry and wet deposition, respectively (Schroeder et al. 1987). Bulk deposition reportedly ranges from 0.002–3.01 kg/ha/year or 0.0005–0.825 mg/m^2/day (Golomb et al. 1997; Landing et al. 1995; Schroeder et al. 1987). For rural areas, the range of bulk deposition reportedly is 0.018–0.5 kg/ha/year or 0.0049–0.1 mg/m^2/day, and wet deposition is 0.033 kg/ha/year or 0.0090 mg/m^2/day. The washout ratio is 140–751 (Schroeder et al. 1987).

Levels of airborne copper measured at a rural site in Bondville, IL were similar to regional background levels in other urban study sites with only episodic increases, depending on wind speed and direction and location relative to local point sources. In one urban study site (East St. Louis), smelters were the primary source of copper. Copper depositional fluxes followed an exponential decay as one transitions from urban to rural settings (Sweet et al. 1993). Soil was not the major source of copper in cities or nearby rural soils but was the predominant source for copper in the atmosphere over more remote areas (Fergusson and Stewart 1992). However, high copper concentrations in snow and aerosols from polar snowfields and

remote locations has been attributed to airborne pollution and long-range transport (Annibaldi et al. 2007; Dinu et al. 2020). Sources of copper in urban areas include coal combustion, soil, tire wear, and automobile emissions (Kim and Fergusson 1994). Long-range transported emissions from combustion processes are typically associated with fine particles; however, there can be instances where the highest concentrations of copper are measured in coarse particles near point sources (Paode et al. 1998).

Estimates of depositional velocities for fine particles (<2.5 µm) and coarse particles (2.5–10 µm) in urban (Chicago) and rural (Kankalee, Illinois) areas have been made (Pirrone and Keeler 1993). The estimated depositional velocities are urban, 0.25–0.46 cm/second and rural, 0.18–0.25 for fine particles; and urban, 1.47–2.93 cm/second and rural, 0.87–1.71 cm/second for coarse particles. The differences in depositional velocities are thought to be due to higher surface roughness and wind velocities in Chicago.

Copper concentrations in particulates formed in a controlled study of waste oil combustion were (in µg/g): 687±11 (10 µm diameter), 575±8 (50 µm diameter), 552±12 (100 µm diameter), 568±9 (300 µm diameter), and 489±8 (500 µm diameter). Approximately 25% of copper was in the 10 µm fraction and ~18% was in each of the larger fractions (e.g., 50, 100, 300, and 500 µm diameter) (Nerín et al. 1999). More recent data on transport were not found.

Water. The average concentrations of copper in Lakes Superior, Erie, and Ontario are 760, 870, and 830 ng/L, respectively (Georgopoulos et al. 2001; Nriagu et al. 1996). These values were derived from measurements taken from 11, 11, and 9 nearshore and offshore sampling sites at different points in the water column up to depths of 251, 55, and 145 meters for Lakes Superior, Erie, and Ontario, respectively (Nriagu et al. 1996). In Lake Ontario, the highest copper concentrations were found at nearshore sampling sites neighboring Buffalo, New York (887–1,051 ng/L), Rochester, New York (1,041– 1,098 ng/L), and Kingston, Ontario (921–1,026 ng/L). The lowest concentrations of copper in Lake Ontario were measured in an offshore sampling site (540–710 ng/L) that was approximately 40 km from the Buffalo sampling site.

The atmospheric input of copper into the Great Lakes is 330–1,470 ng/m^2/year, which amounts to a total deposition of 8.00–35.6x10^{13} ng/year (80.0–356 kg/year). This input of copper accounts for 60–80% of the anthropogenic input into Lake Superior and 20–70% into Lakes Erie and Ontario (Georgopoulos et al. 2001; Nriagu et al. 1996). The mean residency times of copper in sediments are estimated to be 15 years in Lake Erie and 101 years in Lake Superior (Georgopoulos et al. 2001; Nriagu et al. 1996).

Much of the copper discharged into waterways is bound to particulate matter and settles out. In the water column and in sediments, copper adsorbs to organic matter, hydrous iron and manganese oxides, and clay.

In the open water column, a significant fraction of the copper is adsorbed within the first hour of introduction, and in most cases, a steady state is obtained within 24 hours (Harrison and Bishop 1984). Most dissolved copper in POTW effluent and surface runoff is mostly already in complexed form (Sedlak et al. 1997). Copper in wastewater discharged into Back River leading into Chesapeake Bay, Maryland contained 53 ppb of copper, of which 36 ppb (based on weight) were in the form of settleable solids (Helz et al. 1975). The concentration of copper rapidly decreased downstream of the outfall so that 2–3 km from the outfall, the copper concentration had fallen to 7 ppb. The concentration of copper in sediment 2–3 km downstream from the outfall was about a factor of 10 higher than in uncontaminated areas (e.g., Rappahannock River). Based on their data and the results from other studies, Helz et al. (1975) estimated that approximately 200 metric tons of copper entered the Chesapeake Bay from the effluent discharged from waste treatment plants annually. Whitall et al. (2010) concluded that copper released from antifouling paint on boats was a likely source of copper measured in the Choptank river estuary, a tributary of the Chesapeake Bay.

Copper binds primarily to organic matter in estuarine sediment unless the sediment is low in organic matter content. Davies-Colley et al. (1984) determined copper's absorptivity to model phases in artificial seawater in order to estimate copper distributions between estuarine sedimentary phases and water. The model phases included hydrous iron and manganese oxides, clay, aluminosilicates, and organic matter. The binding affinities varied by over a factor of 10,000 and were in the following order: hydrous manganese oxide >organic matter >hydrous iron oxide >aluminosilicates >clay (montmorillonite). The partition coefficients at pH 7 for the more strongly binding phases (manganese oxide, iron oxide, and estuarine humic material), were 6,300, 1,300, and 2,500, respectively. The affinity increased somewhat with pH but did not vary appreciably when the salinity was reduced from 35 to 5%. Considering the typical compositional characteristics of estuarine sediment in terms of binding capacity, the results indicate that copper binds predominantly to organic matter (humic material) and iron oxides. Manganese oxide contributes only 1% to the binding because of its generally low concentration in sediment; the other phases are usually unimportant. These findings concur with results of selective extraction experiments (Badri and Aston 1983) and studies of the association of copper with humic material (Raspor et al. 1984).

Sediment and Soil. Most copper deposited on soil from the atmosphere, agricultural use, and solid waste and sludge disposal is retained in the upper 5–10 centimeters of soil in comparison to lower soil depths, except in sandy soils where the lability of bound copper is greater (Breslin 1999; Giusquiani et al. 1992; Hutchinson 1979; Luncan-Bouché et al. 1997; Levy et al. 1992; Perwak et al. 1980). Copper's movement in soil is determined by a host of physical and chemical interactions of copper with the soil components. In general, copper will adsorb to organic matter, carbonate minerals, clay minerals, or

hydrous iron and manganese oxides (EPA 1979; Fuhrer 1986; Janssen et al. 1997; Petruzzelli 1997; Tyler and McBride 1982). Sandy soils with low pH have the greatest potential for leaching. In a laboratory study, Luncan-Bouché et al. (1997) have shown that between 55 and 85% of copper bound to sand (with no other soil components added) is remobilized upon reduction of the pH from 9 to 4. In most temperate soils, the pH, organic matter, concentrations of metal oxyhydroxides, and ionic strength of the soil solutions are the key factors affecting adsorption (Elliot et al. 1986; Fuhrer 1986; Gerritse and Van Driel 1984; Janssen et al. 1997; Rieuwerts et al. 1998; Tyler and McBride 1982). The ionic strength and pH of the soil solution affect the surface charge of soils and thereby influence ionic interaction (Rieuwerts et al. 1998). Soil microorganisms also affect the absorption of copper in soils due to the uptake and assimilation of the metal by these microorganisms (Rieuwerts et al. 1998). However, it is not known how the rate of uptake and absorption capacity of the microorganisms for copper compares with the binding capacity and affinities of copper by organic matter in soils, such as humic and fulvic acids. When the amount of organic matter is low, the mineral content or Fe, Mn, and Al oxides become important in determining the adsorption of copper. Fuhrer (1986) reported that, in oxidized estuarine sediment, adsorption of copper is dominated both by amorphous iron oxide and humic material.

Copper binds strongly to soils with high organic content (14–34% organic matter, dry weight), and the distribution of copper in the soil solution is less affected by changes in pH (within the range of pH normally encountered in the environment) than other metals are (Gerritse and Van Driel 1984). In a laboratory study of competitive adsorption and leaching of metals in soil columns of widely different characteristics, copper eluted in a 0.01 M $CaCl_2$ leaching solution much more slowly and in much lower quantities than Zn, Cd, and Ni from a low-pH and a high-pH mineral soils and not at all from peat soil, which contained the greatest amount of organic matter (Tyler and McBride 1982). Elliot et al. (1986) investigated pH-dependent adsorption of the divalent transition metal cations cadmium, copper, lead, and zinc in two mineral soils (silty clay loam, 0.5 g/kg organic dry weight, and sandy clay, 1.6 g/kg organic) and two soils containing considerable organic matter (loamy sand, 20.5 g/kg organic, and silt loam, 42.5 g/kg organic). Adsorption increased with pH, and copper and lead were much more strongly retained than cadmium and zinc. Reduction in absorptivity after removal of the organic matter demonstrated the importance of organic matter in binding copper. In a study of clay soils, Wu et al. (1999) observed preferential copper binding to organic matter but found higher binding affinities to fine (<0.2 μm) clay fractions once the organic matter had been removed.

To determine the factors affecting copper leachability in soil, Hermann and Neumann-Mahlkau (1985) performed a study in the industrial Ruhr district of West Germany, which has a high groundwater table (10–80 cm from the surface) and a history of heavy metal pollution. Groundwater samples were taken

from six locations and two soil horizons, an upper oxidizing loam, and a lower reducing loam. Total copper concentrations were high in the upper soil horizons and low in the lower horizons. Copper showed a pronounced leachability only in the oxidizing environment. In the reducing environment, the mobility was low, possibly due to the formation of sulfides.

The mobility of copper from soils was also found to increase following the introduction of 10–100 mM sodium chloride or calcium magnesium acetate deicing salts into soil (Amrhein et al. 1992). The concentration of sodium chloride or calcium magnesium acetate used in the study approximate those in runoff water produced from the melting of snow along salted roadways.

For concentrations up to 2 mg of copper per liter of water, 25–75% of copper entering POTWs is removed in sludge, much of which is disposed of by spreading on land. Thus, it is useful to ascertain whether copper in sludge is apt to leach into soil. This did not appear to be the case: leachate collected from sludge-amended soil contained <12 ppb of copper (Perwak et al. 1980). Older studies found that small amounts of copper were found in leachate from soils treated with copper-containing sludge, and copper is typically confined to the upper 5-10 cm of soil (Breslin 1999; Davis et al. 1988; Giusquiani et al. 1992; Ritter and Eastburn 1978). In soils receiving long-term, heavy applications of sludge, high copper concentrations (471 mg/kg in comparison to 19.1 mg/kg in unamended control soils) were reported to depths of up to 25 cm (Richards et al. 1998). Brown et al. (1983) found that copper remained in the upper 12.7 cm of soil treated with sewage sludge for a year. The mobility of copper into soil from sludge was found to be determined mainly by the amount of soil organic carbon and soil surface area (Domergue and Védy 1992; Gao et al. 1997). In addition, soils amended by sludge with low metal content were found to have increased sorption of copper due to the increased binding capacity provided by the “low metal” organics in the sludge (Petruzzelli et al. 1994). From the results of other work, the major portion of the copper (40–74%) is expected to be associated with the organic Fe-Mn-oxide and carbonate fractions of most soils (Ma and Rao 1997).

Recent studies on the long-term effects of soil treated with organic amendments such as sludge, manure,

and compost, on copper availability have been published. Smolders et al. (2012) found that copper availability in soil treated long-term with organic amendments is lower than that in soil that has been spiked with Cu^{2+} salts because of its lower availability in the original matrix and due to aging reactions. Cagnarini et al. (2021) simulated long-term metal concentrations in soil treated with organic amendments in Switzerland. Copper concentrations have decreased over time and are projected to remain nearly constant or in decline through 2100 (Cagnarini et al. 2021). The model suggests that although

concentrations of copper in soil treated with sewage sludge are expected to decrease, historic inputs of sewage sludge would result in exceedances of the threshold concentration that would persist through 2100. Copper availability in soil to which stabilized sewage sludge or biosolids has been applied has also been more recently studied; concentrations of copper in biosolid treated clay, calcareous, and sandy soil were significantly higher than in control samples (Mahdy et al. 2007).

Other Media. The bioconcentration factor (BCF) of copper in fish obtained in field studies is 667 in marine fish and 50-200 in freshwater fish, suggesting a low potential for bioconcentration (Perwak et al. 1980). The BCF is higher for mollusks such as hard-shell clams and squid with BCFs of 30,000 and $2.1x10^7$, respectively (Perwak et al. 1980). This may present a major dietary source of copper that could be of concern for those individuals who regularly consume oysters, clams, or squid. Since mollusks are filter feeders and copper concentrations are higher in particulates than in water, this is to be expected (Perwak et al. 1980). For example, a study was conducted with white suckers and bullheads, both bottom-feeding fish, in two acidic Adirondack, New York, lakes (Heit and Klusek 1985). These lakes were known to have received elevated loadings of copper, but the suckers and bullhead had average copper levels of only 0.85 and 1.2 ppm (dry weight) in their muscle tissue. The biomagnification ratio (the concentration of copper in fish compared to that in their potential food sources on a wet weight/wet weight basis) was <l, indicating no biomagnification in the food chain. The copper content of muscle tissue of fish from copper contaminated lakes near Sudbury, Ontario, did not differ significantly from that of the same fish species in lakes far from this source (Bradley and Morris 1986). In a commercial catfish pond where copper was applied as an algaecide, only 0.01% of the copper applied was taken up by the fish (Liu et al. 2006). Similarly, the copper concentration in shrimps in a shrimp farm with high copper bioavailability did not differ from other shrimp populations (Lacerda et al. 2009).

No evidence of bioaccumulation was obtained from a study of various pollutants in the muscle and livers of 10 mammal species in Donana National Park in Spain (Hernandez et al. 1985). The park is impacted by organochlorine compounds and heavy metals emitted from anthropogenic activities that surround the park. For example, the Guadalquivir River that flows through the park first flows through a major mining region in addition to a large urban area and industrial areas, potentially carrying with it contaminants acquired from these sites. The animal species in the study were classified into three categories (herbivorous, omnivorous, and carnivorous) to ascertain if the pollutants were showing biomagnification in higher trophic levels of animals. No evidence of copper biomagnification in the food chain was observed. Likewise, in a study of a food web in a beech tree forest in Northern Germany, there was no evidence of biomagnification in tertiary consumers (e.g., vole, shrew, and mouse) compared to secondary consumers (e.g., earthworm, snail, beetle, and isopod) (Scharenberg and Ebeling 1996). A study of heavy

metals in cottontail rabbits on mined land treated with sewage sludge showed that, while the concentration of copper in surface soil was 130% higher than in a control area, the elevation was relatively little in foliar samples. No significant increase in copper was observed in rabbit muscle, femur, kidney, or liver. Apparently, copper was not bioaccumulating in the food chain of the rabbit (Dressler et al. 1986).

At the lowest levels of the food chain, there is little evidence of copper bioaccumulation. In a study of copper uptake in earthworms as a function of copper concentration (6–320 mg/kg dry weight) in sludge amended soils, a bioconcentration factor of <1 (0.67) was obtained (Neuhauser et al. 1995). In another example, a study of earthworms and soil from 20 diverse sites in Maryland, Pennsylvania, and Virginia, copper concentrations in earthworms showed a poor correlation with that in soil (Beyer and Cromartie 1987). These results are consistent with the results of another study that also showed no clear correlation between copper concentrations in earthworm tissues and two soils that were heavily contaminated with heavy metals (copper concentrations of 242 and 815 mg/kg dry weight) (Marinussen et al. 1997).

However, there is some evidence in one study for bioconcentration of copper at low copper concentrations in soil. Even though Scharenberg and Ebeling (1996) showed that there was no evidence for biomagnification of copper in a forest food web, their results did show that the total concentrations of copper in the secondary (18.3–192.0 mg/kg dry weight) and tertiary consumers (9.9–17.4 mg/kg dry weight) were higher than the concentrations of the metal in the dominant vegetation (5.3–10.9 mg/kg dry weight) and soil (1.8–5.8 mg/kg dry weight) in the ecosystem.

Diks and Allen (1983) added copper to four sediment/water systems and studied the distribution of copper among five geochemical phases, namely, absorbed/exchangeable, carbonate, easily reducible (Mn-oxides and amorphous Fe-oxides), organic, and moderately reducible (hydrous Fe-oxides). The investigators then attempted to correlate the concentration in each phase with the copper uptake by tubificid worms. Only copper extracted from the manganese oxide/easily reducible phase correlated with the copper content of worms at the 95% confidence level. This result suggests that the redox potential and pH in the gut of the worm is such that manganese oxide coatings are dissolved. The copper in the dissolved manganese oxide phase could be assumed to be soluble and available for uptake by other organisms.

5.4.2 Transformation and Degradation

Air. Data is available on the speciation of copper in airborne particulates. It is generally assumed that metals of anthropogenic origin, especially those from combustion sources, exist in the atmosphere as oxides because metallic species are readily attacked by atmospheric oxidants. As these oxides age,

sulfurization may occur, but only when SO_X gases are present in the atmosphere in sufficient amount. For example, in Arizona, atmospheric copper oxide levels near copper smelters were strongly correlated with co-emitted sulfur (Schroeder et al. 1987). Copper was primarily bound to organics and sulfides in dry deposition near a smelter in China, and dust from the smelter and in deposition samples showed sulfides and oxides (Liu et al. 2021). Copper has been observed bound to fine aerosol particles as the sulfate and nitrate (Osan et al. 2010). The form of copper in the coarse fraction could be used to trace its source to soil resuspension or brake pad wear erosion (Osan et al. 2010).

In fog water, Cu(II) is reduced to Cu(I) by sulfite, which becomes enhanced by the fact that sulfite is also a ligand of and binds to Cu(I) (Xue et al. 1991). Concentrations of Cu(I) in fog water ranged between 0.1 and 1 μM or, respectively, 4 and >90% of copper in the Cu(I) state. The reduction of Cu(II) to Cu(I) is pH dependent and occurs rapidly at pHs>6 (Xue et al. 1991).

Water. Free Cu^+ ion is unstable in aqueous solution, tending to disproportionate to Cu^{2+} and copper metal unless a stabilizing ligand is present (EPA 1979; Kust 1978). The only cuprous compounds stable in water are insoluble ones such as Cu_2S, CuCN, and CuF. Therefore, human exposures to copper will predominately be in the form of Cu(II). Copper in its Cu(II) state forms coordination compounds or complexes with both inorganic and organic ligands. Ammonium and chloride ions can form stable ligands with copper. Copper also forms stable complexes with organic ligands such as humic acids, binding to -NH_2 and -SH functional groups and, to a lesser extent, with -OH functional groups. Copper binding to humic and fulvic substances appears as both ionic binding and chelation. Natural waters contain varying amounts of inorganic and organic species. This affects the complexing and binding capacity of the water and the types of complexes formed. In seawater, organic matter is generally the most important complexing agent (Coale and Bruland 1988). In water, the presence of ligands may affect other physicochemical processes such as adsorption, precipitation, and oxidation-reduction (EPA 1979). More specific information on the transformation and degradation of copper in its cupric [Cu(II)] and cuprous [Cu(I)] states is given below.

At the pH values and carbonate concentrations characteristic of fresh surface waters, most dissolved Cu(II) exists as carbonate complexes rather than as free (hydrated) cupric ions (Stiff 1971).

Based on the results of a theoretical model, the major species of soluble copper found in freshwater, seawater, and a 50:50 combination of the freshwater and seawater over a pH range of 6.5–7.5 is Cu^{2+}, $Cu(HCO_3)^+$, and $Cu(OH)_2$ (Long and Angino 1977).

The concentration of dissolved copper depends on factors such as pH, the oxidation-reduction potential of the water, and the presence of competing cations (Ca^{2+}, Fe^{2+}, Mg^{2+}, etc.), anions (OH^-, S^{2-}, PO_4^{3-}, CO_3^{2-}), and soluble cupric-organic and -inorganic complexing agents. If the combination of a particular anion with copper forms an insoluble salt, precipitation of that salt will occur. The most significant precipitate formed in fresh surface waters is malachite ($Cu_2[OH]_2CO_3$) (Sylva 1976). Other important precipitates are $Cu(OH)_2$ (and ultimately CuO) and azurite ($Cu_3[OH]_2[CO_3]_2$). In anaerobic waters, Cu_2S, Cu_2O, and metallic copper forms and settles out (EPA 1979). The combined processes of complexation, adsorption, and precipitation control the level of free Cu(II) in water. The chemical conditions in most natural water are such that, even at relatively high copper concentrations, these processes will reduce the free Cu(II) ion concentration to extremely low values.

As a result of the previously described physico-chemical processes, copper in water may be dissolved or associated with colloidal or particulate matter. Copper in particulate form includes precipitates, insoluble organic complexes, and copper adsorbed to clay and other mineral solids. In a survey of nine rivers in the United Kingdom, 43–88% of the copper was in the particulate fraction (Stiff 1971). A study using suspended solids from the Flint River in Michigan found that the fraction of adsorbed copper increased sharply with pH, reaching a maximum at a pH of 5.5–7.5 (McIlroy et al. 1986).

The soluble fraction of copper in water is usually defined as that which will pass through a 0.45 µm filter. It includes free copper and soluble complexes as well as fine particulates and colloids. The soluble fraction may be divided according to the lability (e.g., the relative ability of the copper to dissociate from the bound form to the free ion) of the copper forms in the water. Categories range from the very labile metal (e.g., free metal ion, ion pairs, inorganic or organic complexes) to slowly or nonlabile metal (e.g., colloidally bound to inorganic colloidal phases of other metals such as $Fe(OH)_3$ or FeOOH, or bound to high molecular weight organic material) (Tan et al. 1988). For example, in a typical study, 18–70% of dissolved copper in river water was labile and 13–30% was slowly labile (Tan et al. 1988). Various techniques may be used to classify the lability of different fractions of soluble copper; these techniques include solvent extraction, ion-specific electrodes, ion exchange, ultrafiltration, electrochemical methods such as anodic stripping voltammetry, and gel filtration chromatography (Harrison and Bishop 1984). Newer technologies include hyphenated ICP-MS (Agilent Technologies 2012). The resulting classification depends on the specific procedure employed. Therefore, a comparison of the results of different researchers should be done in general terms.

The nature of copper's association with inorganic and organic ligands will vary depending on the pH, copper concentration, concentration of competing ligands, binding capacity of the ligands, and hardness

or salinity of the water (Breault et al. 1996; Cao et al. 1995; Gardner and Ravenscroft 1991; Giusti et al. 1993; Lores and Pennock 1998; Town and Filella 2000). In river water from the northwestern United States that had a relatively high pH (7.0–8.5) and alkalinity (24–219 ppm as $CaCO_3$), inorganic species like CO_3^{2-} and OH^- were the most important ligands at high copper concentrations (McCrady and Chapman 1979). However, other species such as organic compounds were important at low copper concentrations. On the other hand, copper in samples from surface water of lakes and rivers in southern Maine with a relatively low pH (4.6–6.3) and alkalinity (1–30 ppm as $CaCO_3$) was largely associated with organic matter (Giesy et al. 1978). The binding of copper to dissolved organics was found to be dependent on the specific organic chemical species (e.g., fulvic acid) and their concentrations in the surface water, the number of available binding sites per fulvic acid carbon, and the hardness of the water (Breault et al. 1996). Increasing water hardness results in deceased fulvic acid binding sites. This effect is due more to the depression of the solubility of high molecular weight fulvic acid in the presence of Ca and Mg ions than to competition of these ions with copper for fulvic acid binding sites. Changing pH from 8 to 6 resulted in a 7-fold increase in the binding constant for Cu(II) with humic acid (Cao et al. 1995).

The extent to which copper binds to inorganic and organic ligands can be altered by materials carried in runoff. For example, after a period of rain in southeastern New Hampshire, inorganic constituents contributed more to copper binding in lakes and rivers than did dissolved organic matter (Truitt and Weber 1981). A green precipitate, confirmed to be malachite ($Cu_2[OH]_2CO_3$), was formed in river water in Exeter, NH. The water had a high alkaline pH (7.4) with 43.5 mg/L $CaCO_3$ as a buffering agent that was higher than six other surface waters (e.g., three rivers, two reservoirs, a pond, and a swamp) with pH values of 5.7– 7.4 and 1.7–41 mg/L, respectively. A computer simulation of the copper species in water of a pond and water obtained from an artesian well that fed the pond predicted that 98% of the copper in the artesian well water would exist as the free copper ion (Cu^{+2}), whereas 88 and 63% of the copper in pond water would be bound to organics in the spring and fall, respectively (Giesy et al. 1983). These estimates were based on experimentally determined binding capacities of the organic matter in the two water sources and stability constants for the copper-organic matter complexes.

Seawater samples obtained in a transect of the uppermost Narragansett Bay in August 1980 were analyzed for dissolved, particulate, and organically bound copper to investigate the geochemistry of copper-organic complexes (Mills and Quinn 1984). Narragansett Bay is a partly mixed estuary in Massachusetts and Rhode Island that receives organic matter and metals from rivers, municipal and industrial effluents, and from runoff. The Fields Point waste treatment facility accounts for 90% of the copper input into the bay through the Providence River with dissolved copper representing 60% of the

total copper input. The concentrations of dissolved and organic copper ranged from 16.4 and 2.3 µg/kg in the Providence River to 0.23 and 0.12 µg/kg in Rhode Island Sound. Particulate copper concentrations in Narragansett Bay ranged from 0.06 to 2.42 µg/kg and generally comprised 40% of the total copper in the bay. Analysis of the data indicated that ~75% of the dissolved copper that enters the bay from the Providence River is removed within the bay.

Organic ligands can contain a variety of binding sites, and the strength of the resulting copper complexes will vary accordingly. Over 99.7% of the total dissolved copper in ocean surface water from the northeast Pacific was associated with organic ligands (Coale and Bruland 1988). The dominant organic complex, limited to surface water, was a strong ligand of biological origin. A second, weaker class of organic ligand was of geologic origin. An independent study showed that copper binds to humic material at a number of sites. The binding strength of the sites varied by two orders of magnitude (Giesy et al. 1986). The humic material in this study was derived from nine surface waters in the southeastern United States. Soluble copper in water discharged from a nuclear power station was primarily complexed with organic matter in the 1,000–100,000 molecular weight range (Harrison et al. 1980). Ten to 75% of the discharged copper was in particulate form.

The bioavailability of Cu(I) is difficult to access due to its thermodynamic instability in the environment (Xue et al. 1991). Cu(I) is a reactive reducing agent, and its concentrations in the environment is typically determined both by its reaction with oxygen and other oxidants in the aqueous environment to form Cu(II) and its rate of production through the reaction of Cu(II) with reducing agents (Sharma and Millero 1988). Investigators have shown the presence of Cu(I) in seawater, which is thought to occur through the reduction of Cu(II) to Cu(I) by photochemical processes (Moffett and Zika 1987; Xue et al. 1991). The detection of Cu(I) in seawater is likely the result of the stabilization of Cu(I) through complex formation with chloride ions. Cu(II)-organic complexes absorb radiation at wavelengths >290 nm and can undergo charge transfer reactions where the Cu(II) is reduced and a ligand is oxidized. Photochemically-generated reducing agents such as O^{2-} and H_2O_2 in the surface water of oceans and possibly other natural waters (e.g., lakes) may contribute to the reduction of Cu(II) to Cu(I) in these waters (Moffett and Zika 1987; Sharma and Millero 1988).

Cu(I) concentration is highest in the surface layer of seawater, and the hydrogen peroxide concentration increases in parallel to that of Cu(I) (Moffett and Zika 1987). In addition, the percentage of free Cu(I) is highest on the surface. Sharma and Millero (1988) measured the rate of Cu(I) oxidation in seawater as a function of pH, temperature, and salinity. The rate of reaction increased with pH and temperature and decreased with increasing ionic strength (or higher salinity) (Sharma and Millero 1988). The results

suggested that the rates are controlled by Mg^{2+}, Ca^{2+}, Cl^-, and HCO_3 through their involvement in complex formation and ligand exchange (Sharma and Millero 1988).

Sediment and Soil. The adsorption of copper to soil and sediment was discussed in Section 5.4.1 under transport and partitioning. It is important to understand the transport and fate of copper and its compounds in soils and sediments because these compartments tend to be large reservoirs of copper and could have an impact on human exposures. Copper concentrations in drinking water obtained from groundwater can be affected by the leaching of copper from soil. Reservoir sediments have been shown to be sources of copper in drinking water (Georgopoulos et al. 2001). Although much of the copper is bound to inorganic or organic matrices in soils and sediments, there is the potential for release of copper into pore water within soils and sediments depending on soil conditions and the forms of the copper present. There is evidence to suggest that copper binding in soil is correlated with pH, cation exchange capacity, the organic content of the soil, the presence of manganese and iron oxides, and even the presence of inorganic carbon such as carbonates (Petruzzelli 1997; Rieuwerts et al. 1998). At pH levels above 5, absorption of copper from pore water onto soil components becomes a significant process, whereas at pH levels below 5, copper largely remains in pore water and is, therefore, mobile in soil (Perwak et al. 1980). However, broad generalizations about the mobility of copper in soils are not possible since the situation will differ among different soil types and environmental conditions. More specific information on the lability (e.g., extractability) of copper from differing soils and conditions is given below.

There are several ways for determining the forms of copper in soil, the most common method being the measuring of the extractability of the copper with different solvents. Extractability is a function of the nature of the soil and the form of copper deposited in the soil. If a relatively labile form of copper is applied, binding to inorganic and organic ligands can occur, as well as other transformations. The capacity of soil to remove copper and the nature of the bound copper were evaluated by incubating 70 ppm of copper with 5 g samples of soil for 6 days (King 1988). Twenty-one samples of soils (10 mineral and 3 organic) from the southeastern United States were included in the study. Some soil samples were taken from the subsoil as well as the surface. The amount of adsorbed copper ranged from 36 to 100%, of which 13–100% was nonexchangeable when extracted with KCl. Removal of copper from solution was much higher with surface soils than with subsurface sandy soils; 95–100% of the copper was removed by five of the mineral surface soils and all three organic soils. The percentage of copper that was nonexchangeable was relatively high in all but some of the acid subsoils. While the fraction of exchangeable copper was not dependent on pH in surface soils, 96% of the variation in exchangeability was correlated with pH in subsoils. The soil/water partition coefficient for copper was >64 for mineral soils and >273 for organic soils. Of the 8 heavy metals in the study, only lead and antimony had higher

partition coefficients than copper. Most of the copper in Columbia River estuary sediment and soil was associated with inorganic carbon (e.g., carbonate), but not with the amount of extractable Fe or the organic carbon content of the sediment (Fuhrer 1986).

The amount of ammonium acetate- and DTPA-extractable copper in wetland soil/sediment resulting from atmospheric deposition from smelters in Sudbury, Ontario showed the same pattern as total copper, despite random variations in soil pH, redox potential, and organic carbon (Taylor and Crowder 1983). Therefore, in this case, soil characteristics were not the dominant factors determining extractability and availability, but rather the form of copper that was deposited. The median concentrations of total copper, ammonium acetate-extractable copper, and DTPA-extractable copper at 25 sample sites were 371, 49, and 98 ppm, respectively.

In another study of copper partitioning in nine different contaminated soils, sequential extractions were used to operationally define six soil fractions in decreasing order of copper availability: water soluble >exchangeable >carbonate >Fe-Mn oxide >organic >residual (Ma and Rao 1997). The results of this study showed that the distribution of copper in these six soil fractions differed depending on the total copper concentration in the soil. As the copper concentration increased above 240 mg/kg, between 69 and 74.4% of the total copper was found in the water soluble, carbonate, Fe-Mn oxide, and organic fractions. In relatively uncontaminated soils (<240 mg/kg copper), between 97.6 and 99.6% of the copper was found to be associated with the residual fraction.

In estuarine environments, anaerobic sediments are known to be the main reservoir of trace metals. Under anaerobic conditions, Cu(II) salts will reduce to Cu(I) salts. The precipitation of cupric sulfide and the formation of copper bisulfide and/or polysulfide complexes determine copper's behavior in these sediments (Davies-Colley et al. 1985). In the more common case where the free sulfide concentration is low due to the controlling coexistence of iron oxide and sulfide, anaerobic sediment acts as a sink for copper, that is, the copper is removed from water and held in the sediment as an insoluble cuprous sulfide. However, in the unusual situation where the free sulfide concentration is high, soluble cuprous sulfide complexes may form, and the copper concentration in sediment pore water may then be high.

In sediment, copper is generally associated with mineral matter or tightly bound to organic material (Kennish 1998). As is common when a metal is associated with organic matter, copper generally is associated with fine, as opposed to coarse, sediment. Badri and Aston (1984) studied the association of heavy metals in three estuarine sediments with different geochemical phases. The phases were identified by their extractability with different chemicals and termed easily or freely leachable and exchangeable;

oxidizable-organic (bound to organic matter); acid-reducible (Mn and Fe oxides and possibly carbonates); and resistant (lithogenic). In the three sediments, the nonlithogenic fraction accounted for ~14–18% of the total copper and the easily exchangeable component was 5% of the total copper. In addition, the compositional associations of copper in sediment samples taken from western Lake Ontario were analyzed employing a series of sequential extractions (Poulton et al. 1988). The mean (±standard deviation) percentages of copper in the various fractions were exchangeable, 0±0; carbonate salt, 0.1±0.3; iron or manganese oxide-bound, 0.2±0.3; organic-bound, 40±11; and residual, 60±8. Another study found that 10–20% of the copper in Lake Ontario sediment samples was bound to humic acids, with virtually all of the copper bound to organic matter (Nriagu and Coker 1980). The concentration of copper associated with humic acids was 21–40 times greater than in the sediment as a whole.

Other Media. Copper is an essential nutrient for plant growth and metabolism. Therefore, uptake of copper from soil by plants through the roots is a natural and necessary process, actively regulated by the plant (Clemens 2001). However, loss of biodiversity has been reported in environments contaminated with copper. Naveed et al. (2014) found that increasing copper pollution resulting from a former wood preservation plant had a negative impact on plant growth and species. Earthworms, bacteria, nematodes, and fungi showed a similar response to increasing copper concentrations. Results of this study showed that there was a 10% loss in soil biodiversity within a copper concentration range of 110 to 800 mg/kg (Naveed et al. 2014).

The uptake of copper into plants is dependent on the concentration and bioavailability of copper in soils. The bioavailability of copper is determined largely by the equilibrium between copper bound to soil components and copper in soil solution. As noted in the discussion of copper binding in soils, this is determined by copper concentrations in soil, soil type, soil components, pH, oxidation-reduction potential of the soil, concentrations of other cations and anions in the soil, etc. (Rieuwerts et al. 1998). Other factors involved root surface area, plant genotype, stage of plant development, weather conditions, interaction with other nutrients in the soil, and the water table (Gupta 1979). Using lime (calcium carbonate) to adjust soil pH is another factor that affects copper uptake. For example, liming acidic soils can increase copper uptake in hay, but decrease copper uptake in wheat (Gupta 1979). However, the effect of liming on increasing soil pH does not appear to be the overriding factor behind the changes in copper uptake by plants, even though there is evidence that the addition of lime to soil to increase the pH to 7 or 8 reduces copper bioavailability to some plants (Perwak 1980). This is evidenced by the fact that changes in pH (5.4–8.0) have little effect on copper concentrations in plant tissues (Gupta 1979).

It appears that microorganisms are able to transform copper and affect the copper bioavailable for plant uptake (Mulder and van Veen 1968). Hydrogen sulfide (H_2S) forming microorganisms may be involved in soil copper precipitation as nearly insoluble sulfide salts. Bacteria of the genera *Thiobacillus* and *Ferrobacillus* are able to oxidize *CuS* to *$CuSO_4$.* Johnson et al. (2017) carried out experiments to study the redox transformation of copper by acidophilic bacteria and found that oxidation and reduction of copper were mediated by acidophilic bacteria indirectly. Copper (I) accumulated in aerobic cultures of sulfur-grown *Acidithiobacillus* spp. More copper (I) was produced by *At. Caldus* than by the other species. Reduction of copper (II) by aerobic cultures of sulfur-grown *Acidithiobacillus* spp. Was more pronounced as culture pH declined. *Acidithiobacillus* grown anaerobically on hydrogen and *Acidiphilium cryptum* grown micro-aerobically on glucose only reduced copper (II) when iron (III) was included. Copper (I) was only oxidized by growing cultures of *Acidithiobacillus* spp. When iron (II) was included.

5.5 LEVELS IN THE ENVIRONMENT

Reliable evaluation of the potential for human exposure to copper depends, in part, on the reliability of supporting analytical data from environmental samples and biological specimens. Concentrations of copper in unpolluted atmospheres and in pristine surface waters are often so low as to be near the limits of current analytical methods. In reviewing data on copper levels monitored or estimated in the environment, it should also be noted that the amount of chemical identified analytically is not necessarily equivalent to the amount that is bioavailable.

Table 5-9 shows the limit of detections typically achieved by analytical analysis in environmental media. Presented in Table 5-10 is a summary of the range of concentrations detected in environmental media at NPL sites.

Table 5-10. Lowest Limit of Detection for Copper Based on Standards[a]

Media	Detection limit	Reference
Metal and nonmetal dust on MCE filters in workplace	0.07 µg/sample[b]	NIOSH 2020; Method 7302, Issue 1
Metal and nonmetal dust on PVC filters	0.08 µg/sample[b]	NIOSH 2020; Method 7304, Issue 1
Biological tissues (nail, liver, lungs, etc.)	6 µg/g[c]	NIOSH 2020; Method 8200, Issue 1
Water, wastewater, and solid wastes	5.4 mg/L[d]	EPA 1994a; Method 200.7
Drinking water	0.2 µg/L[e]	EPA 2003; Method 200.5
Ground waters, surface waters and drinking water	0.01-0.5 µg/L 0.2 mg/kg[f]	EPA 1994b; Method 200.8
Ground water, surface water, drinking water, storm runoff, industrial and domestic wastewater	0.7 µg/L[g]	EPA 1994c; Method 200.9

Table 5-10. Lowest Limit of Detection for Copper Based on Standards[a]

Media	Detection limit	Reference
Air	0.00001 µg/L[h]	EPA 2020b
Food	6.02 µg/kg[f]	FDA 2020
Serum	2.5 µg/dL[i]	CDC 2018

[a]Detection limits based on using appropriate preparation and analytics. These limits may not be possible in all situations.
[b]Inductively Coupled Argon Plasma, Atomic Emission Spectroscopy (ICP-AES)
[c]Inductively-Coupled Plasma Atomic Emission Spectroscopy (ICP-AES)
[d]Inductively-Coupled Plasma Atomic Emission Spectrometry (ICP-AES)
[e]Axially viewed inductively coupled plasma-atomic emission spectrometry (AVICP-AES)
[f]Inductively Coupled Plasma – Mass Spectrometry
[g]Graphite Furnace Atomic Absorption (GFAA)
[h]Inductively Coupled Plasma – Mass Spectrometry or X-ray Fluorescence
[i]Inductively coupled plasma dynamic reaction cell mass spectrometry (ICP-DRC-MS)

Table 5-11. Copper Levels in Water, Soil, and Air of National Priorities List (NPL) Sites

Medium	Median	Geometric mean	Geometric standard deviation	Number of quantitative measurements	NPL sites
Water (µg/L)	2.40×10^2	371	16.2	357	195
Soil (mg/kg)	4.10×10^2	431	17.4	462	245
Air (µg/m^3)	0.293	0.915	86.6	27	19

[a]Concentrations found in ATSDR site documents from 1981 to 2019 for 1,867 NPL sites (ATSDR 2019). Maximum concentrations were abstracted for types of environmental media for which exposure is likely. Pathways do not necessarily involve exposure or levels of concern.

5.5.1 Air

Human exposure to copper in air comes from both natural and anthropogenic sources. The concentrations of copper in air can be higher in the proximity of major sources such as smelters, mining operations, and combustion sources (e.g., power plants, incinerators, automobiles, etc.). Data from EPA's Air Quality System (AQS) for the years 2016 through 2019 are reported in Table 5-12. Most monitors are in California and a few others have been located in Michigan in varying years including three in 2016, six in 2017, two in 2018, and two in 2019. Based on these data, the general population is expected to be exposed to copper concentrations in air below 0.8 µg/m^3.

Table 5-12. Percentile Distribution of Annual Mean Copper (TSP) Concentrations (µg/m^3) Measured in Ambient Air at Locations Across the United States

Year	Number of U.S. locations	25th	50th	75th	95th	Maximum
2016	17	0.021	0.027	0.035	0.051	0.208
2017	19	0.020	0.030	0.057	0.289	0.792
2018	15	0.022	0.024	0.031	0.054	0.066
2019	15	0.013	0.018	0.020	0.045	0.047

TSP = Total suspended particles
Source: EPA 2020a

One recent study found that the mean concentration of copper in ambient air from 13 U.S. cities was 0.005 µg/m^3, and concentrations ranged from 0.002 to 0.006 µg/m^3 (Chen and Lippmann 2009). The results of several studies in which concentrations of copper in air were reported are described below and summarized in Table 5-13. It should be noted that older data may not be representative of current concentrations, given the reduction of ambient air pollution in the United States.

Table 5-13. Outdoor Air Monitoring Data for Copper

Location(s)	Geographic type	Date(s)	Mean concentration (ng/m^3)	Notes	Reference
United States	Urban	1977	207.5	4,648 samples[a]	EPA 1984
United States	Urban	1978	200.8	3,615 samples[a]	
United States	Urban	1979	259.3	2,507 samples[a]	
United States	Nonurban	1977	193.2	709 samples[a]	
United States	Nonurban	1978	265.7	458 samples[a]	
United States	Nonurban	1979	141.7	235 samples[a]	
Smokey Mountain National Park	Remote	1979	1.6	Above canopy, crustal enrichment factor 31	Davidson et al. 1985
Olympic National Park	Remote	1980	5.6	Crustal enrichment factor 76	
Camden, NJ	Urban	Summer 1981 & 82	16.0-18.0[b]		Lioy et al. 1987
Elizabeth, NJ	Urban	Summer 1981 & 82	21.0-29.0[b]		
Newark, NJ	Urban	Summer 1981 & 82	25.0-33.0[b]		
Ringwood, NJ	Rural	Summer 1981 & 82	13.0-63.0[b]		
Camden, NJ	Urban	Winter 1982 & 83	17.0-21.0[b]		
Elizabeth, NJ	Urban	Winter 1982 & 83	28.0-36.0[b]		
Newark, NJ	Urban	Winter 1982 & 83	21.0-27.0[b]		
Ringwood, NJ	Rural	Winter 1982 & 83	6.0-18.0[b]		

[a]Samples from National Survey

Table 5-13. Outdoor Air Monitoring Data for Copper

Location(s)	Geographic type	Date(s)	Mean concentration (ng/m^3)	Notes	Reference

[b]Concentrations reported by Lioy et al. (1987) are geometric means.

Davies and Bennett (1985) reported average atmospheric copper concentrations of 5–50 ng/m^3 in rural areas and 20–200 ng/m^3 in urban locations. Data from many urban locations in the United States show concentrations of copper associated with particulate matter ranging from 3 to 5,140 ng/m^3 (Schroeder et al. 1987). Remote and rural areas have concentrations of 0.029–12 and 3–280 ng/m^3, respectively (Schroeder et al. 1987). In remote areas such as national parks, differences in copper concentrations have been attributed to greater vegetative cover and higher moisture and larger amounts of exposed rock and soil (Davidson et al. 1985). Copper follows the same pattern as other heavy metals, in that increased copper levels are present in urban areas in winter and in rural areas in summer (Evans et al. 1984; Lioy et al. 1987.

Anderson et al. (1988) performed a study of the atmospheric aerosols collected at a site in Chandler, Arizona. Several major copper smelters are located ~120 km to the southeast, which were upwind of the sampling site during approximately 50% of the study period. The most abundant type of Cu-bearing particle, representing 74% of the total, was associated with sulfur. However, the analysis was not able to specify the form of sulfur present. Anderson et al. (1988) concluded that the smelters to the southeast were the probable source. Mine waste dump sites are another source of airborne copper (Mullins and Norman 1994). Particle size distribution and the concentration of copper in particle size ranges differ depending on the mine waste site (Mullins and Norman 1994).

Mean concentration ranges of copper in remote (any area of lowest copper concentration such as the Antarctic or Arctic) and rural (any site that represents a regional background that is not directly influenced by local anthropogenic emissions) precipitation ranges were 0.013–1.83 and 0.68–1.5 ppb, respectively, based on a weight per unit volume basis (Barrie et al. 1987). Although an earlier survey referred to by these investigators (Galloway et al. 1982) yielded much higher values of 0.060 and 5.4 ppb, these were ascribed to sample contamination. The mean concentration of copper in rain reported in an extensive study in southern Ontario, Canada was 1.57±0.36 ppb during 1982 (Chan et al. 1986). These concentrations showed little spatial variability. Concentration of copper in cloud water over Olympic Peninsula in Washington State has been measured at 1.7±1.6 µg/L (air equivalent mean concentration of 0.5 ng/m^3) (Vong et al. 1997). Copper concentrations in precipitation may be affected by proximity to

industry, but concentrations do not appear to be affected by proximity to automobile emissions. Elevated levels of copper in fog water have been observed 3 km downwind from a refuse incinerator in Switzerland (Johnson et al. 1987). The concentration of copper in rain samples taken within 2–15 km downwind of the Claremont, New Hampshire municipal waste incinerator was found to range from 0.11 to 2.12 µg/L with a mean concentration of 0.87 µg/L. (Feng et al. 2000). Cu(II) concentrations in fog water from the central valley of California ranged from 1.7 to 388 ppb (Miller et al. 1987). The source of the copper was not investigated. The highest values were recorded just as the fog was dissipating.

Copper deposition from automobile emissions, as measured by the concentration of copper in snow, did not vary significantly as a function of distance (15–150 meters) from an expressway in Montreal, Canada (Loranger et al. 1996).

Airborne concentrations of copper in the indoor atmosphere within homes located in Suffolk and Onondaga counties in New York average between 8 and 12 ng/m^3 (Koutrakis et al. 1992). The concentration was significantly affected by the use of kerosene heaters, which were found to emit copper into the indoor air at a rate of 15,630 ng/hour (Koutrakis et al. 1992).

5.5.2 Water

Copper is widely distributed in water since it is a naturally occurring element. The results of several studies in which concentrations of copper in water were reported are described below and summarized in Tables 5-14, 5-15, and 5-16. Data from older studies may have been analyzed with instrumentation with high detection limits, and samples were often contaminated during collection, treatment, and analysis.

Groundwater collected from wells from 2013 to 2016 by USGS for the National Water-Quality Assessment Project show that copper concentration ranges from 0.2 to 98.4 µg/L (USGS 2020b). Copper concentrations in drinking water can vary widely (≤5–10,200 ppb) and can exceed the action level of 1,300 ppb (1.3 mg/L) that is the regulatory Maximum Contaminant Level Goal (MCLG) for copper in drinking water (EPA 1991; EPA 2021, 40 CFR Part 141). Copper was found at concentrations greater than EPA's Treatment Technology Action Level of 1.3 mg/L in 0.06 percent of domestic wells sampled by USGS from 1991 to 2004 (USGS 2009b). An Action Level is the concentration of a contaminant in potable water, which if exceeded in ten percent of monitoring systems requires treatment for corrosion control and public notification (EPA 2018).

Table 5-14. Surface Water Monitoring Data for Copper

Location(s)	Type	Date(s)	Range (µg/L)[a]	Mean Concentration (µg/L)	Notes	Reference
United States	USGS survey stations	Not specified	Not reported	4.2	53,862 occurrences	Eckel and Jacob 1988
New Jersey	Representative sample	1977-1979	Maximum = 261.0	Not reported	1603 samples taken from 600 sites. Median = 3.0	Page 1981
East Arctic Ocean	Ocean water	1980	32-489 ng/kg	93 ± 38 ng/kg	26 locations 0.5–1 m depth. Mean concentration at depth was 400 ng/kg Unfiltered samples	Mart and Nurnberg 1984
Atlantic Ocean	Ocean water	Not specified	0.79-3.9 nM	Not reported	20 sites, 2 cruises, 0–1 m depth Unfiltered samples	Yeats 1988
Massachusetts	Pond water	April 1971-March 1972	<10–105	Not reported	Low in summer, high in winter	Kimball 1973
Canada	Lake water	November 1976-January 1977	1-8	2	Acid sensitive lakes	Reed and Henningson 1984
Lake Superior	Lake water	August-September 1991	629-834	756	3 samples Filtered samples	Nriagu et al. 1996
Lake Erie	Lake water	August 1993	703-1,061	870	9 samples Filtered samples	Nriagu et al. 1996
Lake Ontario	Lake water	May-June 1993; October 1993	540-1,098	830	14 samples Filtered samples	Nriagu et al. 1996
Indiana	Stream and pond water, near acidic mine drainage		32-1,200	736	12 samples taken from streams and ponds near abandoned Cerbat Mountain coal mines Filtered samples	Allen et al. 1996
Cerbat Mountains, northwestern Arizona	Surface water in a copper mining area	March 1995, September 1995	100-69,000	Not reported	Samples obtained from the Cerbat Mountains mining area; 15 surface water sites with 14 sites downstream from old tailings and adits. Median=1,200	Rosner 1998

[a] Range is µg/L unless otherwise stated
USGS = United States Geological Survey

Table 5-15. Copper Concentrations in Groundwater Water Monitoring Data

Location(s)	Type	Date(s)	Range (µg/L)	Median (µg/L)	Notes	Reference
New Jersey	Representative sample	1977-1979	Maximum = 2783.0	5.0	1,063 samples, 90th percentile 64.0 ppb, groundwater may or may not be used for drinking water	Page 1981
Denver, Colorado	Shallow monitoring well	1993	<1-14	2.0	30 monitoring wells, 22 with PVC casings and 8 with metal casings; samples obtained after purging well 20 minutes Filtered pesticide samples and unfiltered VOC samples	Bruce and McMahon 1996

Table 5-16. Copper Concentrations in Drinking Water Monitoring Data

Location(s)	Type	Date(s)	Range (µg/L)	Mean (µg/L)	Notes	Reference
Nova Scotia, four communities	Running tap water from private wells Standing tap water from private wells	NS NS	40-200 130-2,450	NR NR	53% of homes exceeded Canada's maximum permissible limit for copper (1.0 mg/L)	Maessen et al. 1985
New Bedford, Massachusetts	Running tap water from private wells	April 1987, 1992, 1993 July 1992	NR	230-560	24 sample areas included	Yannoni and Piorkowski 1995
Canada (National Survey)	Raw, treated, and distributed water	November 1976-January 1977	5.0-620	NR	Sampled raw, treated, and distributed water from 70 municipalities; median concentration was 20; noted differences based on source and type of water Filtered samples	Meranger et al. 1979

Table 5-16. Copper Concentrations in Drinking Water Monitoring Data

Location(s)	Type	Date(s)	Range (µg/L)	Mean (µg/L)	Notes	Reference
New Jersey	School drinking water	1991-1992	BD-10.2	NR	Sampled 2 water fountains in each of 50 schools. Median concentration ranged from 0.068 to 0.26 depending on time of day. Noted differences based on time of day and corrosivity of samples	Murphy 1993
Berlin, Germany	Running tap water from municipal water supply	June 1998-March 2001	0.009-4.2	0.436-0.561	2619 samples from 2944 households were tested.	Zietz et al. 2003a
Lower Saxony, Germany	Tap water from municipal water supply	January 1997-November 1999	>0.01-6.40	0.106-0.183	1619 stagnated water samples and 1660 random daytime samples.	Zietz et al. 2003b

BD = below detection; NR = not reported

Copper concentrations in drinking water vary widely as a result of variations in pH, hardness of the source water, and copper released from the water distribution system materials (Davies and Bennett 1985; Yannoni and Piorkowski 1995). A Canadian national survey of copper and other metals in drinking water was conducted from November 1976 to January 1977 (Meranger et al. 1979). Supplies from 70 municipalities representing 38% of the Canadian population were included in the survey, including 50 derived from river or lake water and 20 derived from groundwater. Unfiltered raw, treated, and distributed drinking waters were analyzed. Whether the water was derived from river, lake, or well water did not significantly affect the copper concentration in the raw water. Only in a few supplies did copper levels in raw water exceed 20 ppb and only one of these was derived from groundwater. The results in groundwater contrast with those of Page (1981) in New Jersey, in which over 100 wells contained copper levels in excess of 64 ppb. However, that study included groundwater that was a source of drinking water, in addition to groundwater that was not. The copper concentration in Canadian treated water was generally ~10 ppb (Meranger et al. 1979). In 20% of the samples, the copper level in distributed water was significantly higher than the treated water. The increase was greater in areas where the water was soft and corrosive, thus enhancing leaching of copper from the distribution system.

Elevated concentrations of copper in drinking water can result as a consequence of leaching processes

that occur in water distribution systems. Data from 208 U.S. households indicates that about a third of U.S. homes have drinking water containing more than 0.1 ppm copper (Brewer 2010). A study of 1,000 water samples from random households in Ohio found that ~30% contained copper levels >1 ppm (Strain et al. 1984). The highest copper level in the study was 18 ppm. In a study of private water wells in four communities in Nova Scotia, Maessen et al. (1985) found that the concentrations of copper increased in water that remained in the distribution system overnight, indicating that copper was mobilized from the distribution system. Whereas the level of copper in running water was generally very low, that in standing water was variable and exceeded 1.0 ppm in 53% of the homes. Similar results were reported for U.S. cities (Maessen et al. 1985; Schock and Neff 1988; Strain et al. 1984). In a study in Seattle, Washington, the mean copper concentrations in running and standing water were 0.16 and 0.45 ppm, respectively, and 24% of the standing water samples exceeded 1.0 ppm (Maessen et al. 1985). The difference in copper level between standing and flushed systems became evident at pH 7 and increased with decreasing pH (Strain et al. 1984). Copper levels in school drinking water were found to differ by 3-fold between first draw and 10-minute flush water samples, irrespective of the corrosiveness of the water (Murphy 1993). However, the concentration of copper in both first draw and 10-minute flush samples decreased by approximately 10-fold as the corrosiveness of the water decreased. Increasing pH in water distribution lines has been found to result in an overall decrease in metal concentrations. For example, increasing the pH of water from 7.5 to 8.5 in distribution lines decreased copper concentration by 50% (Yannoni and Piorkowski 1995).

In homes with copper piping, the mean concentration of copper in tap water has been shown to decline with the age of the home. In a sampling of tap water of 2,619 households in Berlin, Germany that are supplied with municipal drinking water, the mean concentration of copper decreased from 0.77 ppm in homes with stated ages of 0–<5 years to 0.23 ppm in homes with stated ages of 35–<40 years (Zietz et al. 2003a). In another study of 1,619 homes in Lower Saxony, Germany, the mean concentration of copper in first draw tap water decreased from 0.37 ppm in homes with stated ages of 0–<5 years to 0.05 ppm in homes with stated ages of 35–<45 years (Zietz et al. 2003b). These decreases of copper concentration with age were attributed to a buildup of a surface layer on the piping that reduced corrosion. However, in these same two studies, it was found that the concentration of copper in tap water began to increase with increasing age in homes with stated ages of >45 years. This increase in copper concentration was attributed to the increased probability of repair or partial placement (or unknown total replacement) of piping in these homes.

In a study of groundwaters and surface waters throughout New Jersey in which >1,000 wells and 600 surface sites were sampled, the median copper levels in groundwater and surface water were 5.0 and 3.0

ppb, respectively (Page 1981, Table 1). The respective 90th percentile and maximum levels were 64.0 and 2,783.0 ppb for groundwater and 9.0 and 261.0 ppb for surface water. The pattern of contamination in surface water correlates with light hydrocarbons, while that in groundwater correlates with heavy metals. This suggests that the sources of contamination of surface water and groundwater are different. The nature of the sites with elevated levels of copper was not indicated.

Copper levels in surface water range from 0.5–1,000 ppb, with a median of 10 ppb; seawater contains <1–5 ppb (Davies and Bennett 1985; Mart and Nurnberg 1984; Page 1981; Perwak et al. 1980; Yeats 1988). USGS collects data on the bottom material and surface water of streams for the Regional Stream Quality Assessment. Copper in bottom material ranged from 0.005 to 306 mg/kg (USGS 2018). The geometric mean, standard deviation, and median concentration of dissolved copper in surface water based on 53,862 occurrences in the Water Quality Portal are 4.2±2.71 and 4.0 ppb, respectively (WQP 2020). Higher concentrations tend to be found in New England, the western Gulf, and the lower Colorado River.

Copper concentrations were measured in surface water obtained from sampling sites in the Spearfish Creek, Whitewood Creek, and Bear Butte Creek watersheds. These watersheds are affected by water leaching from tailings and acid mine drainage from gold mining operations in the Black Hills of South Dakota. Copper concentrations of <0.24–28 µg/L were measured in surface water, whereas concentrations in sediments were much higher, ranging from 7.8 to 159 mg/kg (May et al. 2001).

In a survey of sources of copper in stormwater, measurements of copper concentrations in stormwater samples were taken from various urban locations in Birmingham, Alabama. Copper concentrations were generally low in filtered samples (dissolved copper), ranging between 1.4 and 20 µg/L; however, they were much higher in unfiltered samples (copper bound to particulate matter) with mean values (in µg/L) of 280 (street runoff), 135 (vehicle service areas), 116 (parking areas), 110 (roof areas), 81 (landscaped areas), 50 (urban creeks), and 43 (retention ponds) (Pitt et al. 1995).

As a result of improvements in controlling the quality of discharges from municipal and industrial wastewater treatment plants mandated in the Clean Water Act, copper concentrations have been declining in surface waters. For example, median copper concentrations in the Hudson River estuary have fallen 36– 56% between the mid-1970s and the mid-1990s (Sañudo-Wilhelmy and Gill 1999).

The copper concentration in some bodies of water evidently varies with season. In a study of a small pond in Massachusetts from April of 1971 to March 1972, the concentration of copper was found to vary, decreasing during the spring and early summer to lows of <10–30 ppm in early August and then increasing when the pond was under the cover of ice to maximum values of 80–105 ppb in late January

and early February (Kimball 1973). Similar seasonal variations were noted in the epilimnion of the offshore waters of the Great Lakes (Nriagu et al. 1996). In both examples, the cycling of copper concentrations is thought to be a response to biological need and copper uptake during the growing season and its subsequent release from seasonal die-off and decay of biota.

Copper concentrations in seawater usually are in the 1–5 ppb range (Perwak et al. 1980). Copper levels are overall lower in the Pacific Ocean versus the Atlantic Ocean and higher near the continental shelf than in the open ocean. Copper concentrations in surface water at a depth of 1 meter transected on a cruise from Nova Scotia to the Sargasso Sea ranged from 57.2 to 210 parts per trillion (ppt) (Yeats 1988). The mean value in surface water sampled at a depth of 1 meter of the eastern Arctic Ocean was 93 ppt (Mart and Nurnberg 1984). As noted in a review by Kennish (1998), concentrations of copper in estuarine and coastal waters in the United States were 0.3–3.8 and 0.1–2.5 ppb, respectively.

5.5.3 Sediment and Soil

Copper occurs naturally in the Earth's crust at a mean concentration of approximately 50 ppm (Henckens and Worrell 2020). Rauch and Graedel (2007) estimate that 9.9 $x10^{11}$ Gg (9.9$x10^{8}$ kg) of copper exists in the earth's crust. Several databases report copper levels in soil and sediment in the United States. The National Geochemical Database by USGS (2016) reports that copper occurs in soils at levels of 0.005 to 200,000 ppm in sediment at levels of 0.001 to 150,000 ppm. The median level of copper in soils and sediments reported to the National Geochemical Database is 30 ppm in soils and sediments (USGS 2016). The National Water Information System by USGS reports copper in soil at levels of 0.84 to 9.8 mg/kg (WQP 2020). Copper occurs in sediments at levels of 0.12 to 35,700 mg/kg (WQP 2020). EPA reports levels in soil of 0.58 to 334 mg/kg (WQP 2020). In 2007, USGS conducted a geochemical and mineralogical survey of soils of the conterminous United States. The mean concentration of copper calculated from the 4841 samples taken was 17.9 mg/kg, with values ranging from <0.5 to 996 mg/kg (USGS 2013).

Copper concentrations in soil may be much higher in the vicinity of a source of copper emissions, such as a mining operation or smelter. Concentrations in the top 5 cm of soil near the boundary of a secondary copper smelter were 2,480±585 ppm (Davies and Bennett 1985). Maximum wetland soil/sediment copper concentrations were 6,912 ppm in the immediate vicinity of a Sudbury, Ontario smelter but the concentration decreased logarithmically with increasing distance from the smelter (Taylor and Crowder 1983). The observation that the copper concentrations were highest in soils within 1–2 km from the smelter and decreased exponentially with increasing distance from the plant suggests that copper in the soil from the study area was primarily derived from particulate emissions from the smelter.

Copper and its compounds were reported at 929 of 1,867 hazardous waste sites on the NPL of highest priority sites for possible remedial action (ATSDR 2019). Since copper is commonly found in soil, technically it occurs at all sites. In past work, data analysis of metal concentrations measured in soil from hazardous waste sites taken from the 1980–1983 Contract Laboratory Program (CLP) Analytical Results Data Base (CARD) was conducted to ascertain whether elemental concentrations at hazardous waste sites were elevated above that which normally would be expected in soil of similar composition and derivation. Of the 1,307 samples in CARD, 10.5 and 7.3% (95 and 99% confidence intervals, respectively) had copper concentrations exceeding the number normally expected in soil (Eckel and Langley 1988).

In a study of 340 soil samples collected from diverse land-use situations, the average copper concentrations were 25 mg/kg in agricultural land, 50 mg/kg in suburban/residential land, 100 mg/kg in mixed industrial/residential land, and 175 ppm in industrial/inner urban areas (Haines 1984). From an analysis of the spatial distribution of the copper concentrations in soils where lowest copper soil concentrations are observed for rural (agricultural) soils and highest in soils obtained from industrialized urban areas, it was concluded that most of the contamination was a result of airborne deposition from industrial sources. Soil samples from urban gardens in New York had concentrations of copper ranging from 16.9 to 171 mg/kg, and an orchard had copper concentrations ranging from 19.7 to 62.8 mg/kg (Cai et al. 2016).

The concentrations of copper in soils and sediments were assessed as part of the National Water-Quality Assessment Program (Rice 1999). The median concentrations of copper at 541 sites throughout the conterminous United States ranged from 5 to 70 µg/g (dry weight). At nonurban indicator sites, the median concentrations ranged from 13 to 47 µg/g. The same study derived an average crustal abundance of copper of 60 µg/g (60 ppm).

Sediment is an important sink and reservoir for copper. In areas where there is no known input of copper obtained from anthropogenic sources, sediment generally contains <50 mg/kg copper. The level can reach several thousand ppm in polluted areas (Harrison and Bishop 1984). The mean copper level in surficial sediment of Penobscot Bay, Maine was 14.1 mg/kg (dry weight), while that in estuaries or bays in other New England locations ranged from 4.4 to 57.7 mg/kg (Larsen et al. 1983b). Levels reflect anthropogenic input as well as the mineral content of the regional bedrock. Copper levels in sediment from 24 sites along the New Jersey coast ranged from <1.0 to 202 mg/kg, with a mean value of 66 mg/kg (Renwick and Edenborn 1983). The texture of the sediment varied from 94% clay to 100% sand, and the copper level was correlated negatively with the percentage of sand in the sediment.

Surficial sediment in lakes in the Sudbury region of northeastern Ontario, where several smelters operate, decreased rapidly with increasing distance from the smelters (Bradley and Morris 1986). Three lakes, 10 km from the Sudbury smelters, contained copper concentrations in sediment approaching 2,000 mg/kg dry weight, over 100 times the concentration in a baseline lake 180 km away.

An analysis of the Coastal Sediment Database (COSED) showed that 73% of coastal waterways had copper concentrations below 42 µg/g; 25% had copper concentrations between 42 and 210 µg/g; and 2% were above 210 µg/g. These higher concentrations were associated with locations of high ship traffic, industrial activity, and relatively poor water flushing (Daskalakis and O'Connor 1995). In coastal areas receiving persistently high influxes of contaminants, high concentrations of copper (151 ppm) have been measured in sediments to depths of 54 cm (Bopp et al. 1993). Combined sewer outflows can also contribute significantly to the copper content of sediments. For example, mean (arithmetic) copper concentrations of 180, 208, 280, and 284 mg/kg were measured in sediment samples obtained near four sewer outflows in the lower Passaic River, New Jersey (Iannuzzi et al. 1997). In Jamaica Bay, New York, copper concentrations in sediments were 151–406 mg/kg, with a concentration of 151 ppm in sediment core samples obtained at a depth of 52–54 cm (Bopp et al. 1993). The highest concentrations were found in the middle depths (16–44 cm) ranging from 280 to 406 mg/kg during a period when untreated industrial effluents and sewage outflows entered the bay. However, copper concentrations in surface sediments (0–2 cm) were measured at 208 mg/kg. The decrease in copper concentration in the surface sediments suggests that efforts to reduce metal contaminants from sewage outflows have been making an impact on the copper concentrations in receiving waters and their sediments.

5.5.4 Other Media

In addition to the ingestion of drinking water, the consumption of food is the other primary route for copper intake in the general population. Copper is an essential nutrient present in many plant and animal foods and available as a dietary supplement. The Recommended Dietary Allowance (RDA) and Tolerable Upper Intake Level (UL) by life stage group are presented in Table 5-17. Copper intake per day based on NHANES data is provided in Table 5-18. Voluntary food fortification in the United States increases the probability of consuming copper and is associated with greater risk of exceeding the UL for children (Sacco et al. 2013).

Table 5-17. Dietary Reference Intakes for Copper

Life stage group[a]	RDA (µg/day)	UL (µg/day)
1-3 years	340	1,000
4-8 years	440	3,000
9-13 years	700	5,000
14-18 years	890	8,000
19 years and over	900	10,000
Pregnant females, 18 years and younger	1,000	8,000
Pregnant females, 19-50 years	1,000	10,000
Lactating females, 18 years and younger	1,300	8,000
Lactating females, 19-50 years	1,300	10,000

Source: Institute of Medicine 2006
RDA = Recommended Dietary Allowance, UL = Upper Intake Level
[a]RDAs are not estimated for ages 0 to 12 months. Adequate intake at this life stage is 220 µg/day.

Table 5-18. Mean Amount of Copper Consumed per Individual by Gender and Age

Gender and age (years)	Amount consumed (mg)	Standard error
Males		
2-5	0.8	0.02
6-11	0.9	0.05
12-19	1.1	0.03
20-29	1.2	0.05
30-39	1.4	0.06
40-49	1.4	0.07
50-59	1.5	0.07
60-69	1.3	0.05
70 and over	1.3	0.05
Females		
2-5	0.7	0.02
6-11	0.9	0.03
12-19	0.9	0.04
20-29	1.1	0.04
30-39	1.1	0.04
40-49	1.0	0.03
50-59	1.1	0.06
60-69	1.2	0.05
70 and over	1.1	0.03

Source: USDA 2020

The FDA Total Diet Survey provides copper concentration in various foods, examples of which are given in Table 5-19 (FDA 2017). The copper content in baby food is given in Table 5-20. The highest concentrations of dietary copper were found in liver; in some oat and bran cereals; in some legumes and nuts; and in chocolate syrup, candy, and some desserts. Coleman et al. (1992) reported copper concentrations in the edible tissues of livestock and poultry with the highest mean concentrations (mg/kg) found in liver (cow 3.7; lamb 89.8; chicken 4.60; turkey 7.14), followed by kidney (cow 8.15; lamb 5.39;

chicken 3.07; turkey 3.68), and muscle (cow 1.41; lamb 1.47; chicken 0.67; turkey 0.83) (Coleman et al. 1992).

Table 5-19. Copper Content of Selected Foods (mg/kg)

Food description	Mean±SD	Food description	Mean±SD
Liver (beef/calf), pan-cooked with oil	135.00±40.78	Pear, canned in light syrup	0.55±0.22
Sunflower seeds (shelled), roasted, salted	19.23±0.40	Pepper, sweet, green, raw	0.55±0.28
Walnuts, shelled	11.70±0.00	Beef with vegetables in sauce, from Chinese carry-out	0.54±0.24
Peanut butter, creamy	4.94±0.38	Cornbread, homemade	0.54±0.03
Peanuts, dry roasted, salted	4.82±0.16	Meatloaf, beef, homemade	0.54±0.08
Raisin bran cereal	4.32±0.31	Pie, pumpkin, fresh/frozen	0.54±0.04
Syrup, chocolate	3.82±0.26	Salmon, steaks/fillets, baked	0.53±0.12
Candy bar, milk chocolate, plain	3.67±0.33	Cream of wheat (farina), enriched, cooked	0.52±0.17
Shredded wheat cereal	3.66±0.38	Tomato, raw	0.52±0.22
Potato chips	3.57±0.35	Chicken potpie, frozen, heated	0.51±0.03
Oat ring cereal	3.55±0.23	Corn flakes cereal	0.51±0.04
Brownie	3.43±0.17	Frankfurter (beef/pork), boiled	0.51±0.08
Raisins	3.30±0.35	Tomato juice, bottled	0.51±0.06
Pinto beans, dry, boiled	3.18±0.39	Chicken breast, fried, fast-food (with skin)	0.50±0.07
Avocado, raw	2.96±0.59	Pineapple, canned in juice	0.50±0.05
Granola with raisins	2.90±0.38	Chicken nuggets, fast-food	0.49±0.10
Bread, whole wheat	2.77±0.02	Pie, apple, fresh/frozen	0.48±0.07
White beans, dry, boiled	2.71±0.58	Pineapple juice, frozen concentrate, reconstituted	0.48±0.05
Chocolate chip cookies	2.70±0.70	Collards, fresh/frozen, boiled	0.47±0.06
Bread, multigrain	2.64±0.16	Potatoes, mashed, prepared from fresh	0.47±0.23
Granola bar, with raisins	2.47±0.85	Orange juice, frozen concentrate, reconstituted	0.46±0.06
Cake, chocolate with icing	2.38±0.17	Blueberries, raw	0.45±0.00
Mushrooms, raw	2.34±0.50	Tuna, canned in water, drained	0.45±0.05
Sweet potato, baked, peel removed	2.31±0.00	Beets, canned	0.44±0.12
Candy bar, chocolate, nougat, and nuts	2.25±0.11	Brussels sprouts, fresh/frozen, boiled	0.43±0.04
Popcorn, microwave, butter-flavored	2.23±0.21	Fruit cocktail, canned in light syrup	0.43±0.02
Pork and beans, canned	2.10±0.14	Watermelon, raw/frozen	0.43±0.33
Lima beans, immature, frozen, boiled	2.08±0.20	Orange (navel/Valencia), raw	0.42±0.07
Sandwich cookies with creme filling	2.06±0.42	Orange juice, bottled/carton	0.42±0.02
Ice cream, chocolate	1.85±0.00	Strawberries, raw/frozen	0.40±0.13
Refried beans, canned	1.85±0.13	Turkey breast, oven-roasted	0.40±0.07
Crisped rice cereal	1.82±0.14	Peach, canned in light/medium syrup	0.39±0.03
Crackers, graham	1.79±0.20	Carrot, fresh, peeled, boiled	0.36±0.09
Meal replacement, liquid RTD, any flavor	1.70±0.62	Green beans, canned	0.36±0.08
Pretzels, hard, salted	1.66±0.15	Onion, mature, raw	0.36±0.06
Black olives	1.63±0.37	Soup, tomato, canned, condensed, prepared with water	0.36±0.03
Tomato catsup	1.55±0.17	Chicken breast, oven-roasted (skin removed)	0.35±0.05
Rice, brown, cooked	1.52±0.00	Eggplant, fresh, peeled, boiled	0.34±0.25
Chili con carne with beans, canned	1.51±0.03	Applesauce, bottled	0.33±0.06
Lamb chop, pan-cooked with oil	1.51±0.10	Bologna (beef/pork)	0.33±0.02
Bagel, plain, toasted	1.48±0.14	Cantaloupe, raw/frozen	0.33±0.22
Bread, rye	1.46±0.29	Cheese, Swiss, natural	0.33±0.05
French fries, fast-food	1.43±0.33	Corn, fresh/frozen, boiled	0.33±0.24
Shrimp, boiled	1.40±0.56	Grapefruit, raw	0.33±0.04
Crackers, saltine	1.38±0.14	Carrot, baby, raw	0.32±0.23
English muffin, plain, toasted	1.35±0.14	Apricots, canned in heavy/light syrup	0.30±0.06

Table 5-19. Copper Content of Selected Foods (mg/kg)

Food description	Mean±SD	Food description	Mean±SD
Noodles, egg, enriched, boiled	1.34±0.29	Soup, vegetable beef, canned, condensed, prepared with water	0.30±0.06
Soup, bean with bacon/pork, canned, condensed, prepared with water	1.28±0.02	Broccoli, fresh/frozen, boiled	0.29±0.22
Spaghetti, enriched, boiled	1.25±0.34	Dill cucumber pickles	0.27±0.03
Bread, white, enriched	1.23±0.16	Grapefruit juice, bottled	0.27±0.06
Beef steak, loin/sirloin, broiled	1.18±0.46	Cod, baked	0.26±0.00
Pineapple, raw/frozen	1.16±0.00	Sweet & sour sauce	0.26±0.23
Spaghetti with meat sauce, homemade	1.16±0.10	Cheese, cheddar, natural (sharp/mild)	0.25±0.18
Bread, white roll/bun (hamburger/hotdog)	1.15±0.00	Lettuce, leaf, raw	0.23±0.46
Beef stroganoff with noodles, homemade	1.14±0.25	Milk, chocolate, low-fat, fluid	0.21±0.14
Fruit-flavored cereal, presweetened	1.13±0.20	Grape juice, frozen concentrate, reconstituted	0.20±0.03
Crackers, butter-type	1.12±0.11	Prune juice, bottled	0.20±0.01
Burrito with beef, beans, and cheese, from Mexican carry-out	1.11±0.21	Cucumber, peeled, raw	0.16±0.18
Pizza, cheese, fast-food	1.11±0.00	Cake, yellow with icing	0.15±0.17
Quarter-pound hamburger on bun, fast-food	1.09±0.04	Cream substitute, non-dairy, liquid/frozen	0.13±0.23
Tortilla, flour	1.08±0.11	Wine, dry table, red/white	0.12±0.04
Turkey, ground, pan-cooked	1.08±0.00	Cheese, American, processed	0.11±0.20
Peas, green, fresh/frozen, boiled	1.05±0.19	Corn, canned	0.11±0.18
Pizza, cheese and pepperoni, regular crust, from pizza carry-out	1.03±0.05	Apple juice, bottled	0.10±0.03
Corn/tortilla chips	1.02±0.10	Sour cream dip, any flavor	0.10±0.17
Fried rice, meatless, from Chinese carry-out	1.02±0.18	Turnip, fresh/frozen, boiled	0.10±0.17
Quarter-pound cheeseburger on bun, fast-food	1.02±0.12	Catfish, pan-cooked with oil	0.09±0.18
Doughnut, cake-type, any flavor	1.00±0.28	Clam chowder, New England, canned, condensed, prepared with whole milk	0.09±0.15
Salami, luncheon-meat type (not hard)	0.96±0.12	Cottage cheese, creamed, low-fat (2% milk fat)	0.09±0.15
Asparagus, fresh/frozen, boiled	0.93±0.15	Luncheon meat (chicken/turkey)	0.09±0.16
Pork bacon, oven-cooked	0.93±0.22	Sorbet, fruit-flavored	0.09±0.15
Tortilla, corn	0.92±0.00	Soup, chicken noodle, canned, condensed, prepared with water	0.09±0.15
Potato, baked (with peel)	0.89±0.15	Lettuce, iceberg, raw	0.08±0.15
Banana, raw	0.87±0.15	Cabbage, fresh, boiled	0.07±0.14
Beef roast, chuck, oven-roasted	0.87±0.09	Fruit juice blend (100% juice), canned/bottled	0.05±0.05
Biscuits, refrigerated-type, baked	0.86±0.01	Cranberry juice cocktail, canned/bottled	0.03±0.05
Rice, white, enriched, cooked	0.85±0.15	Milk, low-fat (2%), fluid	0.03±0.04
Chicken leg, fried, fast-food (with skin)	0.84±0.12	Lemonade, frozen concentrate, reconstituted	0.02±0.04
Pork sausage (link/patty), oven-cooked	0.84±0.05	Tea, from tea bag	0.02±0.03
Sweet potatoes, canned	0.84±0.17	Milk, skim, fluid	0.01±0.03
Iced cinnamon roll	0.83±0.08	Milk, whole, fluid	0.01±0.03
Lasagna with meat, frozen, heated	0.83±0.07	Apple (red), raw (with peel)	0.00±0.00
Tomato sauce, plain, bottled	0.83±0.05	Beer	0.00±0.00
Fish sticks or patty, frozen, oven-cooked	0.82±0.19	Bottled drinking water (mineral/spring), not carbonated or flavored	0.00±0.00
Salami, dry/hard	0.82±0.00	Brown gravy, canned or bottled	0.00±0.00
Mustard, yellow, plain	0.81±0.06	Butter, regular (not low-fat), salted	0.00±0.00
Egg, cheese, and ham on English muffin, fast-food	0.80±0.13	Candy, hard, any flavor	0.00±0.00
Oatmeal, plain, cooked	0.77±0.20	Carbonated beverage, cola, low-calorie	0.00±0.00

Table 5-19. Copper Content of Selected Foods (mg/kg)

Food description	Mean±SD	Food description	Mean±SD
Peach, raw/frozen	0.77±0.18	Carbonated beverage, cola, regular	0.00±0.00
Tomato salsa, bottled	0.77±0.06	Carbonated beverage, fruit-flavored, regular	0.00±0.00
Chicken filet (broiled) sandwich on bun, fast-food	0.75±0.13	Cauliflower, fresh/frozen, boiled	0.00±0.00
Chicken leg, fried, fast-food (with skin)	0.75±0.00	Celery, raw	0.00±0.00
Macaroni salad, from grocery/deli	0.74±0.15	Cheese, Monterey jack	0.00±0.00
Biscuits, fast-food	0.73±0.00	Cheese, mozzarella	0.00±0.00
Potato salad, mayonnaise-type, from grocery/deli	0.71±0.27	Coffee, decaffeinated, from ground	0.00±0.00
Potato, boiled (without peel)	0.71±0.26	Coffee, from ground	0.00±0.00
Tuna noodle casserole, homemade	0.71±0.09	Coleslaw, mayonnaise-type, from grocery/deli	0.00±0.00
Chicken thigh, oven-roasted (skin removed)	0.70±0.15	Corn/hominy grits, enriched, cooked	0.00±0.00
Fish sandwich on bun, fast-food	0.69±0.06	Cream cheese	0.00±0.00
Okra, fresh/frozen, boiled	0.69±0.03	Cream, half & half	0.00±0.00
Taco/tostada with beef and cheese, from Mexican carry-out	0.69±0.08	Fruit drink (10% juice), canned or bottled	0.00±0.00
Pancakes, frozen, heated	0.68±0.05	Fruit drink, from powder	0.00±0.00
Pear, raw (with peel)	0.68±0.07	Gelatin dessert, any flavor	0.00±0.00
Breakfast tart/toaster pastry	0.67±0.08	Honey	0.00±0.00
Soup, Oriental noodles (ramen noodles), prepared with water	0.67±0.04	Ice cream, light, vanilla	0.00±0.00
Squash, winter (Hubbard or acorn), fresh/frozen, boiled	0.67±0.15	Ice cream, regular (not low-fat), vanilla	0.00±0.00
Beef, ground, regular, pan-cooked	0.66±0.09	Jelly, any flavor	0.00±0.00
Pork roast, loin, oven-roasted	0.64±0.02	Margarine, regular (not low-fat), salted	0.00±0.00
Eggs, boiled	0.63±0.18	Mayonnaise, regular, bottled	0.00±0.00
Eggs, scrambled with oil	0.63±0.10	Milk shake, vanilla, fast-food	0.00±0.00
Pork chop, pan-cooked with oil	0.62±0.06	Olive oil	0.00±0.00
Sugar cookies	0.62±0.06	Popsicle, fruit-flavored	0.00±0.00
Summer squash, fresh/frozen, boiled	0.62±0.23	Pudding, ready-to-eat, flavor other than chocolate	0.00±0.00
Grapes (red/green), raw	0.60±0.13	Salad dressing, creamy/buttermilk type, low-calorie	0.00±0.00
Green beans, fresh/frozen, boiled	0.60±0.18	Salad dressing, creamy/buttermilk type, regular	0.00±0.00
Spinach, fresh/frozen, boiled	0.60±0.14	Salad dressing, Italian, regular	0.00±0.00
Ham, cured (not canned), baked	0.59±0.11	Sour cream	0.00±0.00
Milk shake, chocolate, fast-food	0.59±0.35	Sugar, white, granulated	0.00±0.00
Mixed vegetables, frozen, boiled	0.58±0.12	Syrup, pancake	0.00±0.00
Chicken with vegetables in sauce, from Chinese carry-out	0.57±0.13	Tea, decaffeinated, from tea bag	0.00±0.00
Muffin, blueberry	0.56±0.12	Tilapia, baked	0.00±0.00
Luncheon meat, ham	0.55±0.07	Vegetable oil	0.00±0.00
Macaroni and cheese, prepared from box mix	0.55±0.15	Yogurt, frozen, vanilla	0.00±0.00
Pear, canned in light syrup	0.55±0.22	Yogurt, low-fat, fruit-flavored	0.00±0.00

SD = standard deviation
Source : FDA 2017

Table 5-20. Copper Content of Selected Baby Foods (mg/kg)

Food description	Mean	SD
Teething biscuits	1.60	0.53
Sweet potatoes	1.24	0.22
Arrowroot cookies	1.08	0.08
Cereal, mixed, dry, prepared with water	1.01	0.18
Cereal, oatmeal with fruit, prepared with water	0.96	0.10
Pears	0.95	0.08
Peaches	0.92	0.14
Turkey and rice	0.92	0.22
Cereal, oatmeal, dry, prepared with water	0.91	0.21
Mixed vegetables	0.90	0.10
Peas	0.89	0.07
Infant formula, soy-based, RTF	0.86	0.06
Bananas	0.84	0.22
Macaroni and cheese with vegetables	0.70	0.27
Chicken noodle dinner	0.67	0.09
Pears and pineapple	0.67	0.08
Plums/prunes with apples or pears	0.65	0.18
Apricots with mixed fruit	0.64	0.20
Carrots	0.64	0.17
Macaroni, tomato, and beef	0.64	0.08
Chicken with rice	0.61	0.06
Vegetables and turkey	0.61	0.16
Apples with fruit other than berries	0.57	0.14
Green beans	0.57	0.05
Infant formula, milk-based, iron fortified, rtf	0.56	0.07
Vegetables and beef	0.56	0.10
Squash	0.55	0.09
Fruit yogurt dessert	0.53	0.23
Vegetables and chicken	0.51	0.14
Cereal, rice, dry, prepared with water	0.46	0.04
Beef and broth/gravy	0.39	0.10
Applesauce	0.36	0.06
Juice, pear	0.34	0.04
Chicken and broth/gravy	0.33	0.02
Apples with berries	0.26	0.23
Turkey and broth/gravy	0.15	0.26
Juice, apple	0.11	0.03
Juice, grape	0.02	0.04

SD = standard deviation
Source : FDA 2017

The contribution of food groups to copper intake varies depending on the age group (Pennington and Schoen 1996). For example, animal flesh only contributes to 18% of the copper intake for a 2-year-old child but contributes to 38% of the copper intake for a 60–65-year-old male.

Wu et al. (2018) conducted a review of the literature to determine nutrient composition in human milk in the United States and Canada from 1980 to 2017. Average copper levels ranged from 0.02–0.08 µg per 100 g of human milk in women one to six months postpartum and from 0.017–0.02 µg per 100 g of human milk in women seven to twelve months postpartum.

High concentrations of copper have been measured in shellfish and crustacean species such as shrimp and prawns which use a copper-containing protein, hemocyanin, as an oxygen-transport molecule (Olmedo et al. 2013; Venugopal and Gopakumar 2017). Median copper concentrations ranged from 0 mg/kg wet weight in canned frigate to 6.865 mg/kg wet weight in frozen prawn (Olmedo et al. 2013). The calculated intake of copper from fish and shellfish is 0.117 mg/day, which is not expected to pose a risk to the average consumer (Olmedo et al. 2013). Shellfish provide between 7 to 378% of percent daily values of copper, with the highest contributions from oysters, squid, and lobster (Venugopal and Gopakumar 2017). The concentrations of copper in the soft tissue in mussels and oysters collected as part of the U.S. Mussel Watch Program in 1976–1978 were 4–10 ppm (dry weight) for mussels and 25–600 ppm for oysters (Goldberg 1986). Copper concentrations in mussels collected from 11 sites near Monterey Bay, California were 4.63–8.93 ppm (dry weight) (Martin and Castle 1984). Perwak et al. (1980) reported similar results for mussels (3.9–8.5 ppm) and for clams (8.4–171 ppm). Recent measurements of copper concentrations in zebra and quagga mussels taken from Lakes Erie and Ontario in 1997 ranged from 21 to 41 ppm (dry weight) (Rutzke et al. 2000). In the National Oceanic and Atmospheric Administration (NOAA) Mussel Watch Project, copper concentrations were quantified in mollusks (*M. edulis*, *M. californianus*, *C. virginica*, and *Ostrea equestris*) from 113 sites around the United States in 1993 and compared to copper concentrations measured in mollusks taken from the same site in the EPA2 Mussel Watch Program, 1976–1978 (Lauenstein and Daskalakis 1998). The results of the comparison indicate that the decreasing and increasing trends in copper concentrations in mollusks were approximately equal among the sites, except in California, where increasing trends were noted at five sites.

As a part of the National Contaminant Biomonitoring Program of the U.S. Fish and Wildlife Service, eight species of freshwater fish were collected at 112 stations in the United States in 1978–1979 and 1980–1981 (Lowe et al. 1985). The geometric mean concentrations of copper in ppm (wet weight, whole

fish) for these two periods were 0.86 and 0.68, respectively; the 85th percentiles were 1.14 and 0.90, respectively, and the ranges were 0.29–38.75 and 0.25–24.10, respectively. The highest concentration, 38.75 and 24.10 ppm, during both collecting periods was in white perch from the Susquehanna River; the second highest concentration, 19.3 ppm, was found in white perch from the Delaware River near Trenton, New Jersey. Copper concentrations in common carp and white catfish collected from the same station at the same time were 0.76 and 1.35 ppm, respectively.

Copper residues in muscle of 268 fish specimens from 17 species were analyzed over a 5-year period in several surface water systems in eastern Tennessee (Blevins and Pancorbo 1986). The mean residue levels in the muscle of different species of fish from nine stations ranged from 0.12–0.86 ppm (wet weight). Maximum levels ranged from 0.14 to 2.2 ppm.

Concentrations of copper in three species of fish living in storm treatment ponds have been compared to copper concentrations in controls collected from surrounding surface waters near Orlando, Florida (Campbell 1994). In redear sunfish and bluegill sunfish collected from stormwater ponds, the mean whole body copper concentrations were 6.37 and 2.08 mg/kg wet weight, respectively, and were significantly higher than the mean concentrations of copper, 0.879 and 1.07 mg/kg wet weight, respectively, measured in controls collected in natural lakes or ponds. However, in largemouth bass, the mean copper concentrations in fish collected from stormwater ponds and controls did not significantly differ, with values of 3.81 and 4.71 mg/kg wet weight, respectively.

Respective mean and median copper concentrations of 127 samples of finfish from Chesapeake Bay and its tributaries were 1.66 and 0.36 ppm in 1978, and 1.85 and 0.61 ppm in 1979 (Eisenberg and Topping 1986). In striped bass taken from Turkey Point in the bay, copper levels were below the detection limit of the study (<0.1 µg/g) in muscle, but were higher in liver tissue ranging from 0.86 to 23.5 µg/g. In gonad tissue obtained from tissue from a different site on the bay, there was also an increase in the mean copper concentration in this tissue (4.25 µg/g) as compared to muscle (0.76 µg/g). The copper content of muscle tissue of several species of fish collected from metal-contaminated lakes near Sudbury, Ontario, ranged from 0.5 to 1.4 ppm (dry weight). No major pattern in variation was evident among species or among the study lakes (Bradley and Morris 1986). The copper concentration in the livers ranged from 5 to 185 ppm (dry weight) and differed significantly among species and among lakes. Unlike muscle tissue, liver tissue is a good indicator of copper availability, although the data suggest that there are other factor(s) that influence the availability and bioaccumulation of copper in these fish.

The copper concentrations in the liver of lake trout and grayling taken from four freshwater lakes in Alaska did not correlate well with the concentrations of copper in the sediments of these lakes (Allen-Gil et al. 1997). Lake trout were found to have significantly higher burdens ($p<0.05$) of copper in their livers than grayling, and the concentrations of copper in the livers of trout varied considerably depending on the lake from which they were collected. The species and site differences in copper concentrations in fish livers have been attributed to differences in diet (grayling consume mainly insects, whereas trout consume a mix of snails, insects, and small fish) and time spent at various depths of the water column.

Although the concentrations of copper in plants vary widely, they usually range from 1 to 50 ppm (dry weight) (Davies and Bennett 1985) and from 1 to 143 ppm (dry weight) in edible plants (Perwak et al. 1980). Concentration ratios of copper in plants relative to soil (concentration factors or CF) demonstrate that copper uptake differs significantly between plants. For example, CF values have been found to vary from 0.02 (onion), 0.13 (celery), 0.21 (lettuce), and 0.30 (potato) to 2 (grapes), 4.5 (alfalfa), and 6.8 (grass) (Pinochet et al. 1999). Concentration factors in rice grown in Japan were found to vary among soil types (0.59–3.58) with copper concentrations in rice ranging from 1.7 to 5.1 µg/g (Herawati et al. 2000). Copper concentrations in rice grain from the Yangtze delta in China have been found to increase significantly from 1.4 to 15.5 µg/g when copper concentrations in wastewater irrigated soils increased from 17.0 mg/kg (wet weight) to 101.2 mg/kg (wet weight) (Cao and Hu 2000).

Studies of copper in human tissues suggest that copper content in a 70 kg adult ranges from 50–70 mg (Davies and Bennett 1985). Wise and Zeisler (1984) reported an average copper concentration of 10 ppm in the human liver in 36 samples. Despite the wide variation in copper concentrations in the environment, the copper concentration in the liver only varied by a factor of 2–3.5. The concentration of copper in blood is not expected to be predictive of the total body burden of copper: Saltzman et al. (1990) found that the correlation between copper concentrations measured in blood and total body burden was poor ($r=0.54$).

Copper content in 25 tea samples from China ranged from 7.73 to 63.71 mg/kg (Zhong et al. 2015). In a study of copper release from the inner surface of copper teapots, Ni and Li (2008) found that Cu_2O was a main mineral component of the corrosion by-products.

The range of copper concentrations in the filler tobacco of 10 cigarette brands manufactured by British American Tobacco and International Tobacco Company was 18.26–34.94 µg/cigarette (Benson et al. 2017). The range in the filters after smoking was 1.77-36.48 µg/g. The mean copper content of tobacco in Finnish cigarettes was 24.7±10.8 ppm (Mussalo-Rauhamaa et al. 1986). However, only 0.2% of this

copper passes into mainstream smoke. This translates to a daily exposure of approximately 1 µg of copper in a pack of 20 cigarettes.

In an EPA-sponsored study conducted to determine the metal concentration in sewage sludge (Feiler et al. 1980), copper concentrations in primary sludge at seven POTWs were reported to be 3.0–77.4 ppm, with a median concentration of 20.5 ppm. The plant with the highest copper concentrations received wastes from plating industries, foundries, and coking plants. In a comprehensive survey of heavy metals in sewage sludge, 30 sludges from 23 American cities were analyzed (Mumma et al. 1984). The copper concentration in the sludges ranged from 126 to 7,729 ppm (dry weight), with a median value of 991 ppm. Gutenmann et al. (1994) report similar concentrations (217–793 ppm, dry weight) in sewage sludge obtained from 16 major cities in the United States. The proposed limit for copper in sludge spread on agricultural land is 1,000 ppm (Mumma et al. 1984). The concentration of copper in cow's manure is ~5 ppm (Mumma et al. 1984).

In municipal solid waste compost obtained from nine sites in the United States, a mean copper concentration of 281 mg/kg (dry weight) was obtained with range of 36.4–424 mg/kg (He et al. 1995). Lisk et al. (1992) reported copper concentrations in composts formed from yard waste ranging from 22.7 to 327 ppm, from sewage sludge ranging from 432 to 1,019 ppm and from municipal solid waste ranging from 191 to 1,143 ppm.

Bolan et al. (2003) analyzed copper in farm effluent and sludge samples at dairy and pig farms that utilized copper hydroxide and at farms that did not use copper hydroxide. The concentration of total copper was higher at farms that used the compound. Copper concentration was higher in the sludge samples than the effluent. At dairy farms utilizing copper hydroxide, copper concentration ranged from 52 to 105 mg/kg in sludge and from 2.5 to 10.5 mg/l in effluent. At pig farms utilizing copper hydroxide, copper concentrations ranged from 12.5 to 526 mg/kg in sludge and from 0.1 to 1.55 mg/l in effluent.

Copper concentrations in waste from the combustion of municipal solid waste and other combustion processes have been reported. Copper in incinerator bottom ash and fly ash has been measured at mean concentrations of 1,700 and 1,000 mg/kg, respectively (Goldin et al. 1992). Buchholz and Landberger (1995) report concentrations of copper of 390–530 µg/g in fly ash, 1,560–2,110 µg/g in bottom ash, and 1,140–1,540 µg/g in combined ash. In sewage sludge incineration process steams, copper concentrations were 4,561 mg/kg in sludge cake, 3,465 mg/kg in bottom ash, 3,707 mg/kg in cyclone ash, 3,684 mg/kg in scrubber particulate matter, and 6,666 mg/kg in stack particulate matter (Balogh 1996). In fossil fuel wastes, copper concentrations of 33–2,200 mg/kg in fly ash, 4–930 mg/kg in bottom ash, 6–340 mg/kg in

flue gas desulfurization sludge, 10–130,000 mg/kg oil ash, and 2–190 mg/kg in coal have been obtained (Eary et al. 1990).

Copper concentrations have been measured in several types of electronic and e-waste. The concentration of copper was 276,186-423,727 mg/kg in discarded basic phones and 268,945-434,628 mg/kg in discarded smartphones (Singh et al. 2019). The average concentration in basic phones and smartphones was 378,406 and 357,560 mg/kg, respectively. The average weight of copper in different electronic devices is: 700,300 mg in plasma TVs, 625,600 mg in color CRT TVs, 206,000 mg in LCD TVs, 102,800 mg in laptop computers, 59,500 mg in LCD monitors, and 18,800 mg in cell phones (Woo et al. 2016). In an assessment of hazardous chemicals in a market-representative set of waste printed circuit boards (WPCBs) originating from computers manufactured from 1996–2010, copper was found ranging from 177,000 to 268,000 mg/kg and was the most abundant metal in the WPCBs (Chen et al. 2016). In WPCBs, copper is used to transmit electric signals and is fundamental but results from the study show that technological innovation modeled by three types of Intel chipsets correlates with an overall decrease in copper concentration (Chen et al. 2016).

Copper may also be found in clothing. Herrero et al. (2020) analyzed 39 swimsuits made in Vietnam, China, Cambodia, Albania, Sri Lanka, Bangladesh, Tunisia, Spain, Morocco, and Myanmar. Copper was detected in 64% of the samples at an average concentration of 27.9 mg/kg, with concentrations ranging from less than 0.15 to 328 mg/kg. Although Herrero et al. (2020) does not specifically discuss the origins of copper in swimsuits, the authors do note that many swimsuits are made of artificial fibers so that they may be water repellant or fast drying. Metals may be used in the textile industry as dyes, antimicrobials, and water repellants (Herrero et al. 2020).

An assessment of trace metals in lip balms, lip glosses, and lipsticks found that copper was one of the three major trace metals found in lip cosmetics (Gao et al. 2018). Copper concentrations ranged from 11.07 to 136.73 mg/kg in the products sampled. The mean concentrations were 61.96 (lip balms), 81.28 (lip glosses), and 93.93 mg/kg (lipsticks).

Copper has been detected in pigments in American tattoo ink (Liszewski and Warshaw 2019). Of 44 distinct pigments identified, four contained copper. All four pigments were phthalocyanine. The most frequently used pigment containing copper is found in 13 tattoo ink brands and in 562 inks; the least frequently used is found in 1 brand and 1 ink.

Concentrations of copper in fertilizers, soil amendments, and other agricultural materials have been measured by Raven and Loeppert (1997). The materials and mean concentrations are urea (<0.6 µg/g),

ammonium nitrate (<0.6 µg/g), ammonium sulfate (<0.6 µg/g), ammonium phosphate (<2– 41.8 µg/g), potassium chloride (<2–3.5 µg/g), potassium-magnesium-sulfate (1.4–5 µg/g), North Carolina rock phosphate (9.6 µg/g), calcite (2.3 µg/g), corn leaves (9.4 µg/g), manure (17.5 µg/g), and austinite (300 µg/g). Copper was measured in cement dust from the United States at an average concentration of 23.66 ± 7.23 µg/g (Ogunbileje et al. 2013).

5.6 GENERAL POPULATION EXPOSURE

Due to the ubiquity of copper in the environment and the general occurrence of copper in airborne particulates, exposure to copper through inhalation is commonplace. Estimates of atmospheric copper concentrations from different source categories (e.g., smelters, ore processing, steel production, and combustion) yielded a maximum annual concentration of 30 µg/m^3 (EPA 1987). If a person is assumed to inhale 20 m^3 of air/day, this would amount to an average daily intake of 600 µg of copper. For the reported range of annual atmospheric copper concentrations, 5–200 ng/m^3 (EPA 1987), the average daily intake by inhalation, would range from 0.1 to 4.0 µg. At the maximum reported ambient air concentration, 100 µg/m^3 for a 24-hour period at a location within one-half mile of a major source (EPA 1987), the average daily intake would rise to 2,000 µg. These estimates assume that all of the copper is attached to particles of inhalable size, less than 10 µm in diameter.

The average daily dietary intake of copper from food is ~2 mg/day. The dietary intake of copper is expected to be above this average for those individuals who regularly consume organ meats (e.g., liver and kidney), nuts, seeds (including cocoa powder), legumes, and bran and germ portions of grains; these intakes are not expected to exceed the maximum recommended limits of 10–12 mg/day (WHO 1996). In the United States, Tolerable Upper Intake Levels vary by life stage, ranging from 1 mg/day for 1-year old children and 10 mg/day for adults and pregnant and lactating females 19-years old and older (Table 7-17). Those individuals who regularly consume oysters or clams may increase their dietary intake of copper by 2–150 mg/day when consuming 250 g of edible tissue per day, based on copper concentrations of 25–600 and 8.4–171 ppm in oysters and clams, respectively (Goldberg 1986; Perwak et al. 1980). Assuming a median copper concentration in drinking water of 75 µg/L, the average daily copper exposure from consumption of 2 L of water per day is 0.15 mg. However, many people may have high levels of copper in their tap water that were acquired during transport through the water distribution system. While corrosion can occur in plumbing of any age, new copper plumbing is a potential source of exposure as copper leaches into drinking water. In the presence of certain water qualities, copper levels in excess of the EPA action level (1.3 mg/L) are most likely to occur in newly constructed homes and buildings with copper plumbing, or at sites that have been recently renovated with new copper plumbing (Edwards et al.

2001; EPA 1995; Grace et al. 2012; Knobeloch et al. 1998; Lagos et al. 2001; Rajaratnam et al. 2002; Schock and Sandvig 2009; Turek et al. 2011). If the system is not permitted to flush out, average intakes from water may be >2 mg/day. It is less likely that high dermal exposures will result from bathing in this tap water because the distribution system will flush itself out as the water is drawn. Data on serum copper from NHANES is presented in Table 5-21.

Table 5-21. Geometric Mean and Selected Percentiles of Serum Copper (in µg/L) for the U.S. Population from the National Health and Nutrition Examination Survey (NHANES) (CDC 2016, 2018)

	Survey years	Geometric mean (95% CI)	Selected Percentiles				Sample size
			50th	75th	90th	95th	
Total	13-14	1148.34 (1122.60-1174.68)	1135	1319	1547	1710	2520
	15-16	1146.60 (1124.94-1168.68)	1130	1314	1538	1692	2436
Age group							
12 – 19 years	13-14	1055.83 (1033.20-1078.96)	1036	1197	1414	1641	418
	15-16	1055.03 (1012.29-1099.57)	1031	1232	1408	1534	371
20 – 59 years	13-14	1161.59 (1129.35-1194.75)	1138	1343	1607	1787	1221
	15-16	1152.32 (1129.72-1175.38)	1124	1315	1600	1794	1165
60 years and older	13-14	1149.31 (1117.16-1182.39)	1165	1310	1472	1599	542
	15-16	1161.35 (1123.66-1200.31)	1150	1327	1479	1617	579
Sex							
Male	13-14	1032.39 (1001.14-1064.63)	1032	1173	1308	1414	1235
	15-16	1042.57 (1021.11-1064.49)	1043	1171	1332	1422	1201
Female	13-14	1271.39 (1246.35-1296.93)	1244	1453	1677	1908	1285
	15-16	1254.96 (1221.69-1289.15)	1241	1429	1672	1903	1235
Race/ethnicity							
Mexican American	13-14	1163.47 (1132.65-1195.12)	1157	1353	1567	1738	431
	15-16	1147.80 (1118.65-1177.70)	1131	1294	1534	1747	464
Other Hispanic	13-14	1181.15 (1126.79-1238.13)	1156	1375	1587	1656	235
	15-16	1142.06 (1115.41-1169.35)	1133	1323	1493	1741	334
Non-Hispanic white	13-14	1131.74 (1104.19-1159-97)	1111	1300	1504	1679	975
	15-16	1134.14 (1107.51-1161.40)	1117	1295	1476	1652	764
Non-Hispanic black	13-14	1250.99 (1217.36-1285.56)	1242	1469	1653	1763	516
	15-16	1270.74 (1228.30-1314.63)	1245	1452	1705	1959	494
Other race	13-14	1105.52 (1075.40-1136-49)	1082	1292	1455	1659	363
	15-16	1091.44 (1053.23-1131.04)	1095	1231	1486	1653	380

CI = confidence interval

A National Occupational Exposure Survey (NOES) conducted by NIOSH from 1981 to 1983 estimated that potentially 505,982 workers, including 42,557 women, were occupationally exposed to copper in the United States (NIOSH 1989). The NOES estimate is provisional because all of the data for trade name

products that may contain copper have not been analyzed. Of the potential exposures, 1,073 are to pure copper, while in the other cases, the molecular form of copper was unspecified. Additionally, according to the NOES, 125,045 workers, including 38,075 women, were potentially exposed to copper sulfate (NIOSH 1988). The NOES was based on field surveys of 4,490 facilities and was designed as a nationwide survey based on a statistically valid sample of virtually all workplace environments in the United States where eight or more persons are employed in all standard industrial codes (SIC) except mining and agriculture. The exclusion of mining and agriculture is significant for estimating exposure to copper since there is a high potential for exposure in these industries. Current occupational exposure limits for copper fume are 0.2 and 1 mg/m^3 for dust and mists (Frazier and Hage 1998).

Children are not small adults. A child's exposure may differ from an adult's exposure in many ways. Children drink more fluids, eat more food, breathe more air per kg of body weight, and have a larger skin surface in proportion to their body volume. A child's diet often differs from that of adults. The developing human's source of nutrition changes with age: from placental nourishment to breast milk or formula to the diet of older children who eat more of certain types of foods than adults. A child's behavior and lifestyle also influence exposure. Children crawl on the floor, put things in their mouths, sometimes eat inappropriate things (such as dirt or paint chips), and spend more time outdoors. Children also are closer to the ground, and they do not use the judgment of adults to avoid hazards (NRC 1993).

Children could be exposed to copper through contact with wood treated with alkaline copper quaternary (ACQ) (Cushing et al. 2007). ACQ, which contains copper oxide, is used to treat residential decks and playsets. Children might ingest ACQ from dislodged wood residues via hand to mouth contact or be exposed via dermal contact.

Exposure of copper through oral routes may differ between children and adults, due to differences in the consumption of various food groups between children and adults and ingestion of dust and soils. The dietary copper intake for infants who receive the major portion of their nutritional requirements from breast milk is likely to be different from infants whose nutritional needs are either supplemented or entirely received through the consumption of formula. Estimates of copper intake from inhalation and ingestion in children in the United States are limited. From the work of Pennington et al. (1986), the copper intakes from food consumption for a 6–11-month-old infant and a 2-year-old child were estimated to be 0.47 and 0.58 mg/day, respectively, values which are lower than the adult intake of ~1 mg/day. One study has provided estimated inhalation and ingestion exposures of copper for 6- to 10-year-old children in India (Raghunath et al. 1997). In this work, mean daily concentrations of copper in particulates in air from six locations were measured at 0.01–0.26 µg/m^3. Based on these measurements, estimated inhalation

exposures of children to copper were calculated to be 0.1–3.2 µg/day. In this same work, exposures to copper through ingestion were estimated to be between 684–1,732 µg/day.

Exposures of children to copper are likely to increase in areas where copper concentrations in air are expected to be high, such as mining sites, waste dump sites, smelters, and foundries. For example, copper burdens in children living near a lead smelter, as measured by copper concentration in teeth, increased closer to the smelter (Blanuša et al. 1990). Children are also at risk for increased copper intake through consumption of drinking water where leaching of copper from the distribution system has occurred (Murphy 1993; Yannoni and Piorkowski 1995). Copper-contaminated drinking water has been reported to create a light blue or blue-green color to water, and may results in a metallic, bitter taste (WHO 2004). This route of copper exposure can be minimized through the flushing of drinking water supply lines or increasing the pH of the water in the distribution system.

Arcega-Cabrera and Fargher (2016) measured copper in blood and urine samples of children in Mexico and found that 79.4% had copper detected in urine and 100% had copper detected in blood. The range of median copper in blood at nine elementary schools was 723.02 to 1143.7 µg/dL, and in urine ranged from below detection limit to 20.62 µg/dL. Using ethnographic data, Arcega-Cabrera and Fargher (2016) identified potential sources and pathways of exposure to metals. They concluded that children from poor or marginalized families tended to be exposed to copper while children from wealthier families tended to be exposed to inorganic copper (copper sulfate). There was a positive correlation between the frequency that children ate fresh fish and copper in blood, while there was a negative correlation between the frequency and copper in urine. This is likely due to the copper in fish being protein-bound. Since copper sulfate is used as a preservative in fresh fish and as a water treatment in ponds and other freshwater surfaces, children who eat fresh fish more often may be exposed to it. Piped or well water in the study was found to contain higher levels of copper than purified water, and children of poorer or more marginalized families who cooked with piped or well water had higher levels of copper in urine. Children from households cooking over open food fires also had higher levels of copper in urine than households cooking with gas.

5.7 POPULATIONS WITH POTENTIALLY HIGH EXPOSURES

In discussing exposure to copper, the important question is whether individuals are exposed to readily available copper, which in general means free (hydrated) Cu(II) ions and perhaps some weakly complexed or adsorbed small particulate copper ions. The data indicate that copper in natural water, sediment, and soil mainly exists in bound form. Even so, the free form of copper can be released readily from ingested materials, for example a child’s sampling of soil contaminated with copper, followed by

exposure to the low acidic pHs encountered in the stomach (Pizarro et al. 2001). Potential for high uptakes of copper in the general population may exist in situations where people consume large amounts of tap water that contains dissolved copper that come from corrosion of copper in the distribution system, or already have a high copper background due to natural or anthropogenic activities (e.g., close proximity to mining activities or mine drainage). Leaching of copper from water distribution system materials is likely to occur where the water is soft and not flushed out of the system by running the water down the drain before collecting some of it for use. In such cases, the initial concentration of copper frequently exceeds 1 ppm. A large fraction of the copper may be in the form of free cupric ion, and uptake will result by ingestion and, perhaps, dermal contact. Soluble cupric salts are used extensively in agriculture and in water treatment. Workers engaged in the formulation and application of these chemicals along with industrial workers, such as those in the plating industry, may come into dermal contact with absorbable copper ions. Exposure to high levels of free Cu(II) can occur, for example, from swimming in water that was recently treated with a copper-containing algaecide.

Serum concentrations of copper were significantly elevated in users of skin-whitening agents (Iyanda et al. 2011). Copper concentrations ranged from 2.27 to 8.48 mg/kg in skin lightening creams sold in Nigeria (Sani et al. 2016; Theresa et al. 2011). Consumers who use skin-whitening agents could be at risk of high exposure to copper.

Based on the available data, people living close to NPL sites may be at greater risk for exposure to copper than the general population. In this case, exposure can occur through inhalation of airborne particulates from the NPL sites, ingestion of water from private wells in close proximity to the sites, ingestion of contaminated soil, and/or uptake of copper into fruits and vegetables raised in gardens of residents living near NPL sites.

People living near copper smelters and refineries, and workers within these and other industries can be exposed to high levels of dust-borne copper by both inhalation and ingestion. In some industries, workers may be exposed to fumes or very fine dust that may be more hazardous than coarse-grained dust, because it can be inhaled and penetrate more deeply into the lung, thereby evading the mucocilliary escalator.

Exposure to ultrafine particles of copper poses a risk to human health due to their smaller size, larger surface area, surface material, and physical characteristics (Schraufnagel 2020). Traffic exhaust is a common source of exposure, although homes near a trash burning site, bedrooms with burning coils for mosquito abatement, homes with smokers, and kitchens during domestic cooking are also sources of exposure to ultrafine particles (Schraufnagel 2020). Particles created by brake wear, including copper

particles, are in the range of 2.8 μm (Wahlin et al. 2006). Copper has been identified in ultrafine particles leading to metal fume fever among welders (Schraufnagel 2020).

CHAPTER 6. ADEQUACY OF THE DATABASE

Section 104(i)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the health effects of copper is available. Where adequate information is not available, ATSDR, in conjunction with NTP, is required to assure the initiation of a program of research designed to determine the adverse health effects (and techniques for developing methods to determine such health effects) of copper.

Data needs are defined as substance-specific informational needs that, if met, would reduce the uncertainties of human health risk assessment. This definition should not be interpreted to mean that all data needs discussed in this section must be filled. In the future, the identified data needs will be evaluated and prioritized, and a substance-specific research agenda will be proposed.

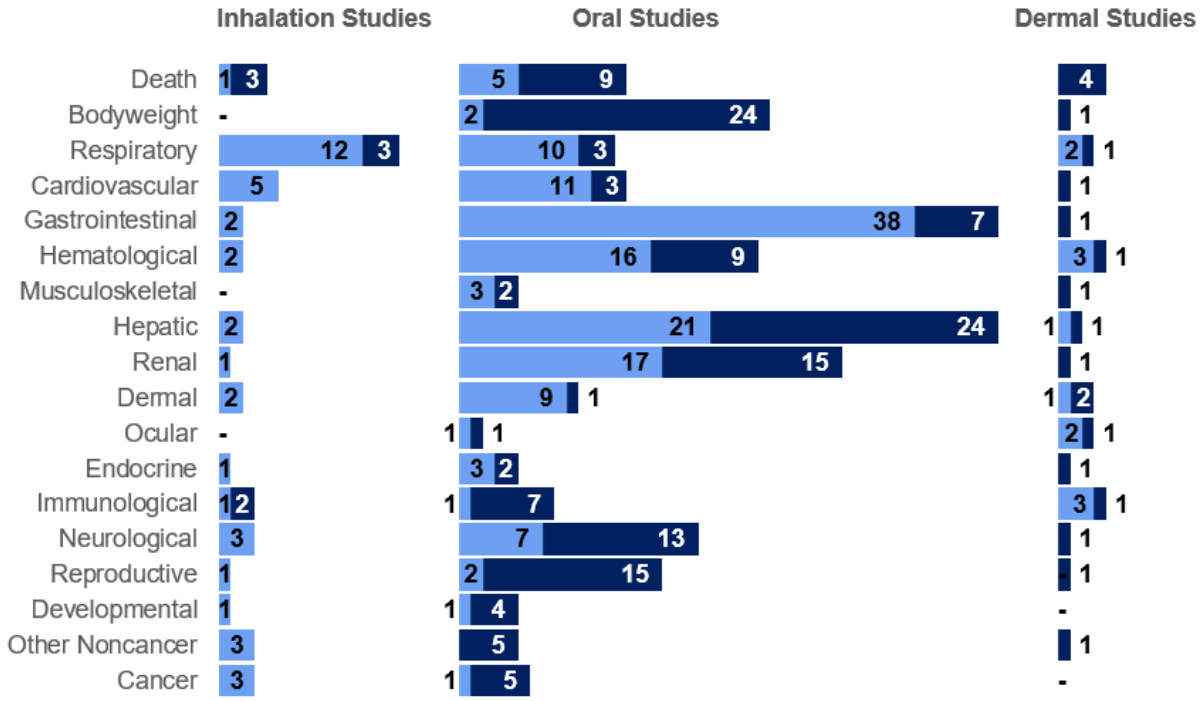
6.1 EXISTING INFORMATION ON HEALTH EFFECTS

Studies evaluating the health effects of inhalation, oral, and dermal exposure of humans and animals to copper that are discussed in Chapter 2 are summarized in Figure 6-1. The purpose of this figure is to illustrate the information concerning the health effects of copper. The number of human and animal studies examining each endpoint is indicated regardless of whether an effect was found and the quality of the study or studies.

As shown in Figure 6-1, information on the health effects in humans exposed to copper primarily apply to oral ingestion. Many of these studies are case reports of individuals who intentionally or accidentally ingested copper or copper-containing substances. Epidemiological and controlled-exposure studies in humans primarily examined effects following ingestion of copper in drinking water. In these studies, gastrointestinal symptoms were the most frequently observed health effect. There are a robust number of experimental studies in animals that examine a wide range of health effects following oral exposure to copper and/or copper compounds, particularly the hepatic and renal toxicity endpoints. Inhalation and dermal studies were limited in both animals and humans, but the results generally support the effects following oral ingestion.

Figure 6-1. Summary of Existing Health Effects Studies on Copper by Route and Endpoint*

Potential gastrointestinal and hepatic effects were the most studied endpoints.
The majority of these studies examined oral exposure in **humans** (versus **animals**).



*Includes studies discussed in Chapter 2; the number of studies includes those finding no effect.

6.2 IDENTIFICATION OF DATA NEEDS

Missing information in Figure 6-1 should not be interpreted as a "data need." A data need, as defined in ATSDR's *Decision Guide for Identifying Substance-Specific Data Needs Related to Toxicological Profiles* (ATSDR 1989), is substance-specific information necessary to conduct comprehensive public health assessments. Generally, ATSDR defines a data gap more broadly as any substance-specific information missing from the scientific literature.

Acute-Duration MRLs. The acute-duration oral human database was adequate for the derivation of an acute-duration oral MRL. Gastrointestinal symptoms occur in humans following acute ingestion of excess copper in drinking water (Araya et al. 2001, 2003a, 2003c; Gotteland et al. 2001; Olivares et al. 2001; Pizarro et al. 1999, 2001). Gastrointestinal toxicity is supported by evidence in the oral database in animals where gastrointestinal symptoms and histological changes in the gut were observed (Cheng et al. 2020; Kadammattil et al. 2018; Khushboo et al. 2018). The acute-duration inhalation database was not adequate for the derivation of an inhalation MRL. The database is limited to one study in humans

examining the immunological endpoint which was of insufficient quality as there was no comparable control group (Markert et al. 2016), and a study in animals examining the respiratory and immunological endpoints (Drummond et al. 1986). Two additional studies only examined the death endpoint (Holbert 1990; Rush 1991). Toxicity studies examining gastrointestinal and hepatic toxicity from inhalation of excess copper would be useful to identify the target of toxicity. Additionally, studies examining a possible concentration-response relationship would be useful especially in occupational settings where such exposures are likely to occur.

Intermediate-Duration MRLs. The intermediate-duration oral animal database was not adequate for the derivation of an intermediate oral MRL. The acute oral MRL was adopted to the intermediate oral MRL. Two studies found increased incidence of gastrointestinal symptoms in humans over a 2-month exposure to copper doses ≥0.106 mg Cu/kg/day (Araya et al. 2003b, 2004). However, the reports as related to gastrointestinal responses were episodic and data for systemic effects are limited. Alternatively, no copper-related effects on the gastrointestinal system, liver or on body weight were observed at doses as low as 0.058–0.3 mg Cu/kg/day (O'Connor et al. 2003; Olivares et al. 1998). A study in nine men showed that oral exposure to 0.1 mg Cu/kg/day for 18 days resulted in reduced antibodies to a strain of influenza following immunization, when compared to non-exposed control (Turnlund et al. 2004). Oral studies in animals examined a wide range of endpoints and data indicates that the liver is a sensitive target of copper toxicity (Hashish and Elgaml 2016; Kvietkauskaite et al. 2005; Wu et al. 2020; Epstein et al. 1982; Shen et al. 2005). The intermediate-duration inhalation database was not adequate for the derivation of an inhalation MRL; it is limited to two studies in rabbits that did not report any adverse health outcomes (Johansson et al. 1983, 1984). It would be useful for toxicity studies to identify the target of copper toxicity following intermediate-duration inhalation exposure to copper, as well as establish concentration-response relationships.

Chronic-Duration MRLs. No studies examining chronic-duration inhalation or oral exposure in humans or animals were adequate for the derivation of chronic MRLs. The oral database is limited to a few studies in animals where decreased lifespan, reduced hemoglobin, and mild hepatic effects were reported (Araya et al. 2012; Massie and Aiello 1984). Since serious health effects have been reported at the acute- and intermediate-durations, studies examining the toxicity of exposure to excess chronic doses of copper may be useful to establish chronic doses that would be harmful to humans. The NIH's tolerable upper intake level for copper in 900 µg/day for adults aged 19 to >70 years. Studies would be helpful to determine effects of ingestion exceeding this level. No inhalation studies in either humans or animals provided any data on copper toxicity. Studies in the inhalation database were limited to studies reporting associations; however, the data is suggestive at best. Studies examining toxicity resulting from chronic

exposure to excess copper in air would be useful for populations living near sites currently processing, using, or manufacturing copper that may enter the local environment, and for workers who handle copper in occupational settings.

Health Effects.

Respiratory. Symptoms of respiratory irritation, including coughing, sneezing and thoracic pain, were observed in workers following inhalation of copper in occupational settings (Askergren and Mellgren 1975; Suciu et al. 1981). Inhalation of copper fumes in an occupational case study resulted in persistent sinus pressure and rhinorrhea (Gibson et al. 2011). A copper sulfate mixture was implicated as the etiologic agent in a unique disease observed among vineyard workers spraying an antimildew agent (Pimentel and Marques 1969; Pimentel and Menezes 1975; Stark 1981; Villar 1974; Villar and Nogueira 1980). Drummond et al. (1986) observed decreased cilia beating in hamsters and alveolar thickening in mice that were exposed repeatedly, and toxicity increased with duration of exposure. Further studies are needed to characterize respiratory toxicity of copper, especially in workers who likely inhale copper dust or fumes in occupational settings. Additionally, concentration-response relationships are yet to be established.

Immunological. Limited evidence in humans and animals suggests that excess copper may be immunotoxic. Reports on humans developing dermatitis after dermal exposure to copper (Barranco 1972; Saltzer and Wilson 1968) suggest that copper is an allergen. This is supported by a report of a woman developing dermatitis after insertion of a copper IUD (Barranco 1972). Increased blood C-reactive protein, an indication of asymptomatic inflammation, was seen in a controlled-study where adult volunteers were exposed to copper-containing welding fumes of 0.53 mg Cu/m^3 for 6 hours, 3 times over 3 weeks (Markert et al. 2016). A study in adult men found that antibodies to an influenza strain were less after immunization when compared to controls following exposure to 0.1 mg Cu/kg/day (Turnlund et al. 2004). Immunological effects were observed in mice following acute inhalation exposure to copper sulfate (Drummond et al. 1986). Copper produced a toxic effect on the antioxidant defense system in mice; decreased percentage of suppressor, natural killer, and its precursor, and increased immunoregulatory index were both reported (Kvietkauskaite et al. 2004). In addition, impaired immune function is observed in mice exposed to copper chloride (Pocino et al. 1991) or copper sulfate (Pocino et al. 1990) in drinking water. Histological changes in the spleen were observed in mice and rats orally exposed to copper compounds (Kadammattil et al. 2018; Khushboo et al. 2018). More studies in humans and animals that examine the immune response to copper exposure would be useful to understand possible dose-response relationships and assess species differences.

Neurological. Neurological impairment was observed in factory workers exposed to copper dust (Donoso et al. 2007). Clinical symptoms of neurotoxicity were observed in a drinking water study of 60 females, with 6 subjects reporting increased salivation (Pizarro et al. 1999). Dizziness, agitation, and drowsiness were noted in case studies of copper ingestion (Malik and Mansur 2011; Du and Mou 2019; Gunay et al. 2006; Yang et al. 2004). No effects on neurobehavioral performance and no histological changes were observed in several oral studies in animals (Kadammattil et al. 2018; Lu et al. 2009; NTP 1993; Seffner et al. 1997). At 4 mg Cu/kg/day, rats showed indications of copper-induced neurotoxicity including neurobehavioral change, impaired muscle strength, and coordination (Kumar et al. 2019). No effects on neurobehavioral performance were observed in rats fed 250 ppm copper in their diet (Murthy et al. 1981). However, this study did find alterations in the levels of a dopamine metabolite, suggesting that copper may adversely affect the nervous system. Serious neurotoxic effects at doses ≥25.5 mg Cu/kg/day included impaired motor coordination and cognitive function (Kalita et al. 2020; Khushboo et al. 2018; Kumar et al. 2015, 2016a, 2016b). Additional studies are needed to further investigate the neurotoxic potential of copper; these studies should assess the potential of copper to perturb dopaminergic pathways and related functions.

A recent *in vitro* study indicates that copper may be critically involved in optimal functioning of the circadian clock by modulating cell metabolism, redox state, transcription, and neuronal activity (Yamada and Prosser, 2020). Alterations in these circadian rhythms have previously been implicated in increased risk for cardiometabolic diseases, cognitive and mood disorders, and sleep disorders (Luojus et al., 2015; Song et al., 2015; Yoshioka et al., 2018; Yukihiro et al., 2020; Abbott et al., 2020). Studies that investigate the effects of inhalation, oral, and dermal exposure to copper that examine its effects on circadian rhythms need to be designed and conducted in animal and human paradigms. These studies need to examine the alterations in circadian machinery and the potential downstream alterations in physiology in the organisms.

There is a growing body of literature that indicates copper may play a role in the development of Alzheimer's disease and other similar neurodegenerative diseases (Pohanka 2019). Medical studies have found evidence that the metabolic balance and distribution of copper is disrupted in individuals with Alzheimer's disease (Coelho et al. 2020). However, the current literature is unclear on how environmental exposures to copper affect the development of Alzheimer's disease. Since the current neurological literature in animals and humans indicate copper can lead to neurological impairment and given that copper can distribute to the brain, epidemiological studies examining the relationship between environmental exposures to copper and Alzheimer's disease would be useful in understanding long term risks of exposure.

Developmental. Developmental studies examining oral exposure to copper in rats (Haddad et al. 1991) and mice (Lecyk 1980) have shown that high copper intakes can result in impaired growth. In rabbit fetuses, mean fetal weight was reduced; 4 fetuses had protrusion of the abdomen; and there was increased incidence of hemivertebrae, delay ossification, and supernumerary ribs compared to controls (Munley 2003a, 2003b). Kadammattil et al. (2018) found no copper-related changes in implantations, non-viable embryos, resorbed embryos, or embryo body weight. The developmental toxicity of copper in humans is not adequately investigated, as animal studies produce strong evidence that copper may be toxic to the reproductive system (Arafa et al. 2019; Babaei et al. 2012; Kadammattil et al. 2018; Khushboo et al. 2018; Liu et al. 2016; Munley 2003a, 2003b; NTP 1993; Sakhaee et al. 2012, 2016a, 2016b). No data were located regarding developmental effects of copper after inhalation or dermal exposures in humans or animals. Multigeneration studies and further investigations in different animal species would provide valuable information on the potential of copper to adversely affect development. Such information might be relevant to humans.

Cancer. Data on the carcinogenicity of copper in humans are limited. A study of copper miners (Chen et al. 1993) and a follow-up to this study (Chen et al. 1995) observed increased risk of cancer, stomach cancer, and lung cancer. Because the workers were also exposed to radon and radon daughters, silica, iron, titanium, sulfur, and arsenic, a causal relationship between copper and increased cancer risk cannot be established. Only one study examined the association between copper ingestion and cancer risk in humans. Odds of leukemia cancer development was not affected by copper levels in carpet dust, which was presumably ingested (Raaschou-Nielsen et al. 2016). This study was very limited in the exposure examined and results are not indicative. A prospective cohort study of populations across Europe found that copper in $PM_{2.5}$ was associated with increased risk of lung cancer (Hazard Ratio = 1.25; 95% CI=1.01-1.53), while PM_{10} was not associated with increased risk of lung cancer (Hazard Ratio = 1.14; 95% CI=0.96-1.35) (Raaschou-Nielsen et al. 2016). These observations were also not indicative of a causal relationship. Several animal studies have examined the carcinogenic potential of ingested copper (Abe et al. 2008; Greene et al. 1987; Kamamoto et al. 1973). These studies are limited in scope; the studies by Green et al. (1987) and Kamamoto et al. (1973) only examined one potential target and tested fairly low doses of copper. No dermal carcinogenicity studies in humans or animals were identified. Additional studies by the inhalation, oral, and dermal routes are needed to assess the carcinogenic potential of copper in humans.

Genotoxicity. No data on the genotoxicity of copper in humans were located; studies of workers or individuals accidentally exposed to high levels of copper would provide value information on its genotoxic potential in humans. The available genotoxicity data suggest that copper is a clastogenic agent

(Agarwal et al. 1990; Bhunya and Jena 1996; Bhunya and Pati 1987; Fahmy 2000; Sideris et al. 1988). However, mixed results are found in point mutation assays (Demerec et al. 1951; Marzin and Phi 1985; Singh 1983; Tso and Fung 1981; Wong 1988). Additional studies are needed to assess copper's potential to induce point mutations. Several studies have also shown that exposure to copper can result in DNA damage (Caicedo et al. 2007; Garrett and Lewtas 1983; Husain and Mahmood 2019; Prasad et al. 2006; Sideris et al. 1988; Sina et al. 1983; Urbino-Cano et al. 2006).

Copper Nanoparticles. The toxicity of copper nanoparticles has not been examined in humans and studies in animals are limited. Data primarily come from *in vivo* and *in vitro* studies examining its genotoxicity and cytology. Oral exposure studies in rats and mice suggest CuNPs cause histological damage to the liver and alter enzyme levels (Anreddy et al. 2018; El Bialy et al. 2020; Chen et al. 2006; De Jong et al. 2019; Tang et al. 2018). Kidney damage is also reported in rats and mice from oral CuNP exposure (El Bialy et al. 2020; Chen et al. 2016; De Jong et al. 2019). These effects are similar to those seen in animals following oral exposure to ionic copper. Further studies are needed to determine if renal and hepatic toxicity is expected at levels lower than those of ionic copper. Additionally, rats and mice show gastrointestinal, respiratory, and neurotoxic effects following oral exposure, which are similar to effects seen in humans following both oral exposure and inhalation exposure, especially in workers. Occupational studies in workers exposed to CuNPs toxicity would help elucidate the effect that particle size has on toxicity, particularly in the respiratory system.

Epidemiology and Human Dosimetry Studies. Several studies have examined the toxicity of inhaled copper in workers (Askergren and Mellgren 1975; Finelli et al. 1981; Suciu et al. 1981). These studies have primarily focused on the respiratory tract, although health examinations revealed other adverse effects (e.g., hepatomegaly). Chen et al. (1993, 1995) examined the carcinogenic potential of inhaled copper. In general, these studies are limited by poor exposure characterization, co-exposure to several toxic and/or carcinogenic compounds (e.g., arsenic, cadmium, radon, lead), and limited number of endpoints examined. Occupational exposure studies examining populations of workers exposed to copper and with minimal exposure to other metals would be useful in assessing the toxicity of inhaled copper. These studies should examine a wide variety of endpoints, particularly the gastrointestinal tract, liver, and kidneys, which are well established targets of toxicity following oral exposure. There are numerous reports of accidental or intentional ingestion of copper, and the most commonly reported effect in these studies is gastrointestinal upset, followed by liver effects. There have been several experimental studies designed to examine gastrointestinal upset following short-term (2 weeks or less) exposure to copper in drinking water (Araya et al. 2001, 2003a, 2003c, 2004; Gotteland et al. 2001; Olivares et al. 2001; Pizarro et al. 1999, 2001). Several studies have examined health effects, mainly gastrointestinal but also hepatic,

in populations exposed to elevated level copper levels in drinking water (Buchanan et al. 1999; Dieter et al. 1999; Eife et al. 1999; Knobeloch et al. 1994, 1998; Pettersson et al. 2003; Pizarro et al. 2007; Zietz et al. 2003a, 2003b). Studies in both children and adults could elucidate the understanding of possible age-related differences in toxicity.

Biomarkers of Exposure and Effect.

Exposure. Copper levels can be measured in tissues, body fluids, excreta, hair, and nails. Whole blood, serum, and urine copper levels have been established in healthy individuals. It has been demonstrated that copper levels in the body increase with increased exposure after acute poisoning. Similarly, increased copper levels were observed in workers after occupational exposure. Serum and urine copper levels, plasma ceruloplasmin levels, and clinical manifestations are specific indicators of copper status. Current biomarkers appear sufficient for assessing copper exposure.

Effect. There are no specific biomarkers for copper toxicity. Individuals with Wilson's disease are usually diagnosed by examining serum and urine copper levels, plasma ceruloplasmin levels, and clinical manifestations. However, the relationship between serum and urine levels of copper and health effects is not known. Studies examining the possible correlation between blood levels or excreta levels of copper with effects would facilitate medical surveillance. Liver enzyme levels can indicate liver damage resulting from copper toxicity; however, these are not specific to copper-induced liver damage.

Absorption, Distribution, Metabolism, and Excretion. The absorption, distribution, metabolism, and excretion of copper administered orally have been studied in animals and, to some extent, in humans. Furthermore, alterations in copper absorption, distribution, and excretion have been studied in deficiency and toxicity states. Despite the information on copper absorption, there is very little information on differences between absorption rates of the various compounds and differences between the bioavailability of copper from food and water.

There is very limited information on copper absorption following inhalation exposure, and data on the absorption of copper through the skin are limited. Further studies in animals on the rate and extent of copper absorption following exposure from both the inhalation route and the dermal route would more fully characterize copper toxicokinetics in animals and by extrapolation in humans.

Comparative Toxicokinetics. The metabolism of copper has been studied in rats, pigs, hamsters, and humans. However, there are no comparative studies on the effects of high copper intakes on the distribution of copper in the body or the development of tolerance to continued high intakes of copper.

Furthermore, the animal species that might serve as the best model for extrapolating results to humans is not known.

Children's Susceptibility. There are some data on the toxicity of copper in infants and children. Severe liver damage has been reported in infants and children. These effects are typically clustered in geographically regions and have been grouped into two syndromes: Indian childhood cirrhosis and idiopathic copper toxicosis. Both of these syndromes have been connected to elevated copper intakes and are believed to have a genetic component. Very high levels of copper are found in the livers of affected children, suggesting that the mechanism of action is related to impaired copper efflux. Additional studies are needed to determine the mechanism of toxicity and to ascertain copper's role in the observed effects.

Physical and Chemical Properties. In general, the available data on the physical and chemical properties of elemental copper and copper sulfate are sufficient for estimating their environmental fate. In general, experimental confirmation is required for predicting copper's fate in the environment. The factors which determine the copper species present or the material to which copper may be bound and the strength of the binding can be site-specific. If the level of detail requires knowledge of, for example, the percentage of copper associated with iron oxides or that which is easily exchangeable, experimental confirmation is necessary.

Production, Import/Export, Use, Release, and Disposal. Information on the production, use, release, and disposal of metallic copper and copper sulfate is generally available. These two forms of copper account for most of the copper used. This information is tabulated by the U.S. Geological Survey every year in the Minerals Yearbook and predictions of future trends in production and use are available. Information on the future of copper demand and implications on copper recycling and production are also available (Ciacci et al. 2020; Schipper et al. 2018). Such information is not available for other copper compounds. The major uses of copper and where these uses occur (e.g., the home, workplace, etc.) is also available.

Environmental Fate. Reliable information on how copper and its compounds partition in the environment (i.e., to soil and sediment), and the type of transformations that occur in different media is extensively available. Data on its transport in the environment is also reliable. Although information on the fate of copper in air, water, and soil is available, the fate of copper is both species- and site-specific. Information concerning the forms of copper (i.e., specific compound, to what it is bound or complexed, or, in the case of air, the particle size) or the lability of the copper in particular media is available from only a few studies. These are sufficient to identify numerous contributors to the fate of copper and its

compounds, but they are insufficiently comprehensive for developing accurate fate maps. In addition, studies of how fate data relate to human exposures, especially in regard to projecting copper toxicity in children, is inadequate.

Bioavailability from Environmental Media. Copper is found in food, water, ambient air, and soil. The bioavailability of copper from food and water has been investigated in animals and humans. Studies on the bioavailability of copper from soil and ambient air would be useful in assessing potential toxicity to people living near a hazardous waste site. The form and lability of copper in the environment is known in only a few site-specific cases that do not include hazardous waste sites. More information on the forms of copper found at industrial sites and hazardous waste sites would be useful. Monitoring groundwater near industries that use highly acid, copper-containing solutions, such as electroplating, electrowinning, and ore leaching industries, is important for the protection of human populations at risk of exposure to their highly mobile and highly bioavailable copper.

Food Chain Bioaccumulation. Because copper occurs in different forms in the environment, its bioaccumulation is expected to vary according to site and species. Data are available on the bioconcentration of copper in aquatic organisms, plants, and animals, as well as biomagnification in food chains. This information is useful in assessing the potential for exposure from ingesting food originating from contaminated areas. However, little information is available on the potential for intoxication from foodstuffs from apparently nonpolluted areas or where they may have accumulated toxic levels of copper through biomagnification resulting from foraging in polluted areas.

Exposure Levels in Environmental Media. Data are available regarding the concentrations of copper in environmental media, including the concentration of copper in soil at some hazardous waste sites. Since copper is naturally present in soil, trace quantitative analytical and statistical techniques can be used to determine whether the copper found at these sites is elevated above normal levels. Monitoring data are reasonably current and human intake of copper from food, water, and air can be estimated.

Exposure Levels in Humans. There are reasonably current data on levels of copper in human tissue and human milk. However, few studies address specific U.S. populations living around hazardous waste sites. There are some quantitative data relating occupation, level, and route of exposure to the form of copper to which people are exposed. There is some limited information correlating copper concentration and form to body burden in the general population. However, more information is needed for occupational and other at-risk populations.

Exposures of Children. Data on copper intake in infants and children is generally up to date. Information on copper intake by infants from human milk also is available. Exposure of children to copper in drinking water has been assessed and methods to decrease this exposure have been identified and implemented. However, only limited information on inhalation is available. Some information on exposure of children to copper near mining, smelting, refining, manufacture facilities, waste sites, and other hazardous sites is available, but not for U.S. populations. This information is needed to better estimate exposures of children in U.S. populations living near these facilities and sites. The use of copper concentrations in toenails and hair has been investigated as a surrogate measure of copper exposure in children and adults, and more research into establishing the validity of these surrogates is underway.

6.3 ONGOING STUDIES

No ongoing studies were identified for copper.

CHAPTER 7. REGULATIONS AND GUIDELINES

Pertinent international and national regulations, advisories, and guidelines regarding copper and copper compounds in air, water, and other media are summarized in Table 7-1. This table is not an exhaustive list, and current regulations should be verified by the appropriate regulatory agency.

ATSDR develops MRLs, which are substance-specific guidelines intended to serve as screening levels by ATSDR health assessors and other responders to identify contaminants and potential health effects that may be of concern at hazardous waste sites. See Section 1.3 and Appendix A for detailed information on the MRLs for copper.

Table 7-1. Regulations and Guidelines Applicable to Copper and Copper Sulfate

Agency	Description	Information	Reference
	Air		
EPA	RfC	Not evaluated	IRIS 1988
WHO	Air quality guidelines	No data	WHO 2010
	Water & Food		
EPA	Drinking water standards and health advisories		EPA 2018
	MCLG	1.3 mg/L	
	MCL or TT	TT[a]	
	National primary drinking water regulations		EPA 2009a
	Treatment Technique Action Level	1.3 mg/L	
	Public Health Goal	1.3 mg/L	
	National secondary drinking water regulations[b]	1.0 mg/L	EPA 2018
	RfD	Not evaluated	EPA 2018
	Groundwater monitoring		EPA 2010
	PQL	0.05 mg/L	
	Clean Water Act – Designation of copper sulfate as a hazardous substance		EPA 2019 40CFR117.3
	RQ	10 lbs.	
	RQ (ammoniated)	100 lbs.	
	National Recommended Water Quality Human Health Criteria		EPA 2002
	Consumption of water and organisms	1300 µg/L	
	Consumption of organism only	No data	
	National Recommended Aquatic Life Criteria		EPA 2007
	Saltwater CMC (acute)	4.8 µg/L	
	Saltwater CCC (chronic)	3.1 µg/L	
	Freshwater CMC	No data	
	Freshwater CCC	No data	
WHO	Drinking water quality guidelines		WHO 2017
	Contaminants from pipes and fittings	2 mg/L (2000 µg/L)	
DOT	Marine pollutant – copper metal powder and copper sulfate, anhydrous, hydrates		DOT 2000 49CFR172.101

Table 7-1. Regulations and Guidelines Applicable to Copper and Copper Sulfate

Agency	Description	Information	Reference
FDA	Allowable level of copper in bottled water	1.0 mg/L	FDA 2019c 21CFR165.110
	Color additives exempt from certification-copper powder for use in externally applied drugs	not less than 95%	FDA 2019b 21CFR73.1647
	Direct food substance affirmed as generally recognized as safe when used as a nutrient supplement or as a processing aid		FDA 2019a 21CFR184
	Copper sulfate		
	Copper gluconate		
	Cancer		
HHS	Carcinogenicity classification	No data	NTP 2016
EPA	Carcinogenicity classification		IRIS 1988
	Copper – Oral exposure	D[c]	
	Copper – Inhalation exposure	D	
IARC	Carcinogenicity classification		IARC 2020
	Copper 8-hydroxyquinoline	Group 3[d]	
	Occupational		
ACGIH	TLV (8-hour TWA)		ACGIH 2018
	Copper (dust and mists, as Cu)	1 mg/m^3	
	Copper fume (as Cu)	0.2 mg/m^3	
OSHA	PEL (8-hour TWA for general industry)		OSHA 2020a 29CFR1910.1000
	Copper dusts and mists	1 mg/m^3	
	Copper fume	0.1 mg/m^3	
	PEL (8-hour TWA for construction industry)		OSHA 2020c 29 CFR 1926.55
	Copper dusts and mists	1 mg/m^3	
	Copper fume	0.1 mg/m^3	
	PEL (8-hour TWA for shipyard industry)		OSHA 2020b 29 CFR 1915.1000
	Copper dusts and mists	1 mg/m^3	
	Copper fume	0.1 mg/m^3	
NIOSH	REL (up to 10-hour TWA)		NIOSH 2014a; 2014b
	Copper (dust and mists, as Cu)	1 mg/m^3	
	Copper fume (as Cu)	0.1 mg/m^3	
	IDLH		
	Dusts and mists	100 mg Cu/m^3	
	Emergency Criteria		
AIHA	ERPGs	No data	AIHA 2016
EPA	AEGLs	No data	AEGLs 2018
DOE	PACs-air		DOE 2018
	Copper		
	PAC-1	3 mg/m^3	
	PAC-2	33 mg/m^3	
	PAC-3	200 mg/m^3	
	Copper sulfate		
	PAC-1	7.5 mg/m^3	
	PAC-2	9.9 mg/m^3	
	PAC-3	59 mg/m^3	
	Copper (II) chloride		
	PAC-1	6.3 mg/m^3	
	PAC-2	69 mg/m^3	
	PAC-3	420 mg/m^3	

Table 7-1. Regulations and Guidelines Applicable to Copper and Copper Sulfate

Agency	Description	Information	Reference
	Other		
DOT	RQ for hazardous substances other than radionuclides		DOT 2000 49CFR172.101
	Copper[e]	5,000 lb (2270 kg)	
	Copper chloride[f]	10 lb (4.54 kg)	
	Cupric sulfate	10 lb (4.54 kg)	
	Quantity limitations for hazardous materials		
	Copper based pesticides, liquid, flammable, toxic, flash point less than 23 degrees C (UN2776, packing group I)		
	Passenger aircraft/rail	Forbidden	
	Cargo aircraft only	30 L	
	Copper based pesticides, liquid, flammable, toxic, flash point less than 23 degrees C (UN2776, packing group II)		
	Passenger aircraft/rail	1 L	
	Cargo aircraft only	60 L	
	Copper based pesticides, liquid, toxic (UN3010, packing group I)		
	Passenger aircraft/rail	1 L	
	Cargo aircraft only	30 L	
	Copper based pesticides, liquid, toxic (UN3010, packing group II)		
	Passenger aircraft/rail	5 L	
	Cargo aircraft only	60L	
	Copper based pesticides, liquid, toxic (UN3010, packing group III)		
	Passenger aircraft/rail	60 L	
	Cargo aircraft only	220L	
	Copper based pesticides, liquid, toxic, flammable, flash point not less than 23 degrees C (UN3009, packing group I)		
	Passenger aircraft/rail	1 L	
	Cargo aircraft only	30 L	
	Copper based pesticides, liquid, toxic, flammable, flash point not less than 23 degrees C (UN3009, packing group II)		
	Passenger aircraft/rail	5 L	
	Cargo aircraft only	60 L	
	Copper based pesticides, liquid, toxic, flammable, flash point not less than 23 degrees C (UN3009, packing group III)		
	Passenger aircraft/rail	60 L	
	Cargo aircraft only	220 L	
	Copper based pesticides, solid, toxic (UN2775, packing group I)		
	Passenger aircraft/rail	5 kg	
	Cargo aircraft only	50 kg	
	Copper based pesticides, solid, toxic (UN2775, packing group II)		
	Passenger aircraft/rail	25 kg	
	Cargo aircraft only	100 kg	

Table 7-1. Regulations and Guidelines Applicable to Copper and Copper Sulfate

Agency	Description	Information	Reference
	Copper based pesticides, solid, toxic (UN2775, packing group III)		
	Passenger aircraft/rail	100 kg	
	Cargo aircraft only	200 kg	
	Copper chloride (UN2802, packing group III)	25 kg	
	Passenger aircraft/rail	100 kg	
	Cargo aircraft only		
FDA	Color additives exempt from certification – copper powder for use in cosmetics		FDA 2019a 21 CFR 73.2647

[a]The MCL is the TT, which is a required process intended to reduce the level of a contaminant in drinking water; the action level for copper is 1.3 mg/L. If more than 10 percent of tap water samples exceed the action level, water systems must take additional steps.
[b]National Secondary Drinking Water Regulations are contaminants tested on voluntary basis. The levels indicated may cause water to appear cloudy or colored, or to taste or smell, however, it is safe to drink.
[c]D: not classified. There are no human data, inadequate animal data from assays of copper compounds, and equivocal mutagenicity data.
[d]Group 3: Not classifiable as to its carcinogenicity to humans.
[e]The RQ for these hazardous substances is limited to those pieces of the metal having a diameter smaller than 100 micrometers (0.004 inches)
[f]Indicates that the name was added by PHMSA because (1) the name is a synonym for a specific hazardous substance and (2) the name appears in the Hazardous Materials Table as a proper shipping name.

ACGIH = American Conference of Governmental Industrial Hygienists; AEGL = acute exposure guideline levels; AIHA = American Industrial Hygiene Association; CCC = Criterion Continuous Concentration; CFR = Code of Federal Regulations; CMC = Continuous Maximum Concentration; HHS = Department of Health and Human Services; DOE = Department of Energy; DOT = Department of Transportation; EPA = Environmental Protection Agency; ERPG = emergency response planning guidelines; FDA = Food and Drug Administration; GRAS = Generally Recognized As Safe; IARC = International Agency for Research on Cancer; IDLH = Immediately Dangerous to Life of Health; IRIS = Integrated Risk Information System; MCL = maximum contaminant level; MCLG = maximum contaminant level goal; NIOSH = National Institute for Occupational Safety and Health; NTP = National Toxicology Program; OSHA = Occupational Safety and Health Administration; PAC = Protective Action Criteria; PEL = permissible exposure limit; PQL = possible quantitation limit; REL = recommended exposure limit; RfC = inhalation reference concentration; RfD = oral reference dose; RQ = reportable quantity; TLV = threshold limit values; TT = treatment technique; TWA = time-weighted average; WHO = World Health Organization

CHAPTER 8. REFERENCES

AAR. 1994. Emergency handling of hazardous materials in surface transportation. Washington, DC: Association of American Railroads, Bureau of Explosives.

Abbott SM, Malkani RG, Zee PC. 2018. Circadian disruption and human health: A bidirectional relationship. Eur J Neurosci. 51: 567-583. 10.111/ejn.14298.

Abe M, Usuda K, Hayashi S, et al. 2008. Carcinogenic risk of copper gluconate evaluated by a rat medium-term liver carcinogenicity bioassay protocol. Arch. Toxicol. 82(8):563-571. 10.1007/s00204-008-0294-x.

Abuja PM, Albertini R. 2001. Methods for monitoring oxidative stress, lipid peroxidation and oxidation resistance of lipoproteins. Clin Chim Acta 306(1-2):1-17. 10.1016/s0009-8981(01)00393-x.

ACGIH. 2018. TLVs and BEIs based on the documentation of the threshold limit values for chemical substances and physical agents and biological exposure indices. Cincinnati, OH: American Conference of Governmental Industrial Hygienists.

AEGLs. 2018. Compiled AEGL values. U.S. Environmental Protection Agency. https://www.epa.gov/sites/production/files/2018-08/documents/compiled_aegls_update_27jul2018.pdf.

Agarwal K, Sharma A, Talukder G. 1990. Clastogenic effects of copper sulfate on the bone marrow chromosomes of mice in vivo. Mutat. Res. 243(1):1-6. https://doi.org/10.1016/0165-7992(90)90115-z.

Agilent Technologies. 2012. Handbook of hyphenated ICP-MS applications. 2nd edition. https://www.ohsu.edu/sites/default/files/2018-08/Handbook-of-Hyphenated-ICP-MS-Applications.pdf.

Ahasan HA, Chowdhury MA, Azhar MA, et al. 1994. Copper sulphate poisoning. Trop Doct 24(2):52-53. 10.1177/004947559402400203.

AIHA. 2016. Current ERPG Values (2016). 2016 ERPG/WEEL Handbook. American Industrial Hygiene Association. https://www.btfire.org/ftp/Documents/2016%20ERPG%20Table.pdf.

Akintonwa A, Mabadeje AFB, Odutola TA. 1989. Fatal poisonings by copper sulfate ingested from "spiritual water". Vet. Hum. Toxicol 31(5):453-454.

Alarifi S, Ali D, Verma A, et al. 2013. Cytotoxicity and genotoxicity of copper oxide nanoparticles in human skin keratinocytes cells. Int J Toxicol 32(4):296-307. 10.1177/1091581813487563.

Alharbi B, Fadda L, Ali HM. 2019. Evaluation of the renoprotective effect of nano turmeric against toxic dose of copper sulfate: Role of vascular cell adhesion molecule-1, kidney injury molecule-1, and signal transducer and activator of transcription 3 protein expressions. J. Biochem. Mol. Toxicol. 33(2):e22243. 10.1002/jbt.22243.

Alhusaini A, Hasan IH, Aldowsari N, et al. 2018a. Prophylactic administration of nanocurcumin abates the incidence of liver toxicity induced by an overdose of copper sulfate: Role of CYP4502E1, NF-kappaB and bax expressions. Dose Response 16(4):1559325818816284. 10.1177/1559325818816284.

Alhusaini A, Fadda L, Hassan I, et al. 2018b. Liposomal curcumin attenuates the incidence of oxidative stress, inflammation, and DNA damage induced by copper sulfate in rat liver. Dose Response 16(3):1559325818790869. 10.1177/1559325818790869.

Allen SK, Allen JM, Lucas S. 1996. Dissolved metal concentrations in surface waters from west-central Indiana contaminated with acidic mine drainage. Bull. Environ. Contam. Toxicol. 56(2):240-243. 10.1007/s001289900036.

Allen-Gil SM, Gubala CP, Landers DH, et al. 1997. Heavy metal accumulation in sediment and freshwater fish in US Arctic lakes. Environmental Toxicology Chemistry: An International Journal 16(4):733-741. https://doi.org/10.1002/etc.5620160418.

Ameh T, Sayes CM. 2019. The potential exposure and hazards of copper nanoparticles: A review. Environ Toxicol Pharmacol 71:103220. 10.1016/j.etap.2019.103220.

Amrhein C, Strong JE, Mosher PA. 1992. Effect of deicing salts on metal and organic matter mobilization in roadside soils. Environmental Science Technology 26(4):703-78a006.

Anchordoquy JM, Anchordoquy JP, Nikoloff N, et al. 2017. High copper concentrations produce genotoxicity and cytotoxicity in bovine cumulus cells. Environ Sci Pollut Res Int 24(24):20041-20049. 10.1007/s11356-017-9683-0.

Anderson JR, Aggett FJ, Buseck PR, et al. 1988. Chemistry of individual aerosol particles from Chandler, Arizona, an arid urban environment. Environ. Sci. Technol. 22(7):811-818. 10.1021/es00172a011.

Annibaldi A, Truzzi C, Illuminati S, et al. 2007. Determination of water-soluble and insoluble (dilute-HCl-extractable) fractions of Cd, Pb and Cu in Antarctic aerosol by square wave anodic stripping voltammetry: distribution and summer seasonal evolution at Terra Nova Bay (Victoria Land). Anal Bioanal Chem 387(3):977-998. 10.1007/s00216-006-0994-0.

Anreddy RNR. 2018. Copper oxide nanoparticles induces oxidative stress and liver toxicity in rats following oral exposure. Toxicol Rep 5:903-904. 10.1016/j.toxrep.2018.08.022.

Arafa MH, Amin DM, Samir GM, et al. 2019. Protective effects of tribulus terrestris extract and angiotensin blockers on testis steroidogenesis in copper overloaded rats. Ecotoxicol. Environ. Saf. 178:113-122. 10.1016/j.ecoenv.2019.04.012.

Araya M, Koletzko B, Uauy R. 2003d. Copper deficiency and excess in infancy: Developing a research agenda. J Pediatr Gastroenterol Nutr 37(4):422-429. 10.1097/00005176-200310000-00005.

Araya M, Peña C, Pizarro F, et al. 2003a. Gastric response to acute copper exposure. Science of The Total Environment 303(3):253-257. 10.1016/s0048-9697(02)00495-3.

Araya M, Olivares M, Pizarro F, et al. 2003b. Gastrointestinal symptoms and blood indicators of copper load in apparently healthy adults undergoing controlled copper exposure. Am J Clin Nutr 77(3):646-650. 10.1093/ajcn/77.3.646.

Araya M, Olivares M, Pizarro F, et al. 2004. Community-based randomized double-blind study of gastrointestinal effects and copper exposure in drinking water. Environ. Health Perspect. 112(10):1068-1073. 10.1289/ehp.6913.

Araya M, McGoldrick MC, Klevay LM, et al. 2001. Determination of an acute no-observed-adverse-effect level (NOAEL) for copper in water. Regul Toxicol Pharmacol 34(2):137-145. 10.1006/rtph.2001.1492.

Araya M, Nunez H, Pavez L, et al. 2012. Administration of high doses of copper to capuchin monkeys does not cause liver damage but induces transcriptional activation of hepatic proliferative responses. J. Nutr. 142(2):233-237. 10.3945/jn.111.140103.

Araya M, Chen B, Klevay LM, et al. 2003c. Confirmation of an acute no-observed-adverse-effect and low-observed-adverse-effect level for copper in bottled drinking water in a multi-site international study. Regul Toxicol Pharmacol 38(3):389-399. 10.1016/j.yrtph.2003.08.001.

Arcega-Cabrera F, Fargher LF. 2016. Education, fish consumption, well water, chicken coops, and cooking fires: Using biogeochemistry and ethnography to study exposure of children from Yucatan, Mexico to metals and arsenic. Sci. Total. Environ 568:75-82. 10.1016/j.scitotenv.2016.05.209.

Arendsen LP, Thakar R, Sultan AH. 2019. The use of copper as an antimicrobial agent in health care, including obstetrics and gynecology. Clin Microbiol Rev 32(4):e00125-e00128. 10.1128/CMR.00125-18.

Armstrong CW, Moore LW, Jr., Hackler RL, et al. 1983. An outbreak of metal fume fever. Diagnostic use of urinary copper and zinc determinations. J Occup Med 25(12):886-888.

Askergren A, Mellgren M. 1975. Changes in the nasal mucosa after exposure to copper salt dust. A preliminary report. Scand J Work Environ Health 1(1):45-49. 10.5271/sjweh.2861.

ATSDR. 1989. Decision guide for identifying substance-specific data needs related to toxicological profiles. Atlanta, GA: Agency for Toxic Substances and Disease Registry, Division of Toxicology

ATSDR. 2004. Toxicological profile for copper. Atlanta, GA: Agency for Toxic Substances and Disease Registry, Public Health Service, U.S. Department of Health and Human Servi2.pdf.

8. REFERENCES

ATSDR. 2019. Copper. Full SPL data. Substance priority list (SPL) resource page. Agency for Toxic Substances and Disease Registry, Centers for Disease Control and Prevention. https://www.atsdr.cdc.gov/SPL/resources/index.html. February 20, 2020.

Aulenbach DB, Meyer MA, Beckwith E, et al. 1987. Removal of heavy metals in POTW. Environ Prog 6(2):91-60210.

Aulerich RJ, Ringer RK, Bleavins MR, et al. 1982. Effects of supplemental dietary copper on growth, reproductive performance and kit survival of standard dark mink and the acute toxicity of copper to mink. J. Anim. Sci. 55(2):337-343. 10.2527/jas1982.552337x.

Babaei H, Abshenas J. 2013. Zinc therapy improves adverse effects of long term administration of copper on epididymal sperm quality of rats. Iran J Reprod Med 11(7):577-582.

Babaei H, Roshangar L, Sakhaee E, et al. 2012. Ultrastructural and morphometrical changes of mice ovaries following experimentally induced copper poisoning. Iran Red Crescent Med J 14(9):558-568.

Badaloni C, Cesaroni G, Cerza F, et al. 2017. Effects of long-term exposure to particulate matter and metal components on mortality in the Rome longitudinal study. Environ. Int. 109:146-154. 10.1016/j.envint.2017.09.005.

Badri MA, Aston SR. 1983. Observations on heavy metal geochemical associations in polluted and non-polluted estuarine sediments. Environmental Pollution Series B, Chemical Physical 6(3):181-1033-2.

Balogh S. 1996. The fate of metals in sewage sludge incinerators. Water, Air, Soil Pollution 91(3-4):249-266261.

Banavi P, Sadeghi E, Garavand F, et al. 2020. Release behavior of metals from tin-lined copper cookware into food simulants during cooking and cold storage. Environ Sci Pollut Res Int 27(31):38591-38601. 10.1007/s11356-020-09970-z.

Barranco VP. 1972. Eczematous dermatitis caused by internal exposure to copper. Arch Dermatol 106(3):386-387.

Barrie LA, Lindberg S, Chan W, et al. 1987. On the concentration of trace metals in precipitation. Atmospheric Environment 21(5):1133-11240-X.

Behera C, Rautji R, Dogra TD. 2007. An unusual suicide with parenteral copper sulphate poisoning: a case report. Med Sci Law 47(4):357-358. 10.1258/rsmmsl.47.4.357.

Behzadfar L, Abdollahi M, Sabzevari O, et al. 2017. Potentiating role of copper on spatial memory deficit induced by beta amyloid and evaluation of mitochondrial function markers in the hippocampus of rats. Metallomics 9(7):969-980. 10.1039/c7mt00075h.

Benson NU, Anake WU, Adedapo AE, et al. 2017. Toxic metals in cigarettes and human health risk assessment associated with inhalation exposure. Environ. Monit. Assess 189(12):6348-x.

Bentur Y, Koren G, McGuigan M, et al. 1988. An unusual skin exposure to copper; clinical and pharmacokinetic evaluation. J Toxicol Clin Toxicol 26(5-6):371-380. 10.1080/15563658809167101.

Beveridge SJ, Boettcher B, Walker WR, et al. 1984. Biodistribution of 64Cu in rats after topical application of two lipophilic anti-inflammatory Cu(II) formulations. Agents Actions 14(2):291-295. 10.1007/BF01966655.

Beyer WN, Cromartie EJ. 1987. A survey of Pb, Cu, Zn, Cd, Cr, As, and Se in earthworms and soil from diverse sites. Environmental Monitoring Assessment 8(1):27-96605.

Bhandari P, Andrews PL. 1991. Preliminary evidence for the involvement of the putative 5-HT4 receptor in zacopride- and copper sulphate-induced vomiting in the ferret. Eur. J. Pharmacol. 2 04(3):273-280. 10.1016/0014-2999(91)90852-h.

Bhave SA, Pandit AN, Tanner MS. 1987. Comparison of feeding history of children with Indian childhood cirrhosis and paired controls. J Pediatr Gastroenterol Nutr 6(4):562-567. 10.1097/00005176-198707000-00013.

Bhave SA, Pandit AN, Pradhan AM, et al. 1982. Liver disease in India. Arch Dis Child 57(12):922-928. 10.1136/adc.57.12.922.

Bhunya SP, Pati PC. 1987. Genotoxicity of an inorganic pesticide, copper sulfate in mouse in vivo test system. Cytologia (Tokyo) 52:801-82.801.

Bhunya SP, Jena GB. 1996. Clastogenic effect of copper sulphate in chick in vivo test system. Mutat. Res. 367(2):57-061-5.

Bilal M, Ali Shah J, Ashfaq T, et al. 2013. Waste biomass adsorbents for copper removal from industrial wastewaster - a review. J Hazard Mater 263(2):322-37.071.

Blanusa M, Ivicic N, Simeon V. 1990. Lead, iron, copper, zinc and ash in deciduous teeth in relation to age and distance from a lead smelter. Bull. Environ. Contam. Toxicol. 45(4):478-485. 10.1007/BF01700618.

Blevins RD, Pancorbo OC. 1986. Metal concentrations in muscle of fish from aquatic systems in East Tennessee, USA. Water, Air, Soil Pollution 29(4):361-383443.

Bolan NS, Khan MA, Donaldson J, et al. 2003. Distribution and bioavailability of copper in farm effluent. Sci. Total. Environ 309(1-3):225-236. 10.1016/S0048-9697(03)00052-4.

Boogaard H, Fischer PH, Janssen NA, et al. 2013. Respiratory effects of a reduction in outdoor air pollution concentrations. Epidemiology 24(5):753-761. 10.1097/EDE.0b013e31829e1639.

Bopp R, Simpson H, Chillrud S, et al. 1993. Sediment-derived chronologies of persistent contaminants in Jamaica Bay, New York. Estuaries 16(3):608-652798.

Borak J, Cohen H, Hethmon T. 2000. Copper exposure and metal fume fever: Lack of evidence for acausal relationship. Am Ind Hyg Assoc J 61(6):832-884594.

Bost M, Houdart S, Oberli M, et al. 2016. Dietary copper and human health: Current evidence and unresolved issues. J Trace Elem Med Biol 35:107-115. 10.1016/j.jtemb.2016.02.006.

Boullata J, Muthukumaran G, Piarulli A, et al. 2017. Oral copper absorption in men with morbid obesity. J Trace Elem Med Biol 44:146-150. 10.1016/j.jtemb.2017.07.005.

Boyden E, Potter VE, Elvehjem CA. 1938. Effect of feeding high levels of copper to albino rats. The Journal of Nutrition 15(4):397-44.397.

Bradley RW, Morris JR. 1986. Heavy metals in fish from a series of metal-contaminated lakes near Sudbury, Ontario. Water, Air, Soil Pollution 27(3-4):341-349416.

Breault R, Colman J, Aiken G, et al. 1996. Copper speciation and binding by organic matter in copper-contaminated streamwater. Environmental Science Technology 30(12):3477-3486.

Breslin V. 1999. Retention of metals in agricultural soils after amending with MSW and MSW-biosolids compost. Water, Air, Soil pollution 109(1-4):163-131978.

Brewer G. 2010. Copper toxicity in the general population. Clin Neurophysiol 121:459-460. 10.1016/j.clinph.2009.12.015.

Brewer GJ, Askari F, Lorincz MT, et al. 2006. Treatment of Wilson disease with ammonium tetrathiomolybdate: IV. Comparison of tetrathiomolybdate and trientine in a double-blind study of treatment of the neurologic presentation of Wilson disease. Arch Neurol 63(4):521-527. 10.1001/archneur.63.4.521.

Brown KW, Thomas JC, Slowey JF. 1983. The movement of metals applied to soils in sewage effluent. Water, Air, Soil Pollution 19(1):43-76794.

Bruce BW, McMahon PB. 1996. Shallow ground-water quality beneath a major urban center: Denver, Colorado, USA. Journal of Hydrology 186(1-4):129-1031-4.

Buchanan SD, Diseker RA, 3rd, Sinks T, et al. 1999. Copper in drinking water, Nebraska, 1994. Int J Occup Environ Health 5(4):256-261. 10.1179/oeh.1999.5.4.256.

Buchholz B, Landsberger S. 1995. Leaching dynamics studies of municipal solid waste incinerator ash. Journal of the Air Waste Management Association 45(8):579-567388.

Budavari S, O'Neil M, Smith A, et al. 2001. The Merck index: an encyclopedia of chemicals, drugs and biologicals. Whitehouse Station, NJ: Merck & Co. Inc.:440, 462.

Buseck PR, Pósfai M. 1999. Airborne minerals and related aerosol particles: effects on climate and the environment. Proc. Natl. Acad. Sci. U. S. A 96(7):3372-3379. 10.1073/pnas.96.7.3372.

8. REFERENCES

Bush J, Mahoney J, Markowitz H, et al. 1955. Studies on copper metabolism.XVI. Radioactive copper studies in normal subjects and in patients with hepatolenticular degeneration. J Clin Invest 34:1766-1703232.

Cadle SH, Mulawa PA, Hunsanger EC, et al. 1999. Composition of light-duty motor vehicle exhaust particulate matter in the Denver, Colorado Area. Environ Sci Technol 33(14):2328-2339. 10.1021/es9810843.

Cagnarini C, Lofts S, D'Acqui LP, et al. 2021. Modelling of long-term Zn, Cu, Cd and Pb dynamics from soils fertilised with organic amendments. SOIL 7(1):107-123. 10.5194/soil-7-107-2021.

Cai M, McBride MB, Li K. 2016. Bioaccesibility of Ba, Cu, Pb, and Zn in urban garden and orchard soils. Environ Pollut 208:145-19.050.

Cai R, Zhang C, Ding W, et al. 2009. Corneal melting induced by a presumed copper-containing foreign body. Clin Exp Ophthalmol 37(3):328-330. 10.1111/j.1442-9071.2009.02027.x.

Caicedo M, Jacobs J, Reddy AH, NJ. 2008. Analysis of metal ion-induced DNA damage, apoptosis, and necrosis in human (Jurkat) T-cells demonstrates Ni2+, and V3+, are more toxic than other metals : Al3+, Be2+, Co2+, Cr3+, Cu2+, Fe3+, Mo5+, Nb5+, Zr2+. J Biomed Mater Res A 86(4):905-931789.

Calabrese E, Moore G. 1979. Can elevated levels of copper in drinking water precipitate acute hemolysis in G-6-PD deficient individuals. MedHypotheses 5(4):493-4116-6.

Campbell K. 1994. Concentrations of heavy metals associated with urban runoff in fish living in stormwater treatment ponds. Archives of Environmental Contamination Toxicology 27(3):352-313171.

Cao ZH, Hu ZY. 2000. Copper contamination in paddy soils irrigated with wastewater. Chemosphere 41(1-2):3-6. 10.1016/s0045-6535(99)00383-5.

Cao Y, Conklin M, Betterton E. 1995. Competitive complexation of trace metals with dissolved humic acid. Environ. Health Perspect. 103(Suppl 1):29-3s129.

CDC. 2016. National Health and Nutrition Examination Survey. NHANES 2013-2014 Laboratory Data. Copper, Selenium & Zinc – Serum. https://wwwn.cdc.gov/nchs/nhanes/search/datapage.aspx?Component=Laboratory&CycleBeginYear=2013. October 21, 2020.

CDC. 2018. National Health and Nutrition Examination Survey. NHANES 2015-2016 Laboratory Data. Copper, Selenium & Zinc – Serum. https://wwwn.cdc.gov/nchs/nhanes/search/datapage.aspx?Component=Laboratory&CycleBeginYear=2015. October 21, 2020.

CEIDARS. 2000. Chemical speciation. California Emission Inventory and Reporting System.

Chan W, Tang AJ, Chung D, et al. 1986. Concentration and deposition of trace metals in Ontario-1982. Water, Air, Soil Pollution 29(4):373-383444.

Chang Y-N, Zhang M, Xia L, et al. 2012. The toxic effects and mechanisms of CuO and ZnO nanoparticles. Materials 5(12):2850-2871. 10.3390/ma5122850.

Chen LC, Lippmann M. 2009. Effects of metals within ambient air particulate matter (PM) on human health. Inhal Toxicol 21(1):1-31. 10.1080/08958370802105405.

Chen R, Wei L, Huang H. 1993. Mortality from lung cancer among copper miners. Br J Ind Med 50(6):505-56.505.

Chen R, Wei L, Chen R-L. 1995. Lung cancer mortality update and prevalence of smoking among copper miners and smelters. Scand J Work Environ Health 21(6):513-5eh.68.

Chen M, Ogunseitan OA, Wang J, et al. 2016. Evolution of electronic waste toxicity: Trends in innovation and regulation. Environ. Int. 89-90:147-154. 10.1016/j.envint.2016.01.022.

Chen Z, Meng H, Xing G, et al. 2006. Acute toxicological effects of copper nanoparticles in vivo. Toxicol Lett 163(2):109-120. 10.1016/j.toxlet.2005.10.003.

Cheng S, Mao H, Ruan Y, et al. 2020. Copper changes intestinal microbiota of the cecum and rectum in female mice by 16S rRNA gene sequencing. Biol Trace Elem Res 193(2):445-455. 10.1007/s12011-019-01718-2.

Cheng TF, Choudhuri S, Muldoon-Jacobs K. 2012. Epigenetic targets of some toxicologically relevant metals: a review of the literature. Journal of Applied Toxicology 32: 643-653. 10.1002/jat.2717.

Chernenkov Y, Bochkova L, Kadymova L, et al. 2018. Copper concentration in the blood serum of low birth weight newborns. Biomedical and Pharmacology Journal 11(4):1807-18/1553.

Cho YS, Moon JM, Jeong YH, et al. 2018. Successful extracorporeal life support in respiratory failure after copper sulphate ingestion. Natl Med J India 31(2):83-85. 10.4103/0970-258X.253166.

Choi YK, Kim JM, Lee JE, et al. 2016. Association of maternal diet with zinc, copper, and iron concentrations in transitional human milk produced by Korean mothers. Clin Nutr Res 5(1):15-25. 10.7762/cnr.2016.5.1.15.

Cholewinska E, Ognik K, Fotschki B, et al. 2018. Comparison of the effect of dietary copper nanoparticles and one copper (II) salt on the copper biodistribution and gastrointestinal and hepatic morphology and function in a rat model. PLoS One 13(5):e0197083. 10.1371/journal.pone.0197083.

Christensen TH, Kjeldsen P, Albrechtsen HJ, et al. 1994. Attenuation of landfill leachate pollutants in aquifers. Critical Reviews in Environmental Science Technology 24(2):119-288463.

Chugh KS, Sakhuja V. 1979. Acute copper intoxication. Int J Artif Organs 2(4):181-182.

Chuttani HK, Gupta PS, Gulati S, et al. 1965. Acute copper sulfate poisoning. Am J Med 39(5):849-854. 10.1016/0002-9343(65)90105-1.

Ciacci L, Fishman T, Elshkaki A, et al. 2020. Exploring future copper demand, recycling and associated greenhouse gas emissions in the EU-28. Global Environ Change 63:102002093.

Clemens S. 2001. Molecular mechanisms of plant metal tolerance and homeostasis. Planta 212(4):475-486. 10.1007/s004250000458.

Clewell HJI, Andersen ME. 1985. Risk assessment extrapolations and physiological modeling.

Coale K, Bruland K. 1988. Copper complexation in the Northeast Pacific. Limnol. Oceanogr 33(5):1084-11.1084.

Coelho FC, Squitti R, Ventriglia M, Cerchiaro G, Daher JP, Rocha JG, Rongioletti MC, Moonen AC. 2020. Agricultural Use of Copper and Its Link to Alzheimer's Disease. Biomolecules, 10: 897. 10.3390/biom10060897

Cole RH, Frederick RE, Healy RP, et al. 1984. Preliminary findings of the priority pollutant monitoring project of the nationwide urban runoff program. Journal (Water Pollution Control Federation) 56(7):898-908.

Coleman M, Elder R, Basu P, et al. 1992. Trace metals in edible tissues of livestock and poultry. J AOAC Int 75(4):615-64.615.

Cordano A. 1998. Clinical manifestations of nutritional copper deficiency in infants and children. Am J Clin Nutr 67(5 Suppl):1012S-1016S. 10.1093/ajcn/67.5.1012S.

Crawford DW, Bonnevie NL, Wenning RJ. 1995. Sources of pollution and sediment contamination in Newark Bay, New Jersey. Ecotoxicol. Environ. Saf. 30(1):85-100. 10.1006/eesa.1995.1010.

Cui JZ, Lifeng. 2007. Metallurgical recovery of metals from electronic waste: A review. Journal of Hazardous Materials 158(2-3):228-22.001.

Cui X, Okayasu R. 2008. Arsenic accumulation, elimination, and interaction with copper, zinc and manganese in liver and kidney of rats. Food and Chemical Toxicology. 46:3646-3650. 10.1016/j.fct.2008.09.040

Cushing CA, Golden R, Lowney YW. 2007. Human Health risk evaluation of ACQ-treated wood. Hum Ecol Risk Assess 13(5):1014-1006173.

Cyr F, Mehra MC, Mallet VN. 1987. Leaching of chemical contaminants from a municipal landfill site. Bull. Environ. Contam. Toxicol. 38(5):775-782. 10.1007/BF01616700.

Danks DM. 1988. Copper deficiency in humans. Annu. Rev. Nutr. 8:235-257. 10.1146/annurev.nu.08.070188.001315.

Daskalakis KD, O'Connor TP. 1995. Distribution of chemical concentrations in US coastal and estuarine sediment. Mar Environ Res 40(4):381-3150-N.

Davenport WG. 2001. Copper Production. In: Buschow KHJ, Cahn RW, Flemings MC, et al., ed. Encyclopedia of materials: Science and technology. Oxford: Elsevier, 1671-1680.

Davidson CI, Wiersma GB, Brown KW, et al. 1985. Airborne trace elements in Great Smoky Mountains, Olympic, and Glacier National Parks. Environmental Science Technology 19(1):27-1a001.

Davies N, Campbell J. 1977. The effect of cadmium on intestinal copper absorption and binding in the rat. Life Sci. 20(6):955-9281-8.

Davies DJA, Bennett BG. 1985. Exposure of man to environmental copper—An exposure commitment assessment. Science of the Total Environment 46(1-4):215-2295-5.

Davies-Colley RJ, Nelson PO, Williamson KJ. 1984. Copper and cadmium uptake by estuarine sedimentary phases. Environmental Science Technology 18(7):491-45a002.

Davies-Colley RJ, Nelson PO, Williamson KJ. 1985. Sulfide control of cadmium and copper concentrations in anaerobic estuarine sediments. Mar Chem 16(2):173-1021-0.

Davis AP, Shokouhian M, Ni S. 2001. Loading estimates of lead, copper, cadmium, and zinc in urban runoff from specific sources. Chemosphere 44(5):997-1009. 10.1016/s0045-6535(00)00561-0.

De Craemer S, Croes K, van Larebeke N, et al. 2017. Metals, hormones and sexual maturation in Flemish adolescents in three cross-sectional studies (2002-2015). Environ. Int. 102:190-199. 10.1016/j.envint.2017.02.014.

De Jong WH, De Rijk E, Bonetto A, et al. 2019. Toxicity of copper oxide and basic copper carbonate nanoparticles after short-term oral exposure in rats. Nanotoxicology 13(1):50-72. 10.1080/17435390.2018.1530390.

De Olivera J, Bonfleur L, Dos Santos C, et al. 2012. Occupational genotoxicity among copper smelters. Toxicol Ind Health 28(9):789-722735.

De Vries DJ, Sewell RB, Beart PM. 1986. Effects of copper on dopaminergic function in the rat corpus striatum. Exp. Neurol. 91(3):546-558. 10.1016/0014-4886(86)90051-8.

Demerec M, Bertani G, Flint J. 1951. A survey of chemicals for mutagenic action on E. coli. Am. Nat. 85(821):119-136.

Denizeau F, Marion M. 1989. Genotoxic effects of heavy metals in rat hepatocytes. Cell Biol Toxicol 5(1):15-25. 10.1007/BF00141061.

Dieter HH, Schimmelpfennig W, Meyer E, et al. 1999. Early Childhood Cirrhoses (ECC) in Germany between 1982-1994 with special consideration of copper etiology. Eur J Med Res 4(6):233-242.

Diks DM, Allen HE. 1983. Correlation of copper distribution in a freshwater-sediment system to bioavailability. Bull. Environ. Contam. Toxicol. 30(1):37-43. 10.1007/BF01610096.

Dinu M, Moiseenko T, Baranov D. 2020. Snowpack as Indicators of Atmospheric Pollution: The Valday Upland. Atmosphere 11(5):462.

DOE. 2018. Table 2: Protective Action Criteria (PAC) Rev. 29a based on applicable 60-minute AEGLs, ERPGs, or TEELs. The chemicals are listed in alphabetical order. June 2018.

Domellof M, Lonnerdal B, Dewey KG, et al. 2004. Iron, zinc, and copper concentrations in breast milk are independent of maternal mineral status. Am J Clin Nutr 79(1):111-115. 10.1093/ajcn/79.1.111.

Domellof M, Hernell O, Abrams SA, et al. 2009. Iron supplementation does not affect copper and zinc absorption in breastfed infants. Am J Clin Nutr 89(1):185-190. 10.3945/ajcn.2008.26887.

Domergue F, Vedy J. 1992. Mobility of heavy metals in soil profiles. Int J Environ Anal Chem 46(1-3):13-26993.

Donahue J. 1997. New ideas after five years of the lead and copper rule: A fresh look at the MCLG for copper. In: Lagos GE & Badilla-Ohlbaum R, ed. Advances in risk assessment of copper in the environment. Proceedings of the International Workshop "Risk Assessment of Copper in the Environment", Renaca, Chile, 7 - 9 May 1997. 265-272.

Donley SA, Ilagan BJ, Rim H, et al. 2002. Copper transport to mammary gland and milk during lactation in rats. Am. J. Physiol. Endocrinol. Metab. 283(4):E667-E675. 10.1152/ajpendo.00115.2002.

Donoso A, Cruces P, Camacho J, et al. 2007. Acute respiratory distress syndrome resulting from inhalation of powdered copper. Clin Toxicol (Phila) 45(6):714-716. 10.1080/15563650701438912.

Dörner K, Dziadzka S, Höhn A, et al. 1989. Longitudinal manganese and copper balances in young infants and preterm infants fed on breast-milk and adapted cow's milk formulas. Br. J. Nutr. 61(3):559-572. 10.1079/bjn19890143.

DOT. 2000. Hazardous materials table, special provisions, hazardous materials communications, emergency response information, training requirements, and security plans. Subpart B-Table of hazardous materials and special provisions. U.S. Department of Transportation. Code of Federal Regulations.

Dressler R, Storm G, Tzilkowski W, et al. 1986. Heavy metals in cottontail rabbits on mined lands treated with sewage sludge 1. J Environ Qual 15(3):278-20014x.

Drummond J, Aranyi C, Schiff L, et al. 1986. Comparative study of various methods used for determining health effects of inhaled sulfates. Environ Res 41(2):514-5146-3.

Du Y, Mou Y. 2019. The role of plasmapheresis in treating lethal cupric sulfate poisoning. Am J Med Sci 357(4):338-342. 10.1016/j.amjms.2018.11.014.

Duby P. 1980. Extractive metallurgy. In:ed. Kirk-Othmer encyclopedia of chemical technology. New York, NY: John Wiley and Sons, 739-767.

Durando J. 2005. Data review for acute oral toxicity testing. Washington, D.C.: U.S. Environmental Protection Agency.

Eary L, Mattigod S, Rai D, et al. 1990. Geochemical factors controlling the mobilization of inorganic constituents from fossil fuel combustion residues: I. Review of the major elements. J Environ Qual 19(2):188-20004x.

Eckel WP, Langley W. 1988. A background-based ranking technique for assessment of elemental enrichment in soils at hazardous waste sites. Superfund'88. Proceedings of the 9th National Conference. Nov. 28-30, 1988. Washington, DC:282-286.

Eckel WP, Jacob TA. 1988. Ambient levels of 24 dissolved metals in US surface and ground waters. Preprints of Papers Presented at National Meeting, Division of Water, Air and Waste Chemistry, American Chemical Society; (USA) 28:371-372.

Edwards M, Powers K, Hidmi L, et al. 2001. The role of pipe ageing in copper corrosion by-product release. Water Supply 1(3):25-32. 10.2166/ws.2001.0050.

Effler S, Litten S, Field S, et al. 1980. Whole lake responses to low level copper sulfate treatment. Water Res 14(10):1489-14015-9.

Eife R, Weiss M, Barros V, et al. 1999. Chronic poisoning by copper in tap water: I. Copper intoxications with predominantly gastrointestinal symptoms. Eur J Med Res 4(6):219-223.

Eisenberg M, Topping JJ. 1986. Trace metal residues in finfish from Maryland waters, 1978-1979. J Environ Sci Health B 21(1):87-102. 10.1080/03601238609372512.

El Bialy BE, Hamouda RA, Abd Eldaim MA, et al. 2020. Comparative toxicological effects of biologically and chemically synthesized copper oxide nanoparticles on mice. Int J Nanomedicine 15:3827-3842. 10.2147/IJN.S241922.

Elliott HA, Liberati MR, Huang CP. 1986. Competitive Adsorption of Heavy Metals by Soils 1. J Environ Qual 15(3):214-219.

EPA. 1979. Water-related environmental fate of 129 priority pollutants. Volume 1: Introduction and technical background, metals and inorganics, pesticides and PCBs. Washington, DC: Office of Water Planning and Standards, Office of Water and Waste Management, U.S. Environmental Protection Agency.

EPA. 1980. Chapter 29 Primary Copper Industry. Industrial process profiles for environmental use. Cincinnati, OH: U.S. Environmental Protection Agency.

EPA. 1981. Treatability manual. Volume 1. Treatability data Washington, DC: Office of Research and Development, U.S. Environmental Protection Agency.

EPA. 1984. Air quality data for metals 1977 through 1979 from the National Air Surveillance Networks. Research Triangle Park, NC: Environmental Monitoring Systems Laboratory, U.S. Environmental Protection Agency.

EPA. 1987. Assessment of copper as a potentially toxic air pollutant. U.S. Environmental Protection Agency. Fed Regist 52(35):5496-5499.

EPA. 1991. Maximum contaminated level, goals and national primary drinking-water regulation for lead and copper. Final rule. Fed Regist 56:438-447.

EPA. 1994a. Method 200.7: Determination of metals and trace elements in water and wastes by inductively coupled plasma-atomic emission spectrometry. Revision 4.4. Cincinnati, OH: Environmental Monitoring Systems Laboratory, Office of Research and Development, U.S. Environmental Protection Age4.pdf.

EPA. 1994b. Method 200.8: Determination of trace elements in waters and wastes by inductively coupled plasma – mass spectrometry. Revision 5.4. Cincinnati, OH: Environmental Monitoring Systems Laboratory, Office of Research and Development, U.S. Environmental Protection Agency. https://www.epa.gov/sites/production/files/2015-08/documents/method_200-8_rev_5-4_1994.pdf. October 21, 2020.

EPA. 1994c. Method 200.9: Determination of trace elements by stabilized temperature graphic furnace atomic absorption. Revision 2.2. Cincinnati, OH: Environmental Monitoring Systems Laboratory, Office of Research and Development, U.S. Environmental Protection Age4.pdf.

EPA. 1995. Effect of pH, DIC, orthophosphate and sulfate on drinking water cuprosolvency. Washington, DC: Office of Research and Development, U.S. Environmental Protection AgeQ.TXT.

EPA. 2002. National Recommended Water Quality Criteria: 2002. Human health criteria calculation matrix. Office of Water, U.S. Environmental Protection Agency.

EPA. 2003. Method 200.5: Determination of trace elements in drinking water by axially viewed inductively coupled plasma - atomic emission spectrometry. Revision 4.2. Cincinnati, OH: National Exposure Research Laboratory, Office of Research and Development, U.S. Environmental Protection Age3.pdf.

EPA. 2005. Toxic chemical release inventory reporting forms and instructions: Revised 2004 version. Section 313 of the Emergency Planning and Community Right-to-Know Act (Title III of the Superfund Amendments and Reauthorization Act of 1986). U.S. Environmental Protection Agency.

EPA. 2007. National recommended water quality criteria-Aquatic life criteria table. U.S. Environmental Protection Agency. https://www.epa.gov/wqc/national-recommended-water-quality-criteria-aquatic-life-criteria-table. October 21, 2020.

EPA. 2009a. National primary drinking water regulations. Office of Groundwater and Drinking Water, U.S. Environmental Protection Agee.pdf.

EPA. 2009b. Reregistration eligibility decision (RED) for coppers. Prevention, Pesticides and Toxic Substances, U.S. Environmental Protection Agency.

EPA. 2010. Lead and copper rule: Monitoring and reporting guidance for public water systems. Office of Water, U.S. Environmental Protection AgeP.PDF.

EPA. 2018. 2018 edition of the drinking water standards and health advisories tables. Washington, DC: Office of Water, U.S. Environmental Protection Age8.pdf.

EPA. 2019. Determination of reportable quantities for hazardous substances. U.S. Environmental Protection Agency. Code of Federal Regulati7.pdf.

EPA. 2020a. Air quality system (AQS): Copper. U.S. Environmental Protection Agency. https://www.epa.gov/aqs. August 18, 2020.

EPA. 2020b. Sampling methods for all parameters. AQS reference table. U.S. Environmental Protection Agency. https://aqs.epa.gov/aqsweb/documents/codetables/methods_all.html. October 21, 2020.

EPA. 2021. 40 CFR Part 141. National Primary Drinking Water Regulations: Lead and Copper Rule Revisions (LCRR). U.S. Environmental Protection Agency. https://www.regulations.gov/document/EPA-HQ-OW-2017-0300-1836. March 21, 2021.

Epstein O, Spisni R, Parbhoo S, et al. 1982. The effect of oral copper loading and portasystemic shunting on the distribution of copper in the liver, brain, kidney, and cornea of the rat. Am J Clin Nutr 35(3):551-555. 10.1093/ajcn/35.3.551.

Evans G, Majors P, Cornatzer W. 1970. Mechanism for cadmium and zinc antagonism of copper metabolism. Biochem. Biophys. Res. Commun. 40(5):1142-11913-7.

Fahmy MA. 2000. Potential genotoxicity in copper sulphate treated mice. Cytologia (Tokyo) 65:235-242.

Fahmy HM, O AA, A AH, et al. 2020. Biodistribution and toxicity assessment of copper nanoparticles in the rat brain. J Trace Elem Med Biol 61:126505. 10.1016/j.jtemb.2020.126505.

Farrer P, Mistilis S. 1967. Absorption of exogenous and endogenousbiliary copper in the rat. Nature 213(5073):291-2291b0.

FDA. 2017. Analytical results of the Total Diet Study. Individual year analytical results: Copper. https://www.fda.gov/food/total-diet-study/analytical-results-total-diet-study. August 18, 2020.

FDA. 2019a. Direct food substances affirmed as generally recognized as safe. Copper sulfate. U.S. Food and Drug Administration. Code of Federal Regulations.

FDA. 2019b. Listing of color additives exempt from certification. Copper powder. U.S. Food and Drug Administration. Code of Federal Regulations.

FDA. 2019c. Beverages. Subpart B-Requirements for specific standardized beverages. U.S. Food and Drug Administration. Code of Federal Regulations.

FDA. 2020. Elemental analysis manual for food and related products. 4.7 Inductively coupled plasma-mass spectrometric determination of arsenic, cadmium, chromium, lead, mercury, and other elements in food using microwave assisted digestion. Version 1.2. U.S. Food and Drug Administration. https://www.fda.gov/media/87509/download. October 21, 2020.

Feiler H, Storch T, Southworth R. 1980. Organics in municipal sludges: survey of forty cities. Proceedings of The National Conference on Municipal Industrial Sludge Utility Disposal:53-57.

Feng X, Melander AP, Klaue B. 2000. Contribution of municipal waste incineration to trace metal deposition on the vicinity. Water Air Soil Pollut 119(1-4):295-320637.

Fergusson J, Stewart C. 1992. The transport of airborne trace elements copper, lead, cadmium, zinc and manganese from a city into rural areas. Science of the Total Environment 121:247-2319-n.

Finelli V, Boscolo P, Salimei E, et al. 1981. Anemia in men occupationally exposed to low levels of copper. 4th Heavy Metals in the Environment, International Conference, Amsterdam:475-478.

Finley EB, Cerklewski FL. 1983. Influence of ascorbic acid supplementation on copper status in young adult men. Am J Clin Nutr 37(4):553-556. 10.1093/ajcn/37.4.553.

Fleckman P. 1985. Anatomy and physiology of the nail. Dermatol Clin 3(3):373-3874-X.

Ford ES. 2000. Serum copper concentration and coronary heart disease among US adults. Am J Epidemiol 151(12):1182-1110168.

Franchitto N, Gandia-Mailly P, Georges B, et al. 2008. Acute copper sulphate poisoning: A case report and literature review. Resuscitation 78(1):92-96. 10.1016/j.resuscitation.2008.02.017.

Franke SI, Pra D, Giulian R, et al. 2006. Influence of orange juice in the levels and in the genotoxicity of iron and copper. Food Chem Toxicol 44(3):425-435. 10.1016/j.fct.2005.08.016.

Frazier L, Hage M. 1998. Appendix 1 Occupational exposure limits for chemicals. In:ed. Reproductive hazards of the workplace. New York: Van Nostrand Reinhold, 537-543.

Fuentealba I, Mullins J, Aburto E. 2000. Effect of age and sex on liver damage due to excess dietary copper in Fischer 344 rats. Clin Toxicol 38(7):709-702384.

Fuhrer G. 1986. Extractable cadmium, mercury, copper, lead, and zinc in the lower Columbia River Estuary, Oregon and Washington. U.S. Geological Survey Water Resources Investigations Report 86(4088). . Portland, OR: U.S. Department of the Interior.

Fukui H, Yamamoto M, Sasaki S, et al. 1994. Possible involvement of peripheral 5-HT4 receptors in copper sulfate-induced vomiting in dogs. Eur. J. Pharmacol. 257(1-2):47-52. 10.1016/0014-2999(94)90692-0.

Furst A. 1971. Trace elements related to specific chronic diseases: Cancer. Environmental geochemistry in health and disease. The Geological Society of America, Inc. 123:109-130.

Galloway J, Thornton J, Norton S, et al. 1982. Trace metals in atmospheric deposition: A review and assessment. Atmospheric Environment 16(7):1677-17262-1.

Gamakaranage CS, Rodrigo C, Weerasinghe S, et al. 2011. Complications and management of acute copper sulphate poisoning; a case discussion. J Occup Med Toxicol 6(1):34. 10.1186/1745-6673-6-34.

Gao S, Walker W, Dahlgren R, et al. 1997. Simultaneous sorption of Cd, Cu, Ni, Zn, Pb, and Cr on soils treated with sewage sludge supernatant. Water Air Soil Pollut 93:331-304765.

Gao P, Lei T, Jia L, et al. 2018. Bioaccesible trace metals in lip cosmetics and their health risks to female consumers. Environ Pollut 238:554-53.072.

Garrett N, Lewtas J. 1983. Cellular toxicity in Chinese hamster ovary cells culture. I. Analysis of cytotoxicity endpoints for twenty-nine priority pollutants. Environ. Res. 32(2):455-465.

Gehring U, Beelen R, Eeftens M, et al. 2015. Particulate matter composition and respiratory health: the PIAMA Birth Cohort study. Epidemiology 26(3):300-309. 10.1097/EDE.0000000000000264.

Georgieva S, Popov B, Petrov V. 2013. Genotoxic effects of copper sulfate in rabbits. Archive Biology Science 65(3):963-93963G.

Georgopoulos PG, Roy A, Yonone-Lioy MJ, et al. 2001. Environmental copper: Its dynamics and human exposure issues. J Toxicol Environ Health B Crit Rev 4(4):341-394. 10.1080/109374001753146207.

Gerritse RG, Van Driel W. 1984. The relationship between adsorption of trace metals, organic matter, and pH in temperate soils. J Environ Qual 13(2):197-20005x.

Gibson A, Faucher L, Schurr M. 2011. Molten copper inhalation. Burns 37(6):e50-e53. 10.1016/j.burns.2011.05.009.

Giesy JP, Briese LA, Leversee GJ. 1978. Metal binding capacity of selected Maine surface waters. Environ Geol 2(5):257-230672.

Giesy JP, Newell A, Leversee GJ. 1983. Copper speciation in soft, acid, humic waters: Effects on copper bioaccumulation by and toxicity to Simocephalus serrulatus (Daphnidae). Science of the Total Environment 28(1-3):23-005-9.

Gill JS, Bhagat CI. 1999. Acute copper poisoning from drinking lime cordial prepared and left overnight in an old urn. Med. J. Aust. 170(10):510. 10.5694/j.1326-5377.1999.tb127863.x.

Gilman J. 1962. Metal carcinogenesis: II. A study on the carcinogenic activity of cobalt, copper, iron, and nickel compounds. Cancer Res 22:158-162.

Giusquiani PL, Gigliotti G, Businelli D. 1992. Mobility of heavy metals in urban waste-amended soils. J Environ Qual 21(3):330-30004x.

Giusti L, Yang YL, Hewitt CN, et al. 1993. The solubility and partitioning of atmospherically derived trace metals in artificial and natural waters: A review. Atmospheric Environment. Part A. General Topics 27(10):1567-1578. 10.1016/0960-1686(93)90156-s.

Gleason RP. 1968. Exposure to copper dust. Am Ind Hyg Assoc J 29(5):461-462. 10.1080/00028896809343035.

Goldberg ED. 1986. The Mussel Watch concept. Environ. Monit. Assess 7(1):91-103. 10.1007/BF00398031.

Goldin A, Bigelow C, Veneman PLJC. 1992. Concentrations of metals in ash from municipal solid waste combusters. Chemosphere 24(3):271-2296-4.

Gollan JL, Deller DJ. 1973. Studies on the nature and excretion of biliary copper in man. Clin Sci 44(1):9-15. 10.1042/cs0440009.

Golomb D, Ryan D, Eby N, et al. 1997. Atmospheric deposition of toxics onto Massachusetts Bay—I. Metals. Atmospheric Environment 31(9):1349-13276-2.

Gorter RW, Butorac MC, Eloy Pulido. 2004. Examination of the cutaneous absorption of copper after the use of copper-containing ointments. Am J Ther 11(6):453-465.e5.

Gotteland MA, M., Pizarro F, Olivares M. 2001. Effect of acute copper exposure on gastrointestinal permeability in healthy volunteers. Dig Dis Sci 46(9):1909-1914390.

Gralak MA, Leontowicz M, Morawiec M, et al. 1996. Comparison of the influence of dietary fibre sources with different proportions of soluble and insoluble fibre on Ca, Mg, Fe, Zn, Mn and Cu apparent absorption in rats. Arch Tierernahr 49(4):293-299. 10.1080/17450399609381892.

Greene F, Lamb L, Barwick M, et al. 1987. Effect of dietary copper on colonic tumor production and aortic integrity in the rat. J Surg Res 42(5):503-5025-4.

Grillo C, Reigosa M, Fernandez Lorenzo de Mele M. 2010. Does over-exposure to copper ions released from metallic copper induce cytotoxic effects on mammalian cells? Contraception 81(4):343-32.003.

Griswold MK, Nordberg A, Babu KM, et al. 2017. Accidental copper sulfate toxicity after flame colorant ingestion. Clin Toxicol (Phila) 55(8):943-945. 10.1080/15563650.2017.1330958.

Gu XY, Wang X, Guo LN, et al. 2012. [The related study of first pregnancy women with Cu-IUD on copper content of blood serum and decidua, chorion tissues]. Zhonghua Yi Xue Za Zhi 92(5):324-326 (Chinese).

Gunay N, Yildirim C, Karcioglu O, et al. 2006. A series of patient in the emergency department diagnosed with copper poisoning: Recognition equals treatment. Tohoku. J. Exp. Med 209(3):243-29.243.

Gupta U. 1979. Copper in agricultural crops. In: Nriagu JO, ed. Copper in the environment. Part I: Ecological Cycling
New York: John Wiley & Sons Inc.

Gupta D, Kerai S, Budoo MS. 2018. A fatal and deceiving case of copper sulphate poisoning. Indian J Anaesth 62(10):819-820. 10.4103/ija.IJA_71_18.

Gutenmann W, Rutzke M, Kuntz T, et al. 1994. Elements and polychlorinated biphenyls in sewage sludges of large cities in the United States. Chemosphere 28(4):725-7225-9.

Guttmann S, Bernick F, Naorniakowska M, et al. 2018. Functional characterization of Novel ATP7B variants for diagnosis of Wilson Disease. Front Pediatr 6:106. 10.3389/fped.2018.00106.

Ha JH, Doguer C, Wang X, et al. 2016. High-iron consumption impairs growth and causes copper-deficiency anemia in weanling Sprague-Dawley rats. PLoS One 11(8):e0161033. 10.1371/journal.pone.0161033.

Haddad DS, Al-Alousi LA, Kantarjian AH. 1991. The effect of copper loading on pregnant rats and their offspring. Funct Dev Morphol 1(3):17-22.

Hagemann C. 1992. 28 day repeated dose dermal toxicity study in the rat. Basel, Switzerland: CIBA-GEIGY Limited, Plant Protection.

Haines R. 1984. Environmental contamination: Surveys of heavy metals in urban soils and hazard assessment. Trace Subst Environ Health 18:450-460.

Hall A, Young B, Bremner I. 1979. Intestinal metallothionein and the mutual antagonism between copper and zinc in the rat. J. Inorg. Biochem. 11(1):57-054-9.

Harada M, Honma Y, Yoshizumi T, et al. 2020. Idiopathic copper toxicosis: Is abnormal copper metabolism a primary cause of this disease? Med Mol Morphol 53:50-55. 10.1007/s00795-019-00227-4.

Hardman B, Michalczyk A, Greenough M, et al. 2007. Distinct functional roles for the Menkes and Wilson copper translocating P-type ATPases in human placental cells. Cell. Physiol. Biochem 20(6):1073-1010718.

Harris ED. 1993. The transport of copper. Essential and Toxic Trace Elements in Human Health and Disease: An Update 380:163-179.

Harrison F, Bishop D. 1984. Review of the impact of copper released into freshwater environments. U.S. Nuclear Regulatory Commission. Livermore, CA: Lawrence Livermore National Lab.

Harrison F, Bishop D, Emerson R, et al. 1980. Concentration and speciation of copper in waters collected near the San Onofre and Diablo Canyon nuclear power stations. Livermore, CA: Lawrence Livermore National Laboratory.

Harvey LJ, Majsak-Newman G, Dainty JR, et al. 2003. Adaptive responses in men fed low- and high-copper diets. Br. J. Nutr. 90(1):161-168. 10.1079/bjn2003887.

Harvey LJ, Dainty JR, Hollands WJ, et al. 2005. Use of mathematical modeling to study copper metabolism in humans. Am J Clin Nutr 81(4):807-813. 10.1093/ajcn/81.4.807.

Haschke F, Ziegler EE, Edwards BB, et al. 1986. Effect of iron fortification of infant formula on trace mineral absorption. J Pediatr Gastroenterol Nutr 5(5):768-773. 10.1097/00005176-198609000-00018.

Hashimyousif E, Obaid HM, Karim AJ, et al. 2019. Toxico pathological study of copper sulfate modulate by zinc oxide and coriandrum sativum plant treatment in mice. Plant Archives 19(1):299-308.

Hashish EA, Elgaml SA. 2016. Hepatoprotective and nephroprotective effect of curcumin against copper toxicity in rats. Indian J. Clin. Biochem 31(3):270-277. 10.1007/s12291-015-0527-8.

Hassan S, Shaikh MU, Ali N, et al. 2010. Copper sulphate toxicity in a young male complicated by methemoglobinemia, rhabdomyolysis and renal failure. J Coll Physicians Surg Pak 20(7):490-491. 07.2010/JCPSP.490491.

Haynes W. 2015. CRC handbook of chemistry and physics: A ready-reference book of chemical and physical data. 95th ed. Boca Raton, FL: CRC Press

Haywood S. 2019. Brain–Barrier Regulation, Metal (Cu, Fe) Dyshomeostasis, and Neurodegenerative Disorders in Man and Animals. Inorganics. 7:108. 10.3390/inorganics7090108.

He XT, Logan TJ, Traina SJ. 1995. Physical and chemical characteristics of selected US municipal solid waste composts. J Environ Qual 24(3):543-552. https://doi.org/10.2134/jeq1995.00472425002400030022x.

Heit M, Klusek CS. 1985. Trace element concentrations in the dorsal muscle of white suckers and brown bullheads from two acidic Adirondack lakes. Water Air Soil Pollut 25:87-96. https://doi.org/10.1007/BF00159627.

Hellman NE, Gitlin JD. 2002. Ceruloplasmin metabolism and function. Annu. Rev. Nutr. 22:439-458. 10.1146/annurev.nutr.22.012502.114457.

Helz GR, Huggett RJ, Hill JM. 1975. Behavior of Mn, Fe, Cu, Zn, Cd and Pb discharged from a wastewater treatment plant into an estuarine environment. Water Res 9(7):631-636. https://doi.org/10.1016/0043-1354(75)90168-2.

Henckens MLCM, Worrell E. 2020. Reviewing the availability of copper and nickel for future generations. The balance between production growth, sustainability and recycling rates. Journal of Cleaner Production 264:121460. https://doi.org/10.1016/j.jclepro.2020.121460.

Herawati N, Suzuki S, Hayashi K, et al. 2000. Cadmium, copper, and zinc levels in rice and soil of Japan, Indonesia, and China by soil type. Bull. Environ. Contam. Toxicol. 64:33-39. https://doi.org/10.1007/s001289910006.

Hermann R, Neumann-Mahlkau P. 1985. The mobility of zinc, cadmium, copper, lead, iron and arsenic in ground water as a function of redox potential and pH. Science of the Total Environment 43(1-2):1-12. https://doi.org/10.1016/0048-9697(85)90027-0.

Herrero M, Rovira J, Esplugas R, et al. 2020. Human exposure to trace elements, aromatic amines and formaldehyde in swimsuits: Assessment of the health risks. Environ. Res. 181:108951. 10.1016/j.envres.2019.108951.

Hickey C, Gordon C, Galdanes K, et al. 2020. Toxicity of particles emitted by fireworks. Part Fibre Toxicol 17:28. 10.1186/s12989-020-00360-4.

Higny J, Vanpee D, Boulouffe C. 2014. Bluish vomiting: A rare clinical presentation of poisoning. Acta Clin Belg 69(4):299-301. 10.1179/2295333714Y.0000000033.

Hirano S, Sakai S, Ebihara H. 1990. Metabolism and pulmonary toxicity of intratracheally instilled cupric sulfate in rats. Toxicology 64(3):223-233. https://doi.org/10.1016/0300-483x(90)90115-w.

Holbert M. 1990. Acute inhalation toxicity study in rats. Houston, Texas: Stillmeadow, Inc.

Holtzman NA, Elliott DA, Heller RH. 1966. Copper intoxication. Report of a case with observations on ceruloplasmin. N Engl J Med 275(7):347-352. 10.1056/NEJM196608182750702.

Hoogenraad T, Koevoet R, de Ruyter Kover E. 1979. Oral zinc sulphate as long-term treatment in Wilson's disease (hepatolenticular degeneration) Eur Neurol 18(3):205-211. https://doi.org/10.1159/000115077.

Hopps H. 1977. The biologic bases for using hair and nail for analyses of trace elements. Sci. Total. Environ 7(1):71-89. https://doi.org/10.1016/0048-9697(77)90018-3.

Horn CC, Meyers K, Lim A, et al. 2014. Delineation of vagal emetic pathways: intragastric copper sulfate-induced emesis and viral tract tracing in musk shrews. Am. J. Physiol. Regul. Integr. Comp. Physiol. 306(5):R341-351. 10.1152/ajpregu.00413.2013.

Hostynek JJ, Dreher F, Maibach HI. 2010. Human skin retention and penetration of a copper tripeptide in vitro as function of skin layer towards anti-inflammatory therapy. Inflamm. Res 59(11):983-988. 10.1007/s00011-010-0214-4.

Huang H-I, Shih H-Y, Lee C-M, et al. 2008. In vitro efficacy of copper and silver ions in eradicating Pseudomonas aeruginosa, Stenotrophomonas maltophilia and Acinetobacter baumannii: Implications for on-site disinfection for hospital infection control. Water Res 42(1):73-80. https://doi.org/10.1016/j.watres.2007.07.003.

Huh C-A. 1996. Fluxes and budgets of anthropogenic metals in the Santa Monica and San Pedro Basins off Los Angeles: Review and reassessment. Science of The Total Environment 179:47-60. 10.1016/s0048-9697(96)90048-0.

Hunt JR, Vanderpool RA. 2001. Apparent copper absorption from a vegetarian diet. Am J Clin Nutr 74(6):803-807. 10.1093/ajcn/74.6.803.

Husain N, Mahmood R. 2019. Copper (II) generates ROS and RNA, impairs antioxidant system and damages membrane and DNA in human blood cells. Environmental Science and Pollution Research 26(20):20654-20668. https://doi.org/10.1007/s11356-019-05345-1.

Hutchinson TC. 1979. Copper contamination of ecosystems caused by smelter activities. In: Nriagu JO, ed. Copper in the environment. Part I: Ecological cycling. New York: John Wiley and Sons Inc.

Iannuzzi TJ, Huntley SL, Schmidt CW, et al. 1997. Combined sewer overflows (CSOs) as sources of sediment contamination in the lower Passaic River, New Jersey. I. Priority pollutants and inorganic chemicals. Chemosphere 34(2):213-231. 10.1016/s0045-6535(96)00373-6.

IARC. 2020. Agents classified by the IARC monographs, volumes 1–127. International Agency for Research on Cancer, World Health Organization. https://monographs.iarc.who.int/list-of-classifications. December 3, 2020.

Igic PG, Lee E, Harper W, et al. 2002. Toxic effects associated with consumption of zinc. Mayo Clin Proc 77(7):713-716. 10.4065/77.7.713.

Institute of Medicine. 2006. Dietary reference intakes: The essential guide to nutrient requirements. Washington, DC: The National Academies Press. https://www.nal.usda.gov/sites/default/files/fnic_uploads/DRIEssentialGuideNutReq.pdf.

IRIS. 1988. Integrated risk information system (IRIS). Chemical assessment summary. Copper; CASRN 7440-50-8. U.S. Environmental Protection Agency, National Center for Environmental Assessment.

Isaac RA, Gil L, Cooperman AN. 1997. Corrosion in drinking water distribution systems: A major contributor of copper and lead to wastewaters and effluents. Environ. Sci. Technol. 31(11):3198-3203. https://doi.org/10.1021/es970185i.

Iyanda AA, Anetor J, Adeniyi FAA. 2011. Altered copper level and renal dysfunction in Nigerian women using skin-whitening agents. Biol Trace Elem Res 143(3):1264-1270. https://doi.org/10.1007/s12011-011-8962-8.

Jaccob A. 2020. Evaluation of lead and copper content in hair of workers from oil product distribution companies in Iraq. Braz J Pharm S ci 56:e18061. https://doi.org/10.1590/s2175-97902019000318061.

Jacob RA, Skala JH, Omaye ST, et al. 1987. Effect of varying ascorbic acid intakes on copper absorption and ceruloplasmin levels of young men. J. Nutr. 117(12):2109-2115. 10.1093/jn/117.12.2109.

Janssen RPT, Posthuma L, Baerselman R, et al. 1997. Equilibrium partitioning of heavy metals in Dutch field soils. II. Prediction of metal accumulation in earthworms. Environ Toxicol Chem 16(12):2479-2488. 10.1002/etc.5620161207.

Jenkins D, Russell LL. 1994. Heavy metals contribution of household washing products to municipal wastewater. Water Environ Res 66(6):805-813.

Jing M, Liu Y, Song W, et al. 2016. Oxidative damage induced by copper in mouse primary hepatocytes by single-cell analysis. Environ Sci Pollut Res Int 23(2):1335-1343. 10.1007/s11356-015-5360-3.

Johansson A, Camner P, Jarstrand C, et al. 1983. Rabbit alveolar macrophages after inhalation of soluble cadmium, cobalt, and copper: A comparison with the effects of soluble nickel. Environ. Res. 31(2):340-354. 10.1016/0013-9351(83)90012-9.

Johansson A, Curstedt T, Robertson B, et al. 1984. Lung morphology and phospholipids after experimental inhalation of soluble cadmium, copper, and cobalt. Environ. Res. 34(2):295-309. 10.1016/0013-9351(84)90098-7.

Johnson CA, Sigg L, Zobrist J. 1987. Case studies on the chemical composition of fogwater: The influence of local gaseous emissions. Atmospheric Environment 21(11):2365-2374. 10.1016/0004-6981(87)90371-4.

Johnson PE, Milne DB, Lykken GI. 1992. Effects of age and sex on copper absorption, biological half-life, and status in humans. Am J Clin Nutr 56(5):917-925. 10.1093/ajcn/56.5.917.

Johnson DB, Hedrich S, Pakostova E. 2017. Indirect redox transformations of iron, copper, and chromium catalyzed by extremely acidophilic bacteria. Front Microbiol 8:211. https://doi.org/10.3389/fmicb.2017.00211.

Kadammattil AV, Sajankila SP, Prabhu S, et al. 2018. Systemic toxicity and teratogenicity of copper oxide nanoparticles and copper sulfate. J Nanosci Nanotechnol 18(4):2394-2404. 10.1166/jnn.2018.14542.

Kalita J, Kumar V, Misra UK, et al. 2020. Movement disorder in copper toxicity rat model: Role of inflammation and apoptosis in the corpus striatum. Neurotox Res 37(4):904-912. 10.1007/s12640-019-00140-9.

Kamamoto Y, Makiura S, Sugihara S, et al. 1973. The inhibitory effects of copper on DL-ethionine carcinogenesis in rats. Cancer Res 33(5):1129-1135.

Kang J, Lin C, Chen J, Liu Q. 2004. Copper induces histone hypoacetylation through directly inhibiting histone acetyltransferase activity. Chemico-Biological Interactions. 148:115-123. 10.1016/j.cbi.2004.05.003

Karlsson B, Noren L. 1965. Ipecacuanha and copper sulphate as emetics in intoxications in children. Acta Paediatr Scand 54:331-335. https://doi.org/10.1111/j.1651-2227.1965.tb06380.x.

Karlsson HL, Cronholm P, Gustafsson J, et al. 2008. Copper oxide nanoparticles are highly toxic: A comparison between metal oxide nanoparticles and carbon nanotubes. Chem Res Toxicol 21(9):1726-1732. 10.1021/tx800064j.

Kasperczyk A, Dobrakowski M, Czuba ZP, et al. 2016. Environmental exposure to zinc and copper influences sperm quality in fertile males. Ann. Agric. Environ. Med. 23(1):138-143. 10.5604/12321966.1196869.

Keeler GJ, Pirrone N. 1993. Deposition of trace metals in urban and rural areas in the lake Michigan basin. Water Sci Technol 28(3-5):261-270. 10.2166/wst.1993.0427.

Keller JC, Kaminski EJ. 1984. Toxic effects of Cu implants on liver. Fundam Appl Toxicol 4(5):778-783. 10.1016/0272-0590(84)90099-x.

Khaghani S, Ezzatpanah H, Mazhari N, et al. 2010. Zinc and copper concentrations in human milk and infant formulas. Iran J Pediatr 20(1):53-57.

Kheirandish R, Askari N, Babaei H. 2014. Zinc therapy improves deleterious effects of chronic copper administration on mice testes: histopathological evaluation. Andrologia 46(2):80-85. 10.1111/and.12047.

Khushboo M, Murthy MK, Devi MS, et al. 2018. Testicular toxicity and sperm quality following copper exposure in Wistar albino rats: ameliorative potentials of L-carnitine. Environ Sci Pollut Res Int 25(2):1837-1862. 10.1007/s11356-017-0624-8.

Kilbride KM, Paveglio FL, Altstatt AL, et al. 1998. Contaminant loading in drainage and fresh water used for wetland management at Stillwater National Wildlife Refuge. Arch. Environ. Contam. Toxicol. 35(2):236-248. 10.1007/s002449900372.

Kim N, Fergusson J. 1993. Concentrations and sources of cadmium, copper, lead and zinc in house dust in Christchurch, New Zealand. Sci. Total. Environ 138(1-3):1-21. 10.1016/0048-9697(93)90400-z.

Kim SY, Park JH, Kim EA, et al. 2012. Longitudinal study on trace mineral compositions (selenium, zinc, copper, manganese) in Korean human preterm milk. J Korean Med Sci 27(5):532-536. 10.3346/jkms.2012.27.5.532.

Kim JS, Adamcakova-Dodd A, O'Shaughnessy PT, et al. 2011. Effects of copper nanoparticle exposure on host defense in a murine pulmonary infection model. Part Fibre Toxicol 8(1):29. 10.1186/1743-8977-8-29.

Kimball KD. 1973. Seasonal fluctuations of ionic copper in Knights Pond, Massachusetts. Limnology and Oceanography 18(1): 169-172. https://doi.org/10.4319/lo.1973.18.1.0169.

Kjaergaard K, Sandah T, Frisch T, et al. 2020. Intravenous and oral copper kinetics, biodistribution and dosimetry in healthy humans studied by [64 Cu] Copper PET/CT. EJNMMI Radiopharmacy and Chemistry 5(1):15. https://dx.doi.org/10.1186%2Fs41181-020-00100-1.

Kline RD, Hays VW, Cromwell GL. 1971. Effects of copper, molybdenum and sulfate on performance, hematology and copper stores of pigs and lambs. J. Anim. Sci. 33(4):771-779. 10.2527/jas1971.334771x.

Knobeloch L, Schubert C, Hayes J, et al. 1998. Gastrointestinal upsets and new copper plumbing - is there a connection? Wis Med J 97(1):49-53.

Knobeloch L, Ziarnik M, Howard J, et al. 1994. Gastrointestinal upsets associated with ingestion of copper-contaminated water. Environ. Health Perspect. 102(11):958-961. 10.1289/ehp.94102958.

Kodama H, Fujisawa C, Bhadhprasit W. 2012. Inherited copper transport disorders: Biochemical mechanisms, diagnosis, and treatment. Curr Drug Metab 13(3):237-250. 10.2174/138920012799320455.

Koo YJ, Pack EC, Lee YJ, et al. 2020. Determination of toxic metal release from metallic kitchen utensils and their health risks. Food Chem Toxicol 145:111651. https://doi.org/10.1016/j.fct.2020.111651.

Krishnan K, Andersen ME, Clewell HJI, et al. 1994. Physiologically based pharmacokinetic modeling of chemical mixtures. In: Yang RSH, ed. Toxicology of Chemical Mixtures.

Kuhn JO. 1989a. Acute oral toxicity in rats. Houston, Texas: Stillmeadow, Inc.

Kuhn JO. 1989b. Acute dermal toxicity study in rabbits. Houston, Texas: Stillmeadow, Inc.

Kumar A, Sharma CB. 1987. Hematological indices in copper-poisoned rats. Toxicol Lett 38(3):275-278. 10.1016/0378-4274(87)90009-9.

Kumar J, Sathua KB, Flora SJS. 2019. Chronic copper exposure elicit neurotoxic responses in rat brain: Assessment of 8-hydroxy-2-deoxyguanosine activity, oxidative stress and neurobehavioral parameters. Cell Mol Biol 65(1):27-35. 10.14715/cmb/2019.65.1.5.

Kumar V, Kalita J, Misra UK, et al. 2015. A study of dose response and organ susceptibility of copper toxicity in a rat model. J Trace Elem Med Biol 29:269-274. 10.1016/j.jtemb.2014.06.004.

Kumar V, Kalita J, Bora HK, et al. 2016a. Temporal kinetics of organ damage in copper toxicity: A histopathological correlation in rat model. Regul Toxicol Pharmacol 81:372-380. 10.1016/j.yrtph.2016.09.025.

Kumar V, Kalita J, Bora HK, et al. 2016b. Relationship of antioxidant and oxidative stress markers in different organs following copper toxicity in a rat model. Toxicol Appl Pharmacol 293:37-43. https://doi.org/10.1016/j.taap.2016.01.007.

Kvietkauskaite R, Dringeliene A, Markevicius A, et al. 2004. Effect of low copper exposure on the antioxidant system and some immune parameters. Vet. Hum. Toxicol 46(4):169-172.

Lacerda LD, Santos JA, Lopes DV. 2009. Fate of copper in intensive shrimp farms: bioaccumulation and deposition in pond sediments. Braz. J. Biol. 69(3):851-858. 10.1590/s1519-69842009000400012.

Lagos GE, Cuadrado CA, Letelier MV. 2001. Aging of copper pipes by drinking water. Journal AWWA 93(11):94-103. https://doi.org/10.1002/j.1551-8833.2001.tb09338.x.

LaGow B, eds ea. 2007. PDR lab advisor: A comprehensive point-of-care guide for over 600 lab tests (Vol. First Edition). Montvale, NJ: Thomson PDR

Lamont DL, Duflou JALC. 1988. Copper sulfate. Not a harmless chemical. Forensic Toxicology 9(3):226-227. https://doi.org/10.1097/00000433-198809000-00010.

Landing WM, Perry JJ, Guentzel JL, et al. 1995. Relationships between the atmospheric deposition of trace elements, major ions, and mercury in Florida: The FAMS project (1992–1993). Water Air Soil Pollut 80(1-4):343-352. 10.1007/bf01189684.

Lapid O. 2008. Copper sulphate burns to the hands, a complication of traditional medicine. J Burn Care Res 29(3):544-547. 10.1097/BCR.0b013e3181711183.

Lavigne A, Freni Sterrantino A, Liverani S, et al. 2019. Associations between metal constituents of ambient particulate matter and mortality in England: An ecological study. BMJ Open 9(12):e030140. 10.1136/bmjopen-2019-030140.

Law LW. 1938. The effects of chemicals on the lethal mutation rate in drosophila melanogaster. Proc. Natl. Acad. Sci. U. S. A 24(12):546-550. https://dx.doi.org/10.1073%2Fpnas.24.12.546.

Lecyk M. 1980. Toxicity of CuSO4 in mice embryonic development. Zool. Pol 28:101-105.

Levenson CW, Janghorbani M. 1994. Long-term measurement of organ copper turnover in rats by continuous feeding of a stable isotope. Anal. Biochem. 221(2):243-249. 10.1006/abio.1994.1408.

Levy DB, Barbarick KA, Siemer EG, et al. 1992. Distribution and partitioning of trace metals in contaminated soils near Leadville, Colorado. J Environ Qual 21(2):185-195. 10.2134/jeq1992.00472425002100020006x.

Lifset RJ, Gordon RB, Graedel TE, et al. 2002. Where has all the copper gone: The stocks and flows project, part 1. Jom 54(10):21-26. 10.1007/bf02709216.

Lifset RJ, Eckelman MJ, Harper EM, et al. 2012. Metal lost and found: dissipative uses and releases of copper in the United States 1975-2000. Sci. Total. Environ 417-418:138-147. https://doi.org/10.1016/j.scitotenv.2011.09.075.

Lioy P, Daisey J, Morandi M, et al. 1987. The airborne toxic element and organic substances (ATEOS) study design. In: Lioy P & Daisey J, ed. Toxic air pollution: A comprehensive study of non-criteria air pollutants. Chelsea, MI: Lewis Publishing, Inc., 3-42.

Liszewski W, Warshaw M. 2019. Pigments in American tattoo inks and their propensity to elicit allergic contact dermatitis. Journal of American Academy of Dermatology 81(2):379-385. https://doi.org/10.1016/j.jaad.2019.01.078.

Liu R, Zhao D, Barnett MO. 2006. Fate and transport of copper applied in channel catfish ponds. Water Air Soil Pollut 176(1-4):139-162. 10.1007/s11270-006-9155-5.

Liu H-L, Zhou J, Li M, et al. 2021. Chemical speciation of trace metals in atmospheric deposition and impacts on soil geochemistry and vegetable bioaccumulation near a large copper smelter in China. J Hazard Mater 413:125346. https://doi.org/10.1016/j.jhazmat.2021.125346.

Liu J, Luan J, Zhou X, et al. 2017. Epidemiology, diagnosis, and treatment of Wilson's disease. Intractable Rare Dis Res 6(4):249-255. 10.5582/irdr.2017.01057.

Liu JY, Yang X, Sun XD, et al. 2016. Suppressive effects of copper sulfate accumulation on the spermatogenesis of rats. Biol Trace Elem Res 174(2):356-361. 10.1007/s12011-016-0710-7.

Lodenius M, Braunschweiler H. 1986. Volatilisation of heavy metals from a refuse dump. Science of The Total Environment 57:253-255. 10.1016/0048-9697(86)90027-6.

Longerich HP, Friel JK, Fraser C, et al. 1991. Analysis of the drinking water of mothers of neural tube defect infants and of normal infants for 14 selected trace elements by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). Canadian Journal of Applied Spectroscopy 36(1):15-21.

Lu J, Wu D-M, Zheng Y-L, et al. 2009. Trace amounts of copper exacerbate beta amyloid-induced neurotoxicity in the cholesterol-fed mice through TNF-mediated inflammatory pathway. Brain Behavior and Immunity 23(2):193-203. https://doi.org/10.1016/j.bbi.2008.09.003.

Lubica C, Rudolf M, Jiri L. 2017. Acute copper sulphate poisoning. J Coll Physicians Surg Pak 27(8):527-528. 2690.

Luncan-Bouche ML, Couderchet M, Vernet G, et al. 1997. The simultaneous influence of pH and temperature on binding and mobilization of metals in sand: 1-Copper. Fresenius. Environ. Bull 6:711-718.

Luo J, Hao S, Zhao L, et al. 2020. Oral exposure of pregnant rats to copper nanoparticles caused nutritional imbalance and liver dysfunction in fetus. Ecotoxicol. Environ. Saf. 206:111206. 10.1016/j.ecoenv.2020.111206.

Ma LQ, Rao GN. 1997. Chemical fractionation of cadmium, copper, nickel, and zinc in contaminated soils. J. Environ. Qual. 26(1):259-264. https://doi.org/10.2134/jeq1997.00472425002600010036x.

Maessen O, Freedman B, McCurdy R. Metal mobilization in home well water systems in Nova Scotia. Journal AWWA 77(6): 73-80. https://doi.org/10.1002/j.1551-8833.1985.tb05557.x.

Mahdy A, Elkhatib E, Fathi N. 2007. Cadmium, Copper, Nickel, and Lead Availability in Biosolids-amended Alkaline Soils. Australian Journal of Basic and Applied Sciences 1(4): 354-363.

Makale MT, King GL. 1992. Surgical and pharmacological dissociation of cardiovascular and emetic responses to intragastric CuSO4. Am. J. Physiol. 263(2 Pt 2):R284-291. 10.1152/ajpregu.1992.263.2.R284.

Malik M, Mansur A. 2011. Copper sulphate poisoning and exchange transfusion. Saudi J Kidney Dis Transpl 22(6):1240-1242.

Mannsville Chemical Products. 1984. Chemical products synopsis: Copper sulfate. Cortland, NY: Mannsville Chemical Products Corp.

Marinussen MPJC, Zee SEATM, Haan FAM, et al. 1997. Heavy metal (copper, lead, and zinc) accumulation and excretion by the earthworm, Dendrobaena veneta. J Environ Qual 26(1):278-284. 10.2134/jeq1997.00472425002600010039x.

Mark AB, Kapolna E, Laursen KH, et al. 2013. Consumption of organic diets does not affect intake and absorption of zinc and copper in men - evidence from two cross-over trials. Food Funct 4(3):409-419. 10.1039/c2fo30247k.

Markert A, Baumann R, Gerhards B, et al. 2016. Single and combined exposure to zinc- and copper-containing welding fumes lead to asymptomatic systemic inflammation. J Occup Environ Med 58(2):127-132. 10.1097/JOM.0000000000000652.

Mart L, Nurnberg HW. 1984. Trace metal levels in the eastern Arctic Ocean. Sci Total Environ 39:1-14.

Marzin D, Phi H. 1985. Study of the mutagenicity of metal derivatives with Salmonella typhimurium. Mutat. Res. 155:49-51. https://doi.org/10.1016/0165-1218(85)90024-2.

Massie HR, Aiello VR. 1984. Excessive intake of copper: influence on longevity and cadmium accumulation in mice. Mech. Ageing Dev. 26(2-3):195-203. 10.1016/0047-6374(84)90093-9.

Mayo Clinic. 2020. Test Definition: CUU. Copper 24Hr, U. In: Mayo Clinic Laboratories.

McArdle H. 1995. The metabolism of copper during pregnancy--a review. Food Chem 54(1):79-84. https://doi.org/10.1016/0308-8146(95)92666-8.

Melino S, Nepravishta R, Bellomaria A, Di Marco S, Paci M. 2009. Nucleic Acid Binding of the RTN1-C C-Terminal Region: Toward the Functional Role of a Reticulon Protein. Biochemistry. 48(2):242-253. 10.1021/bi801407w.

Meranger JC, Subramanian KS, Chalifoux C. 1979. A national survey for cadmium, chromium, copper, lead, zinc, calcium, and magnesium in Canadian drinking water supplies. Environ Sci Tech 13(6):707-711. https://doi.org/10.1021/es60154a009.

Mercer JF, Lazdins I, Stevenson T, et al. 1981. Copper induction of translatable metallothionein messenger RNA. Biosci. Rep. 1(10):793-800. 10.1007/BF01114802.

Mills GL, Quinn JG. 1984. Dissolved copper and copper-organic complexes in the Narragansett Bay estuary. Mar Chem 15(2):151-172. 10.1016/0304-4203(84)90013-6.

Minear RA, Ball RO, Church RL. 1981. Project Summary: Data base for influent heavy metals in publicly owned treatment works. Cincinnati, OH: Municipal Environmental Research Laboratory, U.S. Environmental Protection Agency.

Moffett J, Zika R. 1987. Photochemistry of copper complexes in sea water. Photochemistry of environmental aquatic systems. ACS Sump Ser 327: 116-130. 10.1021/bk-1987-0327.ch009.

Moriya M, Ho YH, Grana A, et al. 2008. Copper is taken up efficiently from albumin and alpha2-macroglobulin by cultured human cells by more than one mechanism. Am. J. Physiol. Cell Physiol. 295(3):C708-C721. 10.1152/ajpcell.00029.2008.

Morris AE, Wadsley M. 2001. Metal extraction: Phase stability diagrams. In: Buschow KHJ, Cahn RW, Flemings MC, et al., ed. Encyclopedia of materials: Science and technology. Oxford: Elsevier, 5362-5377.

Mortazavi F, Jafari-Javid A. 2009. Acute renal failure due to copper sulfate poisoning: A case report. Iran J Pediatr 19(1):75-78.

Motlhatlhedi K, Firth JA, Setlhare V, et al. 2014. A novel and fatal method of copper sulphate poisoning. African Journal of Emergency Medicine 4(4):e23-e25. 10.1016/j.afjem.2014.02.002.

Moussiegt A, Ferreira L, Aboab J, et al. 2020. She has the blues: An unusual case of copper sulphate intoxication. Eur J Case Rep Intern Med 7(2):001394. 10.12890/2020_001394.

Mulder EG, van Veen WL. 1968. Effect of microorganisms on the transformation of mineral fractions in soil. Trans Int Cong Soil Sci 9:651-661.

Müller T, Müller W, Feichtinger H. 1998. Idiopathic copper toxicosis. Am J Clin Nutr 67(5 Suppl):1082s-1086s. 10.1093/ajcn/67.5.1082S.

Müller T, Feichtinger H, Berger H, et al. 1996. Endemic Tyrolean infantile cirrhosis: an ecogenetic disorder. Lancet 347(9005):877-880. 10.1016/s0140-6736(96)91351-3.

Munley SM. 2003a. Copper hydroxide: Pilot developmental toxicity study in rabbits. Newark, DE: E. I. du Pont de Nemours and Company, Haskell Laboratory for Health and Environmental Sciences.

Munley SM. 2003b. Copper hydroxide: Developmental toxicity study in rabbits. Newark, DE: E. I. du Pont de Nemours and Company, Haskell Laboratory for Health and Environmental Sciences.

Murphy EA. 1993. Effectiveness of flushing on reducing lead and copper levels in school drinking water. Environ. Health Perspect. 101(3):240-241. 10.1289/ehp.93101240.

Murthy RC, Lal S, Saxena DK, et al. 1981. Effect of manganese and copper interaction on behavior and biogenic amines in rats fed a 10% casein diet. Chem Biol Interact 37(3):299-308. 10.1016/0009-2797(81)90116-2.

Musacco-Sebio R, Saporito-Magriñá C, Acosta JM, et al. 2017. Iron and copper toxicity in rat liver: A kinetic and holistic overview. Liver Research – Open Journal 2(1):9-13. 10.17140/lroj-2-110.

Musci G, Bonaccorsi di Patti MC, Calabrese L. 1993. The state of the copper sites in human ceruloplasmin. Arch. Biochem. Biophys. 306(1):111-118. 10.1006/abbi.1993.1487.

Myint ZW, Oo TH, Thein KZ, et al. 2018. Copper deficiency anemia: review article. Ann Hematol 97(9):1527-1534. 10.1007/s00277-018-3407-5.

NAS/NRC. 1989. Report of the oversight committee. In:ed. Biologic markers in reproductive toxicology. Washington, DC: National Academy of Sciences, National Research Council, National Academy Press

Naveed M, Moldrup P, Arthur E, et al. 2014. Simultaneous Loss of Soil Biodiversity and Functions along a Copper Contamination Gradient: When Soil Goes to Sleep. Soil Science Society of America Journal 78(4):1239-1250. https://doi.org/10.2136/sssaj2014.02.0052.

Nayak NC, Chitale AR. 2013. Indian childhood cirrhosis (ICC) & ICC-like diseases: the changing scenario of facts versus notions. Indian J. Med. Res 137(6):1029-1042.

Naz S, Gul A, Zia M. 2020. Toxicity of copper oxide nanoparticles: a review study. IET Nanobiotechnol 14(1):1-13. 10.1049/iet-nbt.2019.0176.

Ndilila W, Callan AC, McGregor LA, et al. 2014. Environmental and toenail metals concentrations in copper mining and non mining communities in Zambia. Int J Hyg Environ Health 217(1):62-69. 10.1016/j.ijheh.2013.03.011.

Nerín C, Domeño C, García JI, et al. 1999. Distribution of Pb, V, Cr, Ni, Cd, Cu and Fe in particles formed from the combustion of waste oils. Chemosphere 38(7):1533-1540. 10.1016/s0045-6535(98)00373-7.

Neuhauser EF, Cukic ZV, Malecki MR, et al. 1995. Bioconcentration and biokinetics of heavy metals in the earthworm. Environ. Pollut. 89(3):293-301. 10.1016/0269-7491(94)00072-l.

Ni L, Li S. 2008. Effects of organic matters coming from Chinese tea on soluble copper release from copper teapot. Sci. Total. Environ 389(1):202-207. 10.1016/j.scitotenv.2007.08.039.

NIOSH. 1989. National Occupational Exposure Survey (potential exposure agents list). Cincinnati, OH: U.S. Department of Health, Education and Welfare, Public Health Service, Centers for Disease Control, National Institute for Occupational Safety and Health.

NIOSH. 2014a. Copper (dusts and mists, as Cu). Immediately Dangerous to Life or Health Concentrations (IDLH). Atlanta, GA: The National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention. https://www.cdc.gov/niosh/idlh/7440508.html. October 21, 2020.

NIOSH. 2014b. Copper fume (as Cu). Immediately Dangerous to Life or Health Concentrations (IDLH). Atlanta, GA: The National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention.

NIOSH. 2020. NIOSH manual of analytical methods (NMAM), 5th Edition (Andrews R & O'Connor PF Eds.). National Institute for Occupational Safety and Health, Centers for Disease Control and Prevention

Nishioka H. 1975. Mutagenic activities of metal compounds in bacteria. Mutat. Res. 31(3):185-189. 10.1016/0165-1161(75)90088-6.

Nishito Y, Kambe T. 2018. Absorption Mechanisms of Iron, Copper, and Zinc: An Overview. J Nutr Sci Vitaminol (Tokyo) 64(1):1-7. 10.3177/jnsv.64.1.

Nölte J. 1988. Pollution source analysis of river water and sewage sludge. Environ. Technol. Lett. 9(8):857-868. 10.1080/09593338809384642.

NRC. 2000. Copper in drinking water. Washington, DC: The National Academies Press. https://doi.org/10.17226/9782.

NRC. 1995. Nutrient requirements of laboratory animals, fourth revised edition. Washington, DC: Subcommittee on Laboratory Animal Nutrition, Committee on Animal Nutrition, Board of Agriculture, National Research Council.

Nriagu JO, Pacyna JM. 1988. Quantitative assessment of worldwide contamination of air, water and soils by trace metals. Nature 333(6169):134-139. 10.1038/333134a0.

Nriagu JO, Lawson G, Wong HKT, et al. 1996. Dissolved trace metals in Lakes Superior, Erie, and Ontario. Environ Sci Technol 30(1):178-187. 10.1021/es950221i.

NSF. 2021. NSF product and service listings. NSF/ANSI/CAN 60. Drinking water treatment chemicals - health effects. NSF International. http://info.nsf.org/Certified/PwsChemicals/Listings.asp?ChemicalName=Copper+Sulfate&PlantCountry=UNITED+STATES&. April 9, 2021.

NTP. 2016. Report on carcinogens, Fourteenth Edition. Substances listed in the fourteenth report on carcinogens. Research Triangle Park, NC: National Toxicology Program, Department of Health and Human Services. https://ntp.niehs.nih.gov/ntp/roc/content/listed_substances_508.pdf.

NTP. 1993. NTP Technical Report on toxicity studies of cupric sulfate administered in drinking water and feed to F344/N rats and B6C3F1 mice. Research Triangle Park, NC: United States Department of Health and Human Services.

Occelli F, Lanier C, Cuny D, et al. 2020. Exposure to multiple air pollutants and the incidence of coronary heart disease: A fine-scale geographic analysis. Sci. Total. Environ 714:136608. 10.1016/j.scitotenv.2020.136608.

O'Connor JMB, M.P., Turley E, McKeown A, et al. 2003. Copper supplementation has no effect on markers of DNA damage and liver function in healthy adults (FOODCUE project). Annals of Nutrition & Metabolic 47:201-206. https://doi.org/10.1159/000070486.

O'Donohue JW, Reid MA, Varghese A, et al. 1993. Micronodular cirrhosis and acute liver failure due to chronic copper self-intoxication. Eur J Gastroenterol Hepatol 5(7):561-562.

Ognik K, Cholewińska E, Juskiewicz J, Zduńczyk Z, Tutaj K, Szalak R. 2019. The effect of copper nanoparticles and copper (II) salt on redox reactions and epigenetic changes in a rat model. Journal of animal physiology and animal nutrition/Zeitschrift fuer Tierphysiologie Tierernaehrung und Futtermittelkunde. 102(2):675-686. 10.1111/jpn.13025

Ogra Y, Ohmichi M, Suzuki KT. 1996. Mechanisms of selective copper removal by tetrathiomolybdate from metallothionein in LEC rats. Toxicology 106(1-3):75-83. 10.1016/0300-483x(95)03171-b.

Ogunbileje JO, Sadagoparamanujam VM, Anetor JI, et al. 2013. Lead, mercury, cadmium, chromium, nickel, copper, zinc, calcium, iron, manganese and chromium (VI) levels in Nigeria and United States of America cement dust. Chemosphere 90(11):2743-2749. 10.1016/j.chemosphere.2012.11.058.

Ohgami RS, Campagna DR, McDonald A, et al. 2006. The Steap proteins are metalloreductases. Blood 108(4):1388-1394. 10.1182/blood-2006-02-003681.

Olivares M, Araya M, Uauy R. 2000. Copper hemeostasis in infant nutrition: Deficit and excess. J Pediatr Gastroenterol Nutr 31:102-111. https://doi.org/10.1097/00005176-200008000-00004.

Olivares M, Lonnerdal B, Abrams S. 2002. Age and copper intake do not affect copper absorption, measured with the use of 65Cu as a tracer, in young infants. Am J Clin Nutr 76(3):641-645. https://doi.org/10.1093/ajcn/76.3.641.

Olivares M, Araya M, Pizarro F, et al. 2001. Nausea threshold in apparently healthy individuals who drink fluids containing graded concentrations of copper. Regul Toxicol Pharmacol 33(3):271-275. 10.1006/rtph.2000.1440.

Olivares M, Pizarro F, Speisky H, et al. 1998. Copper in infant nutrition: safety of World Health Organization provisional guideline value for copper content of drinking water. J Pediatr Gastroenterol Nutr 26(3):251-257. 10.1097/00005176-199803000-00003.

Olmedo P, Hernández AF, Pla A, et al. 2013. Determination of essential elements (copper, manganese, selenium and zinc) in fish and shellfish samples. Risk and nutritional assessment and mercury–selenium balance. Food Chem Toxicol 62:299-307. https://doi.org/10.1016/j.fct.2013.08.076.

Oon S, Yap CH, Ihle BU. 2006. Acute copper toxicity following copper glycinate injection. Intern Med J 36(11):741-743. 10.1111/j.1445-5994.2006.01195.x.

Osán J, Meirer F, Groma V, et al. 2010. Speciation of copper and zinc in size-fractionated atmospheric particulate matter using total reflection mode X-ray absorption near-edge structure spectrometry. Spectrochimica Acta Part B: Atomic Spectroscopy 65(12):1008-1013. https://doi.org/10.1016/j.sab.2010.11.002.

OSHA. 2020a. Occupational safety and health standards. Subpart Z - Toxic and hazardous substances. Air contaminants. Table Z-1: Limits for air contaminants. Occupational Safety and Health Administration. Code of Federal Regulations: https://www.osha.gov/annotated-pels/table-z-1. October 21, 2020.

OSHA. 2020b. Occupational safety and health standards for shipyard employment. Subpart Z - Toxic and hazardous substances. Air contaminants. Occupational Safety and Health Administration. Code of Federal Regulations. https://www.osha.gov/laws-regs/regulations/standardnumber/1915/1915.1000. October 21, 2020.

OSHA. 2020c. Safety and health regulations for construction. Subpart D - Occupational health and environment controls. Gases, vapors, fumes, dusts, and mists. Occupational Safety and Health Administration. Code of Federal Regulations. https://www.osha.gov/laws-regs/regulations/standardnumber/1926/1926.55. October 21, 2020.

Ostro BD, Feng WY, Broadwin R, et al. 2008. The impact of components of fine particulate matter on cardiovascular mortality in susceptible subpopulations. Occup Environ Med 65(11):750-756. 10.1136/oem.2007.036673.

Ostro B, Hu J, Goldberg D, et al. 2015. Associations of mortality with long-term exposures to fine and ultrafine particles, species and sources: results from the California Teachers Study Cohort. Environ. Health Perspect. 123(6):549-556. 10.1289/ehp.1408565.

Page GW. 2002. Comparison of groundwater and surface water for patterns and levels of contamination by toxic substances. Environ Sci Technol 15(12):1475-1481. 10.1021/es00094a008.

Pandit A, Bhave S. 1996. Present interpretation of the role of copper in Indian childhood cirrhosis. Am J Clin Nutr 63(5):830S-835S. 10.1093/ajcn/63.5.830.

Paode RD, Sofuoglu SC, Sivadechathep J, et al. 1998. Dry deposition fluxes and mass size distributions of Pb, Cu, and Zn Measured in Southern Lake Michigan during AEOLOS. Environ Sci Technol 32(11):1629-1635. 10.1021/es970892b.

Park KS, Kwon JH, Park SH, et al. 2018. Acute copper sulfate poisoning resulting from dermal absorption. Am J Ind Med 61:783-788. 10.1002/ajim.22892.

Pedersen M, Gehring U, Beelen R, et al. 2016. Elemental Constituents of Particulate Matter and Newborn's Size in Eight European Cohorts. Environ. Health Perspect. 124(1):141-150. 10.1289/ehp.1409546.

Pekelharing H, Lemmens A, Beyen A. 1994. Iron, copper and zinc status in rats fed on diets containing various concentrations of tin. Br. J. Nutr. 71(1):103-109. https://doi.org/10.1079/bjn19940115.

Pennington JA, Young BE, Wilson DB, et al. 1986. Mineral content of foods and total diets: the Selected Minerals in Foods Survey, 1982 to 1984. J Am Diet Assoc 86(7):876-891.

Perwak J, Bysshe S, Goyer M, et al. 1980. An exposure and risk assessment for copper. Washington, DC: U.S. Environmental Protection Agency.

Petruzzelli G. 1997. Chapter 5: Soil sorption of heavy metals. In:ed. Ecological issues and environmental impact assessment. 145-175.

Pettersson R, Rasmussen F, Oskarsson A. 2003. Copper in drinking water: Not strong risk factor for diarrhoea among young children. A population-based study from Sweden. Acta Paediatr 92(4):473-480. https://doi.org/10.1111/j.1651-2227.2003.tb00581.x.

Pimentel JC, Marques F. 1969. "Vineyard sprayer's lung": A new occupational disease. Thorax 24(6):678-688. 10.1136/thx.24.6.678.

Pimentel JC, Menezes AP. 1975. Liver granulomas containing copper in vineyard sprayer's lung. A new etiology of hepatic granulomatosis. Am Rev Respir Dis 111(2):189-195. 10.1164/arrd.1975.111.2.189.

Pirot F, Millet J, Kalia Y, et al. 1996a. In vitro study of percutaneous absorption, cutaneous bioavailability and bioequivalence of zinc and copper from five topical formulations. Skin Pharmacol 9(4):259-269. https://doi.org/10.1159/000211423.

Pirot F, Panisset F, Agache P, et al. 1996b. Simultaneous absorption of copper and zinc through human skin in vitro: Influence of counter-ion and vehicle. Skin Pharmacol 9(1):43-52. https://doi.org/10.1159/000211389.

Pizarro F, Olivares M, Araya M, et al. 2001. Gastrointestinal effects associated with soluble and insoluble copper in drinking water. Environ. Health Perspect. 109(9):949-952. 10.1289/ehp.01109949.

Pizarro F, Olivares M, Uauy R, et al. 1999. Acute gastrointestinal effects of graded levels of copper in drinking water. Environ. Health Perspect. 107(2):117-121. 10.1289/ehp.99107117.

Pizarro F, Araya M, Vasquez M, et al. 2007. Case study of complaints on drinking water quality: Relationship to copper content? Biol Trace Elem Res 116(2):131-145. 10.1007/BF02685926.

Plamenac P, Santic Z, Nikulin A, et al. 1985. Cytologic changes of the respiratory tract in vineyard spraying workers. Eur J Respir Dis 67(1):50-55.

Pohanka M. 2019. Copper and copper nanoparticles toxicity and their impact on basic functions in the body. Bratisl Med J. 120(6); 397-409. 10.4149/BLL_2019_065.

Prá D, Franke SI, Giulian R, et al. 2008. Genotoxicity and mutagenicity of iron and copper in mice. BioMetals 21(3):289-297. 10.1007/s10534-007-9118-3.

Prasad R, Kumar S, Kumar S. 2006. Hydrogen peroxide commences copper induced DNA damage isolated from human blood: In vitro study. Indian J Exp Biol 44(5):377-380.

Prasad AS, Brewer GJ, Schoomaker EB, et al. 1978. Hypocupremia induced by zinc therapy in adults. JAMA 240(20):2166-2168.

Pratt WB, Omdahl JL, Sorenson JR. 1985. Lack of effects of copper gluconate supplementation. Am J Clin Nutr 42(4):681-682. 10.1093/ajcn/42.4.681.

PubChem. 2020. Compound summaries for copper, copper sulfate, and cupric chloride. National Library of Medicine, National Center for Biotechnology Information. https://pubchem.ncbi.nlm.nih.gov/. October 21, 2020.

Puchkova LV, Babich PS, Zatulovskaia YA, et al. 2018. Copper metabolism of newborns is adapted to milk ceruloplasmin as a nutritive source of copper: Overview of the current data. Nutrients 10(11):1591. 10.3390/nu10111591.

Pujol J, Fenoll R, Macia D, et al. 2016. Airborne copper exposure in school environments associated with poorer motor performance and altered basal ganglia. Brain and Behavior. 6(6). 10.1002/brb3.467

Que Hee SS, Finelli VN, Fricke FL, et al. 2006. metal content of stack emissions, coal and fly ash from some eastern and western power plants in the U.S.A. as obtained by ICP-AES. Int J Environ Anal Chem 13(1):1-18. 10.1080/03067318208071579.

Raaschou-Nielsen O, Beelen R, Wang M, et al. 2016. Particulate matter air pollution components and risk for lung cancer. Environ. Int. 87:66-73. 10.1016/j.envint.2015.11.007.

Rader KJ, Carbonaro RF, van Hullebusch ED, et al. 2019. The fate of copper added to surface water: Field, laboratory, and modeling studies. Environ. Toxicol. Chem. 38(7):1386-1399. 10.1002/etc.4440.

Rajaratnam G, Winder C, An M. 2002. Metals in Drinking Water from New Housing Estates in the Sydney Area. Environ Res 89(2):165-170. https://doi.org/10.1006/enrs.2002.4356.

Rana SV, Kumar A. 1980. Biological haematological and histological observations in copper poisoned rats. Ind Health 18(1):9-17. 10.2486/indhealth.18.9.

Ranucci G, Di Dato F, Spagnuolo M, et al. 2014. Zinc monotherapy is effective in Wilson's disease patients with mild liver disease diagnosed in childhood: A retrospective study. Journal of Rare Diseases 9:41. https://doi.org/10.1186/1750-1172-9-41.

Raspor B, Nürnberg HW, Valenta P, et al. 1984. Studies in seawater and lake water on interactions of trace metals with humic substances isolated from marine and estuarine sediments. Mar Chem 15(3):231-249. 10.1016/0304-4203(84)90020-3.

Rauch JNGTE. 2007. Earth's anthrobiogeochemical copper cycle. Global Biogeochemical Cycles 21(2):GB2010. https://doi.org/10.1029/2006GB002850.

Reed JS, Henningson JC. 1984. Acid precipitation and drinking water supplies. J Am Water Works Assoc 76:60-65.

Rice KC. 1999. Trace-element concentrations in streambed sediment across the conterminous United States. Environ Sci Technol 33(15):2499-2504. 10.1021/es990052s.

Richards BK, Steenhuis TS, Peverly JH, et al. 1998. Metal mobility at an old, heavily loaded sludge application site. Environ Pollut 99(3):365-377. 10.1016/s0269-7491(98)00011-6.

Rieuwerts JS, Thornton I, Farago ME, et al. 2015. Factors influencing metal bioavailability in soils: preliminary investigations for the development of a critical loads approach for metals. Chemical Speciation & Bioavailability 10(2):61-75. 10.3184/095422998782775835.

Ritter WF, Eastburn RP. 2008. Leaching of heavy metals from sewage sludge through coastal plain soils. Commun Soil Sci Plant Anal 9(9):785-798. 10.1080/00103627809366853.

Rodriguez-Castro KI, Hevia-Urrutia FJ, Sturniolo GC. 2015. Wilson's disease: A review of what we have learned. World J Hepatol 7(29):2859-2870. 10.4254/wjh.v7.i29.2859.

Rohr U, Senger M, Selenka F, et al. 1999. Four years of experience with silver-copper ionization for control of legionella in a German university hospital hot water plumbing system. Clin Infect Dis 29(6):1507-1511. 10.1086/313512.

Romeu-Moreno A, Aguilar C, Arola L, et al. 1994. Respiratory toxicity of copper. Environ. Health Perspect. 102 (Suppl 3):339-340. 10.1289/ehp.94102s3339.

Romo-Kröger CM, Morales JR, Dinator MI, et al. 1994. Heavy metals in the atmosphere coming from a copper smelter in Chile. Atmospheric Environment 28(4):705-711. 10.1016/1352-2310(94)90047-7.

Rösner U. 1998. Effects of historical mining activities on surface water and groundwater - an example from northwest Arizona. Environ Geol 33(4):224-230. 10.1007/s002540050241.

Rossner Jr P, Vrbova K, Rossnerova A, et al. 2020. Gene Expression and Epigenetic Changes in Mice Following Inhalation of Copper(II) Oxide Nanoparticles. Nanomaterials. 10:550. 10.3390/nano10030550.

Roy WR. 1994. Groundwater contamination form municipal landfills in the USA. In: Adriano D, ed. Contamination of groundwaters: Case studies. Northwood, UK: Scientific Review, 411-446.

Roychoudury S, Massanyi P, Bulla J, et al. 2010. In vitro copper toxicity on rabbit spermatozoa motility, morphology and cell membrane integrity. J Environ Sci Health A Tox Hazard Subst Environ Eng 45(12):1482-1491. https://doi.org/10.1080/10934529.2010.506092.

Rush R. 1990a. Acute dermal toxicity study in rabbits with Komeen. Spencerville, Ohio: Springborn Laboratories, Inc.

Rush R. 1990b. Acute oral toxicity study in rats with Komeen (EPA-FIFRA). Spencerville, Ohio: Springborn Laboratories, Inc.

Rush R. 1991. Acute inhalation toxicity study in rats with komeen (EPA-FIFRA). Spencerville, Ohio: Life Sciences Division, Springborn Laboratories, Inc.

Rush R. 1990c. Acute oral toxicity study in rats with K-Tea. Spencerville, Ohio: Springborn Laboratories, Inc.

Sacco JE, Dodd KW, Kirkpatrick SI, et al. 2013. Voluntary food fortification in the United States: Potential for excessive intakes. Eur J Clin Nutr 67(6):592-597. 10.1038/ejcn.2013.51.

Saenko E, Yaropolov A, Harris E. 1994. Biological function of ceruloplasmin expressed through copper-binding sites and cellular receptor. J Trace Elem Exp Med 7:69-88.

Saha A, Karnik A, Sathawara N, et al. 2008. Ceruloplasmin as a marker of occupational copper exposure. Journal of Exposure Science and Environmetology 18:332-337. https://doi.org/10.1038/jes.2008.2.

Sakhaee E, Emadi L, Siahkouhi H. 2016b. Histopathological evaluation of supportive effects of Rosa damascene on mice testes, following long term administration of copper sulfate. Asian Pacific Journal of Reproduction 5(1):46-50. https://doi.org/10.1016/j.apjr.2015.12.008.

Sakhaee E, Emadi L, Abshenas J, et al. 2012. Evaluation of epididymal sperm quality following experimentally induced copper poisoning in male rats. Andrologia 44(Suppl 1):110-116. 10.1111/j.1439-0272.2010.01147.x.

Sakhaee E, Abshenas J, Emadi L, et al. 2014. Effects of vitamin C on epididymal sperm quality following experimentally induced copper poisoning in mice. Comp Clin Path 23(1):181-186. https://doi.org/10.1007/s00580-012-1592-5.

Sakhaee E, Emadi L, Azari O, et al. 2016a. Effects of Cuminum cyminum L. essential oil on some epididymal sperm parameters and histopathology of testes following experimentally induced copper poisoning in mice. Andrologia 48(5):542-547. 10.1111/and.12476.

Saleha Banu B, Ishaq M, Danadevi K, et al. 2004. DNA damage in leukocytes of mice treated with copper sulfate. Food Chem Toxicol 42(12):1931-1936. 10.1016/j.fct.2004.07.007.

Saltzer EI, Wilson JW. 1968. Allergic contact dermatitis due to copper. Arch Dermatol 98(4):375-376. 10.1001/archderm.1968.01610160049009.

Samuelsson C, Björkman B. 2014. Chapter 7 - Copper recycling. In: Worrell E & Reuter MA, ed. Handbook of recycling. Boston: Elsevier, 85-94.

8. REFERENCES

Sani A, Gaya MB, Abubakar FA. 2016. Determination of some heavy metals in selected cosmetic products sold in kano metropolis, Nigeria. Toxicology Reports 3:866-869. https://doi.org/10.1016/j.toxrep.2016.11.001.

Sansinanea AS, Cerone SI, Elperding A, et al. 1996. Glucose-6-phosphate dehydrogenase activity in erythrocytes from chronically copper-poisoned sheep. Comp. Biochem. Physiol. C Pharmacol. Toxicol. Endocrinol. 114(3):197-200. 10.1016/0742-8413(96)00034-5.

Santschi PH, Nixon S, Pilson M, et al. 1984. Accumulation of sediments, trace metals (Pb, Cu) and total hydrocarbons in Narragansett Bay, Rhode Island. Estuar Coast Shelf Sci 19(4):427-449. 10.1016/0272-7714(84)90095-7.

Scarl RT, Lawrence CM, Gordon HM, et al. 2017. STEAP4: its emerging role in metabolism and homeostasis of cellular iron and copper. J. Endocrinol. 234(3):R123-R134. 10.1530/JOE-16-0594.

Scharenberg W, Ebeling E. 1996. Distribution of heavy metals in a woodland food web. Bull. Environ. Contam. Toxicol. 56(3):389-396. 10.1007/s001289900056.

Scheinberg IH, Sternlieb I. 1996. Wilson disease and idiopathic copper toxicosis. Am J Clin Nutr 63(5):842S-845S. 10.1093/ajcn/63.5.842.

Schilsky M. 2019. Wilson disease: Epidemiology and pathogenesis. UpToDate. https://www.uptodate.com/contents/wilson-disease-epidemiology-and-pathogenesis.

Schipper BW, Lin H-C, Meloni MA, et al. 2018. Estimating global copper demand until 2100 with regression and stock dynamics. Resources, Conservation and Recycling 132:28-36. https://doi.org/10.1016/j.resconrec.2018.01.004.

Schlesinger ME, King MJ, Sole KC, et al. 2011a. Chapter 1 - Overview. In: Schlesinger ME, King MJ, Sole KC, et al., ed. Extractive metallurgy of copper (fifth edition). Oxford: Elsevier, 1-12.

Schlesinger ME, King MJ, Sole KC, et al. 2011b. Chapter 2 - Production and use. In: Schlesinger ME, King MJ, Sole KC, et al., ed. Extractive metallurgy of copper (fifth edition). Oxford: Elsevier, 13-30.

Schlesinger ME, King MJ, Sole KC, et al. 2011c. Chapter 18 - Collection and processing of recycled copper. In: Schlesinger ME, King MJ, Sole KC, et al., ed. Extractive metallurgy of copper (fifth edition). Oxford: Elsevier, 373-387.

Schock MR, Sandvig AM. 2009. Long-term effects of orthophosphate treatment on copper concentration. Journal AWWA 101(7):71-82. https://doi.org/10.1002/j.1551-8833.2009.tb09925.x.

Schraufnagel DE. 2020. The health effects of ultrafine particles. Experimental & Molecular Medicine 52(3):311-371. https://doi.org/10.1038/s12276-020-0403-3.

Schroeder HA, Nason AP, Tipton IH, et al. 1966. Essential trace metals in man: copper. J Chronic Dis 19(9):1007-1034. 10.1016/0021-9681(66)90033-6.

Schroeder WH, Dobson M, Kane DM, et al. 1987. Toxic Trace Elements Associated with Airborne Particulate Matter: A Review. JAPCA 37(11):1267-1285. 10.1080/08940630.1987.10466321.

Schwerdtle T, Hamann I, Jahnke G, et al. 2007. Impact of copper on the induction and repair of oxidative DNA damage, poly(ADP-ribosyl)ation and PARP-1 activity. Mol Nutr Food Res 51(2):201-210. 10.1002/mnfr.200600107.

Sedlak DL, Phinney JT, Bedsworth WW. 1997. Strongly complexed Cu and Ni in wastewater effluents and surface runoff. Environ Sci Technol 31(10):3010-3016. 10.1021/es970271i.

Seffner W, Schiller F, Lippold U, et al. 1997. Experimental induction of liver fibrosis in young guinea pigs by combined application of copper sulphate and aflatoxin B1. Toxicol Lett 92(3):161-172. 10.1016/s0378-4274(97)00052-0.

Sethi S, Grover S, Khodaskar MB. 1993. Role of copper in Indian childhood cirrhosis. Ann Trop Paediatr 13(1):3-5. 10.1080/02724936.1993.11747618.

Seven PT, Baykalir BG, Seven I, et al. 2018. The protective effects of chrysin and flunixin meglumine against excess copper in male rats. Turkish Journal of Veterinary and Animal Sciences 42(5):376-387. 10.3906/vet-1710-70.

Sharda B, Bhandari B. 1984. Copper concentration in plasma, cells, liver, urine, hair and nails in hepatobiliary disorders in children. Indian Pediatr 21(2):167-171.

Sharma A. 2011. Acute copper sulphate poisoning: A case report and review of literature. Medico-Legal Update 11(2):7-8. https://doi.org/10.1016/j.resuscitation.2008.02.017.

Sharma VK, Millero FJ. 1988. Oxidation of copper(I) in seawater. Environmental Science & Technology 22(7):768-771. 10.1021/es00172a004.

Shen SG, Li H, Zhao YY, et al. 2005. The distribution patterns of trace elements in the blood and organs in a rabbit experimental model of copper pollution and study of haematology and biochemistry parameters. Environ Toxicol Pharmacol 19(2):379-384. 10.1016/j.etap.2004.09.008.

Shen XL, Yu JH, Zhang DF, et al. 2014. Positive relationship between mortality from Alzheimer's disease and soil metal concentration in mainland China. J Alzheimers Dis 42(3):893-900. 10.3233/JAD-140153.

Shubber E, Amin NS, El-Adhami BH. 1998. Cytogenetic effects of copper-containing intrauterine contraceptive device (IUCD) on blood lymphocytes. Mutat. Res. 417(2-3):57-63. 10.1016/s1383-5718(98)00090-4.

Shukla AK, Pragya P, Chowdhuri DK. 2011. A modified alkaline Comet assay for in vivo detection of oxidative DNA damage in Drosophila melanogaster. Mutat. Res. 726(2):222-226. 10.1016/j.mrgentox.2011.09.017.

Sideris EG, Charalambous SC, Tsolomyty A, et al. 1988. Mutagenesis; carcinogenesis and the metal elements--DNA interaction. Prog. Clin. Biol. Res 259:13-25.

Sina J, Bean C, Dysart G, et al. 1983. Evaluation of the alkaline elution/rat hepatocyte assay as a predictor of carcinogenic/mutagenic potential. Mutat. Res. 113:357-391.

Singh I. 1983. Induction of reverse mutation and mitotic gene conversion by some metal compounds in Saccharomyces cerevisiae. Mutat. Res. 117:149-152.

Singh N, Duan H, Ogunseitan OA, et al. 2019. Toxicity trends in E-Waste: A comparative analysis of metals in discarded mobile phones. J Hazard Mater 380:120898. 10.1016/j.jhazmat.2019.120898.

Sinkovic A, Strdin A, Svensek F. 2008. Severe acute copper sulphate poisoning: a case report. Arh Hig Rada Toksikol 59(1):31-35. 10.2478/10004-1254-59-2008-1847.

Sirover MA, Loeb LA. 1976. Infidelity of DNA synthesis in vitro: screening for potential metal mutagens or carcinogens. Science 194(4272):1434-1436. 10.1126/science.1006310.

Sood N, Verma P. 2011. Life-threatening haemolysis in a patient with acute copper sulphate poisoning. Indian J Anaesth 55(2):204-205. 10.4103/0019-5049.79878.

Sora S, Carbone M, Pacciarini M, et al. 1986. Disomic and diploid meiotic products induced in Saccharomyuces cerevisiae by the salts of 27 elements. Mutagenesis 1(1):21-28.

Spitalny KC, Brondum J, Vogt RL, et al. 1984. Drinking-water-induced copper intoxication in a Vermont family. Pediatrics 74(6):1103-1106.

Stark P. 1981. Vineyard sprayer's lung - a rare occupational disease. J Can Assoc Radiol 32(3):183-184.

Stephen G, Darren AL, Mark NG. 2012. Control of new copper corrosion in high-alkalinity drinking water. Journal (American Water Works Association) 104(1):E15-E25.

Stephenson T, Lester JN. 1987. Heavy metal behavior during the activated sludge process II. Insoluble metal removal mechanisms. Science of The Total Environment 63:215-230. 10.1016/0048-9697(87)90047-7.

Strain WH, Hershey CO, McInnes S, et al. 1984. Hazards to groundwater from acid rain. Trace Subst Environ Health 18:178-184.

Strickland G, Beckner W, Leu M. 1972. Absorption of copper in homozygotes and heterozygotes for Wilson's disease and controls: isotope tracer studies with 67Cu and 64Cu. Clin Sci 43:617-625.

Suciu I, Prodan L, Lazar V, et al. 1981. Research on copper poisoning. Med Lav 72(3):190-197.

Sugawara N, Li D, Katakura M, et al. 1994. Biliary excretion of copper in Fischer rats treated with copper salt and in Long-Evans cinnamon (LEC) rats with an inherently abnormal copper metabolism. Biol Trace Elem Res 46(1-2):125-134. 10.1007/BF02790073.

Sugawara N, Sugawara C, Katakura M, et al. 1991. Harmful effect of administration of copper on LEC rats. Res Commun Chem Pathol Pharmacol 73(3):289-297.

Sugawara N, Sugawara C, Li D, et al. 1992. Copper metabolism in new mutant Long-Evans Cinnamon (LEC) rats causing hereditary hepatitis: Gastrointestinal absorption and distribution of radioisotopic copper (64Cu). Res Commun Chem Pathol Pharmacol 76(2):233-243.

Suttle NF, Mills CF. 1966. Studies of the toxicity of copper to pigs. 1. Effects of oral supplements of zinc and iron salts on the development of copper toxicosis. Br. J. Nutr. 20(2):135-148.

Suzuki KT, Kanno S, Misawa S, et al. 1995. Copper metabolism leading to and following acute hepatitis in LEC rats. Toxicology 97(1-3):81-92. 10.1016/0300-483x(94)02927-m.

Swastika M, Harahap AR, Panggalo LV, et al. 2020. Determining a critical threshold for G6PD activity below which red blood cell response to oxidative stress is poor. Malar. J. 19(1):208. 10.1186/s12936-020-03272-y.

Sweet CW, Vermette SJ, Landsberger S. 2002. Sources of toxic trace elements in urban air in Illinois. Environ Sci Technol 27(12):2502-2510. 10.1021/es00048a030.

Tang H, Xu M, Shi F, et al. 2018. Effects and mechanism of nano-copper exposure on hepatic cytochrome P450 enzymes in rats. Int J Mol Sci 19(7):2140. 10.3390/ijms19072140.

Tanner MS. 1998. Role of copper in Indian childhood cirrhosis. Am J Clin Nutr 67(5):1074S-1081S. https://doi.org/10.1093/ajcn/67.5.1074S.

Tanner MS, Kantarjian AH, Bhave SA, et al. 1983. Early introduction of copper-contaminated animal milk feeds as a possible cause of Indian childhood cirrhosis. Lancet 2:992-995.

Tapia L, Gonzalez-Aguero M, Cisternas F, et al. 2004. Metallothionein is crucial for safe intracellular copper storage and cell survival at normal and supra-physiological exposure levels. Biochem. J. 378(2):617-624. https://dx.doi.org/10.1042%2FBJ20031174.

Taylor GJ, Crowder AA. 1983. Accumulation of atmospherically deposited metals in wetland soils of Sudbury, Ontario. Water Air Soil Pollut 19(1):29-42. 10.1007/bf00176793.

Taylor AA, Tsuji JS, Garry MR, et al. 2020. Critical review of exposure and effects: Implications for setting regulatory health criteria for ingested copper. Environ. Manage. 65(1):131-159. 10.1007/s00267-019-01234-y.

Theresa OC, Onebunne OC, Dorcas W, et al. 2011. Potentially Toxic Metals Exposure From Body Creams Sold In Lagos, Nigeria. Researcher 3(1): 30-37.

Tian Y, Wu B, Li X, et al. 2019. The resveratrol alleviates the hepatic toxicity of CuSO4 in the rat. Biol Trace Elem Res 187(2):464-471. 10.1007/s12011-018-1398-7.

Tinwell H, Ashby J. 1990. Inactivity of copper sulphate in a mouse bone-marrow micronucleus assay. Mutat. Res. 245(3):223-226. 10.1016/0165-7992(90)90054-n.

Toyokuni S, Tanaka T, Nishiyama Y, et al. 1996. Induction of renal cell carcinoma in male Wistar rats treated with cupric nitrilotriacetate. Lab Invest 75(2):239-248.

TRI18. 2020. TRI explorer: Providing access to EPA's toxics release inventory data. . Washington, DC: Toxics Release Inventory. U.S. Environmental Protection Agency. http://www.epa.gov/triexplorer/. July 28, 2020.

Troost FJ, Brummer RJ, Dainty JR, et al. 2003. Iron supplements inhibit zinc but not copper absorption in vivo in ileostomy subjects. Am J Clin Nutr 78(5):1018-1023. 10.1093/ajcn/78.5.1018.

Tso WW, Fung WP. 1981. Mutagenicity of metallic cations. Toxicol Lett 8(4-5):195-200. 10.1016/0378-4274(81)90100-4.

Turek NF, Kasten L, Lytle DA, et al. 2011. Impact of plumbing age on copper levels in drinking water. Journal of Water Supply: Research and Technology-Aqua 60(1):1-15. 10.2166/aqua.2011.014.

Turnlund JR, Swanson CA, King JC. 1983. Copper absorption and retention in pregnant women fed diets based on animal and plant proteins. J. Nutr. 113(11):2346-2352. 10.1093/jn/113.11.2346.

Turnlund JR, King JC, Gong B. 1985. A stable isotope study of copper absorption in young men: Effect of phytate and alpha-cellulose 1-3. Am J Clin Nutr 42:18-23. https://doi.org/10.1093/ajcn/42.1.18.

Turnlund JR, Keyes WR, Anderson HL, et al. 1989. Copper absorption and retention in young men at three levels of dietary copper by use of the stable isotope 65Cu1-4. Am J Clin Nutr 49(5):870-878. https://doi.org/10.1093/ajcn/49.5.870.

Turnlund JR, Keyes WR, Peiffer GL, et al. 1998. Copper absorption, excretion, and retention by young men consuming low dietary copper determined by using the stable isotope 65Cu. Am J Clin Nutr 67(6):1219-1225. 10.1093/ajcn/67.6.1219.

Turnlund JR, Keyes WR, Kim SK, et al. 2005. Long-term high copper intake: effects on copper absorption, retention, and homeostasis in men. Am J Clin Nutr 81(4):822-828. 10.1093/ajcn/81.4.822.

Turnlund JR, Michel MC, Keyes WR, et al. 1982. Copper absorption in elderly men determined by using stable 65Cu. Am J Clin Nutr 36(4):587-591. https://doi.org/10.1093/ajcn/36.4.587.

Turnlund JR, Wada L, King JC, et al. 1988. Copper absorption in young men fed adequate and low zinc diets. Biol Trace Elem Res 17:31-41. 10.1007/BF02795445.

Turnlund JR, Jacob RA, Keen CL, et al. 2004. Long-term high copper intake: effects on indexes of copper status, antioxidant status, and immune function in young men. Am J Clin Nutr 79(6):1037-1044. 10.1093/ajcn/79.6.1037.

Tyler LD, McBride MB. 1982. Mobility and extractability of cadmium, copper, nickel, and zinc in organic and mineral soil columns. Soil Sci 134(3):198-205.

Urbina-Cano P, Bobadilla-Morales L, Ramirez-Herrera MA, et al. 2006. DNA damage in mouse lymphocytes exposed to curcumin and copper. J. Appl. Genet. 47(4):377-382. 10.1007/BF03194648.

USDA. 2020. What we eat in America, NHANES 2017-2018. Table 1. Nutrient intakes from food and beverages: Mean amounts consumed per individual, by gender and age, in the United States, 2017-2018. U.S. Department of Agriculture, Agricultural Research Service.

USDT. 2018. Title 31- Money and finance. Denominations, specifications, and design of coins. U.S. Department of the Treasury. United States Code. https://www.govinfo.gov/content/pkg/USCODE-2018-title31/pdf/USCODE-2018-title31-subtitleIV-chap51-subchapII-sec5112.pdf.

USGS. 2009–. Copper - A metal for the ages: U.S. Geological Survey fact sheet 2009-3031. U.S. Department of the Interior, U.S. Geological Survey. https://pubs.usgs.gov/fs/2009/3031/FS2009-3031.pdf.

USGS. 2009b. Quality of water from domestic wells in principal aquifers of the United States, 1991–2004: Overview of major findings. Reston, Virginia: US Geological Survey. https://pubs.usgs.gov/circ/circ1332/includes/circ1332.pdf. October 21, 2020.

USGS. 2013. Geochemical and mineralogical data for soils of the conterminous United States: U.S. Geological Survey data series 801. U.S. Department of the Interior, U.S. Geological Survey.

USGS. 2016. National geochemical database: Soil. U.S. Department of the Interior, U.S. Geological Survey.

USGS. 2017a. 2013-2017 county-level pesticide use estimates. Estimated annual agricultural pesticide use. Pesticide national synthesis project. U.S. Department of the Interior, U.S. Geological Survey. https://water.usgs.gov/nawqa/pnsp/usage/maps/county-level/. August 14, 2020.

USGS. 2017b. Copper [Advance Release]. 2015 Minerals Yearbook. U.S. Department of the Interior, U.S. Geological Survey. https://s3-us-west-2.amazonaws.com/prd-wret/assets/palladium/production/mineral-pubs/copper/myb1-2015-coppe.pdf. October 21, 2020.

USGS. 2018. Regional stream quality assessment (RSQA). U.S. Department of the Interior, U.S. Geological Survey. https://webapps.usgs.gov/rsqa/#!/download. July 28, 2020.

USGS. 2020a. Copper data sheet. Mineral commodity summaries. U.S. Department of the Interior, U.S. Geological Survey. https://pubs.usgs.gov/periodicals/mcs2020/mcs2020-copper.pdf.

USGS. 2020b. Datasets from groundwater-quality and select quality-control data from the National Water-Quality Assessment Project, January through December 2016, and previously unpublished data from 2013 to 2015. U.S. Department of the Interior, U.S. Geological Survey. https://doi.org/10.5066/P9W4RR74. October 21, 2020.

Valdes A, Zanobetti A, Halonen JI, et al. 2012. Elemental concentrations of ambient particles and cause specific mortality in Santiago, Chile: A time series study. Environ Health 11:82. https://doi.org/10.1186/1476-069x-11-82.

Valsami S, Stamoulis K, Lydataki E, et al. 2012. Acute copper sulphate poisoning: a forgotten cause of severe intravascular haemolysis. Br. J. Haematol. 156(3):294. 10.1111/j.1365-2141.2011.08881.x.

Van den Berg GJ, Beynen AC. 1992. Influence of ascorbic acid supplementation on copper metabolism in rats. Br. J. Nutr. 68(3):701-715. 10.1079/bjn19920127.

van den Berghe PV, Klomp LW. 2009. New developments in the regulation of intestinal copper absorption. Nutr Rev 67(11):658-672. 10.1111/j.1753-4887.2009.00250.x.

Varada KR, Harper RG, Wapnir RA. 1993. Development of copper intestinal absorption in the rat. Biochem Med Metab Biol 50(3):277-283. 10.1006/bmmb.1993.1069.

Venugopal V, Gopakumar K. 2017. Shellfish: Nutritive Value, Health Benefits, and Consumer Safety. Comprehensive Reviews in Food Science and Food Safety 16(6):1219-1242. https://doi.org/10.1111/1541-4337.12312.

Vieira J, Oliveira P, Juliano Y, et al. 2012. Urinary copper excretion before and after oral intake of d-penicillamine in parents of patients with Wilson's disease. Digestive and Liver Diseases 44(4):323–327. 10.1016/j.dld.2011.11.001.

Villar TG. 1974. Vineyard sprayer's lung. Clinical aspects. Am Rev Respir Dis 110(5):545-555. 10.1164/arrd.1974.110.5.545.

Villar TG, Nogueira T. 1980. Radiology and respiratory function in "vineyard sprayer's lung". Bronchopneumologie 30(1):61-67.

Vincent M, Hartemann P, Engels-Deutsch M. 2016. Antimicrobial applications of copper. Int J Hyg Environ Health 219(7):585-591. 10.1016/j.ijheh.2016.06.003.

Vyskocil V, Viau C. 1999. Assessment of Molybdenum Toxicity in Humans. Journal of Applied Toxicology. 19: 185-192.

Wahlin P, Berkowicz R, Palmgren, F. 2006. Characterisation of traffic-generated particulate matter in Copenhagen. Atmos Environ 40(12):2151-2159. https://doi.org/10.1016/j.atmosenv.2005.11.049.

Wake SA, Mercer JF. 1985. Induction of metallothionein mRNA in rat liver and kidney after copper chloride injection. Biochem. J. 228(2):425-432. 10.1042/bj2280425.

Walker WR, Reeves RR. 1977. Perfusion of intact skin by a saline solution of bis(glycinato) copper(II). Bioinorg Chem 7(3):271-276. 10.1016/s0006-3061(00)80100-3.

Walsh FM, Crosson FJ, Bayley M, et al. 1977. Acute copper intoxication. Pathophysiology and therapy with a case report. Am J Dis Child 131(2):149-151. 10.1001/archpedi.1977.02120150031005.

Wang SC, Borison HL. 1951. Copper sulphate emesis; a study of afferent pathways from the gastrointestinal tract. Am. J. Physiol. 164(2):520-526. 10.1152/ajplegacy.1951.164.2.520.

Wang C, Hao L, Liu C, et al. 2020. Associations between fine particulate matter constituents and daily cardiovascular mortality in Shanghai, China. Ecotoxicol. Environ. Saf. 191:110154. 10.1016/j.ecoenv.2019.110154.

Wang YX, Wang P, Feng W, et al. 2017. Relationships between seminal plasma metals/metalloids and semen quality, sperm apoptosis and DNA integrity. Environ. Pollut. 224:224-234. 10.1016/j.envpol.2017.01.083.

Wapnir R, Devas G, Solans C. 1993. Inhibition of intestinal copper absorption by divalent cations and low-molecular-weight ligands in the rat. Biol Trace Elem Res 36:291-305. https://doi.org/10.1007/BF02783963.

Weant GE. 1985. Sources of copper air emissions. Research Triangle Park, NC: Air and Energy Engineering Research Laboratory, U.S. Environmental Protection Agency.

Weber PM, O'Reilly S, Pollycove M, et al. 1969. Gastrointestinal absorption of copper: Studies with 64Cu, 95Zr, a whole-body counter and the scintillation camera. J Nucl Med 10(9):591-596.

Weiss KC, Linder MC. 1985. Copper transport in rats involving a new plasma protein. Am. J. Physiol. 249(1):E77-E88. https://doi.org/10.1152/ajpendo.1985.249.1.E77.

Whanger PD, Weswig PH. 1971. Effect of supplementary zinc on the intracellular distribution of hepatic copper in rats. J. Nutr. 101(8):1093-1097. 10.1093/jn/101.8.1093.

Whitall D, Hively WD, Leight AK, et al. 2010. Pollutant fate and spatio-temporal variability in the choptank river estuary: Factors influencing water quality. Sci Total Environ 408(9):2096-2108. https://doi.org/10.1016/j.scitotenv.2010.01.006.

Whitehead TP, Ward MH, Colt JS, et al. 2015. Dust metal loadings and the risk of childhood acute lymphoblastic leukemia. J Expo Sci Environ Epidemiol 25(6):593-598. 10.1038/jes.2015.9.

WHO. 2004. Copper in Drinking-water, Background document for development of WHO Guidelines for Drinking-water Quality. Geneva, Switzerland: World Health Organization.

WHO. 2010. WHO guidelines for indoor air quality: Selected pollutants. Geneva, Switzerland: World Health Organization. https://www.euro.who.int/__data/assets/pdf_file/0009/128169/e94535.pdf.

WHO. 2017. Guidelines for drinking-water quality. Fourth edition incorporating the first addendum. Geneva, Switzerland: World Health Organization. https://apps.who.int/iris/bitstream/handle/10665/254637/9789241549950-eng.pdf;jsessionid=8AD40533ABAFCC37B8C6ABE4EFF2AC2F?sequence=1.

Wildemeersch D, Sabbe PJ, Dowsett MG, et al. 2014. Assessment of copper corrosion from frameless copper IUDs after long-term in utero residence. Contraception 90(4):454-459. 10.1016/j.contraception.2014.05.009.

Wilhelm M, Hafner D, Lombeck I. 1991. Monitoring of cadmium, copper, lead and zinc status in young children using toenails: Comparison with scalp hair. Sci. Total. Environ 103(2-3):199-207. https://doi.org/10.1016/0048-9697(91)90145-5.

Windholz M. 1983. The Merck Index. 10th ed. Rahway, NJ: Merck & Co., 358-359; 2484-2485.

Wong PK. 1988. Mutagenicity of heavy metals. Bull. Environ. Contam. Toxicol. 40(4):597-603. 10.1007/BF01688386.

Woo SH, Lee DS, Lim SR. 2016. Potential resource and toxicity impacts from metals in waste electronic devices. Integr Environ Assess Manag 12(2):364-370. 10.1002/ieam.1710.

WQP. 2020. Water quality portal data: Copper. Advisory Committee on Water Information (ACWI); Agricultural Research Service (ARS); Environmental Protection Agency (EPA); National Water Quality Monitoring Council (NWQMC); United States Geological Survey (USGS). . https://www.waterqualitydata.us/portal/. August 18, 2020.

Wu J, Laird DA, Thompson ML. 1999. Sorption and desorption of copper on soil clay components. J Environ Qual 28(1):334-338. 10.2134/jeq1999.00472425002800010041x.

Wu X, Jackson RT, Khan SA, et al. 2018. Human milk nutrient composition in the United States: Current knowledge, challenges, and research needs. Curr Dev Nutr 2(7):nzy025. 10.1093/cdn/nzy025.

Wu H, Guo H, Liu H, et al. 2020. Copper sulfate-induced endoplasmic reticulum stress promotes hepatic apoptosis by activating CHOP, JNK and caspase-12 signaling pathways. Ecotoxicol. Environ. Saf. 191:110236. 10.1016/j.ecoenv.2020.110236.

Xue H, Goncalves MdLS, Reutlinger M, et al. 1991. Copper(I) in fogwater: Determination and interactions with sulfite. Environ Sci Technol 25(10):1716-1722. 10.1021/es00022a006.

Yadla M, John P, Kanth S, et al. 2015. An unusual case of acute kidney injury due to poisoning with blue stone. Hong Kong J Nephrol 17(2):26-27. 10.1016/j.hkjn.2015.08.002.

Yalcin SS, Yalcin S, Gucus AI. 2015. Zinc and copper concentrations in breast milk during the first nine months of lactation: A longitudinal study. Pediatrics 135:S13-S14. https://doi.org/10.1542/peds.2014-3330X.

Yamada Y. Prosser RA. 2018. Copper in the suprachiasmatic circadian clock: A possible link between multiple circadian oscillators. Eur J Neurosci. 51: 47-70. 10.1111/ejn.14181

Yamamoto K, Ngan MP, Takeda N, et al. 2004. Differential activity of drugs to induce emesis and pica behavior in Suncus murinus (house musk shrew) and rats. Physiol. Behav 83(1):151-156. 10.1016/j.physbeh.2004.08.006.

Yamane Y, Sakai K, Umeda T, et al. 1984. Suppressive effect of cupric acetate on DNA alkylation, DNA synthesis and tumorigenesis in the liver of dimethylnitrosamine-treated rats. Gan 75(12):1062-1069.

Yang CC, Wu ML, Deng JF. 2004. Prolonged hemolysis and methemoglobinemia following organic copper fungicide ingestion. Vet. Hum. Toxicol 46(6):321-323.

Yang TH, Yuan TH, Hwang YH, et al. 2016. Increased inflammation in rheumatoid arthritis patients living where farm soils contain high levels of copper. J Formos Med Assoc 115(11):991-996. 10.1016/j.jfma.2015.10.001.

Yang X, Li Y, Li J, et al. 2020. Associations between exposure to metal mixtures and birth weight. Environ Pollut 263. 10.1016/j.envpol.2020.114537.

Yannoni CC, Piorkowski T. 1995. Profile of lead and copper levels in house plumbing and service pipe. J New Engl Water Works Assoc 109(3):192-210.

Yeats PA. 1988. The distribution of trace metals in ocean waters. Sci Total Environ 72:131-149. https://doi.org/10.1016/0048-9697(88)90012-5.

Zhong W-S, Ren T, Zhao L-J. 2015. Determination of Pb (Lead), Cd (Cadmium), Cr (Chromium), Cu, (Copper), and Ni (Nickel) in Chinese tea with high-resolution continuum source graphite furnace atomic absorption spectometry. J Food Drug Anal 24(1):46-55. https://doi.org/10.1016/j.jfda.2015.04.010.

Zietz BP, De Vergara JD, Dunkleberg H. 2003b. Copper concentrations in tap water and possible effects on infant's health - results of a study in lower Saxony, Germany. Environ Res 92(2):129-138. https://doi.org/10.1016/s0013-9351(03)00037-9.

Zietz BP, Dieter HH, Lakomek M, et al. 2003a. Epidemiological investigation on chronic coppertoxicity to children exposed via the public drinking water supply. Sci. Total. Environ 302(1-3): 127-144. https://doi.org/10.1016/s0048-9697(02)00399-6.

APPENDIX A. ATSDR MINIMAL RISK LEVELS AND WORKSHEETS

MRLs are derived when reliable and sufficient data exist to identify the target organ(s) of effect or the most sensitive health effect(s) for a specific duration for a given route of exposure. An MRL is an estimate of the daily human exposure to a hazardous substance that is likely to be without appreciable risk of adverse non-cancer health effects over a specified route and duration of exposure. MRLs are based on non-cancer health effects only; cancer effects are not considered. These substance-specific estimates, which are intended to serve as screening levels, are used by ATSDR health assessors to identify contaminants and potential health effects that may be of concern at hazardous waste sites. It is important to note that MRLs are not intended to define clean-up or action levels.

MRLs are derived for hazardous substances using the NOAEL/uncertainty factor approach. They are below levels that might cause adverse health effects in the people most sensitive to such chemical-induced effects. MRLs are derived for acute (1–14 days), intermediate (15–364 days), and chronic (≥365 days) durations and for the oral and inhalation routes of exposure. Currently, MRLs for the dermal route of exposure are not derived because ATSDR has not yet identified a method suitable for this route of exposure. MRLs are generally based on the most sensitive substance-induced endpoint considered to be of relevance to humans. Serious health effects (such as irreparable damage to the liver or kidneys, or birth defects) are not used as a basis for establishing MRLs. Exposure to a level above the MRL does not mean that adverse health effects will occur.

MRLs are intended only to serve as a screening tool to help public health professionals decide where to look more closely. They may also be viewed as a mechanism to identify those hazardous waste sites that are not expected to cause adverse health effects. Most MRLs contain a degree of uncertainty because of the lack of precise toxicological information on the people who might be most sensitive (e.g., infants, elderly, nutritionally or immunologically compromised) to the effects of hazardous substances. ATSDR uses a conservative (i.e., protective) approach to address this uncertainty consistent with the public health principle of prevention. Although human data are preferred, MRLs often must be based on animal studies because relevant human studies are lacking. In the absence of evidence to the contrary, ATSDR assumes that humans are more sensitive to the effects of hazardous substance than animals and that certain persons may be particularly sensitive. Thus, the resulting MRL may be as much as 100-fold below levels that have been shown to be nontoxic in laboratory animals.

Proposed MRLs undergo a rigorous review process: Health Effects/MRL Workgroup reviews within the Office of Analytics, Toxicology Section, expert panel peer reviews, and agency-wide MRL Workgroup

reviews, with participation from other federal agencies and comments from the public. They are subject to change as new information becomes available concomitant with updating the toxicological profiles. Thus, MRLs in the most recent toxicological profiles supersede previously published MRLs. For additional information regarding MRLs, please contact the Office of Analytics, Toxicology Section, Agency for Toxic Substances and Disease Registry, 1600 Clifton Road NE, Mailstop S102-1, Atlanta, Georgia 30329-4027.

APPENDIX A

MRL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Inhalation
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration inhalation MRL as evidence of a critical effect is limited.

Rationale for Not Deriving an MRL: Human toxicity data following acute-duration inhalation exposure to copper are limited to one experimental study in humans where 5 men exposed to copper-containing welding fume were examined for markers of inflammation (Markert et al. 2016). Several occupation studies report metal fume fever in workers exposed to copper dust or fumes, characterized by chills, fever, aching muscles, dryness in the mouth and throat, and headache (Armstrong et al. 1983; Gleason 1968), and copper levels of 0.075–0.12 mg/m^3 were measured in dust (Gleason 1968). These data are insufficient as the basis of an MRL because the study authors reported that the workers had likely been exposed to concentrations 2 to 3 times higher than what was measured. Respiratory effects have been observed in human case studies following acute exposure to fumes or powder containing copper substances; however, no detailed exposure information was provided (Donoso et al. 2007; Gibson et al. 2011). An animal study examined lethality, respiratory, and immunological effects of acute-duration inhalation exposure to copper in mice, and respiratory effects in hamsters (Drummond et al. 1986). In the mouse studies, high mortality was seen even among the lowest exposure groups, therefore, none of the dose (as these are typically higher) are appropriate for MRL derivation. The study in hamster weas limited to respiratory effects, and the data are insufficient to deem these effects as a critical endpoint (Drummond et al. 1986). Acute inhalation LC_{50} values of 45 and 109 mg Cu/m^3 were determined for female and male rats exposed to a copper-containing herbicide (Rush 1991). Exposure to mixed substances, such as herbicides, are not considered for MRL derivation. No death was reported in rats exposed to 1662 mg Cu/m^3 as copper oxide aerosols (Holbert 1990).

Agency Contact (Chemical Managers): Breanna Alman, MPH

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Inhalation
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration inhalation MRL as the studies for this route and duration are limited two studies in rabbits. These studies do not allow for the determination of a critical effect.

Rationale for Not Deriving an MRL: There were no studies that provided sufficient dose information on copper toxicity in humans following intermediate-duration inhalation exposure. Animal toxicity studies were limited to two studies in rabbits which only identified NOAELs for respiratory and immune effects (Johansson et al. 1983, 1984). There are insufficient toxicity data to derive an MRL as these did not evaluate comprehensive endpoints and were single dose studies testing 8 rabbits each.

Agency Contact (Chemical Managers): Breanna Alman, MPH

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Inhalation
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration inhalation MRL as evidence of a critical effect is limited.

Rationale for Not Deriving an MRL: Occupational studies demonstrate that copper is a respiratory irritant and symptoms of coughing, sneezing, thoracic pain, and runny nose have been observed in workers exposed to high levels (Askergren and Mellgren 1975; Suciu et al. 1981). Decreased hemoglobin and erythrocyte levels were observed in workers exposed to airborne copper levels of 0.64–1.05 mg/m^3 (Finelli et al. 1981). These data are limited as the exposure dose resulting in effects could not be accurately calculated. There were no chronic inhalation animal studies published.

Agency Contact (Chemical Managers): Breanna Alman, MPH

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Oral
Duration: Acute
MRL: 0.02 mg/kg/day (provisional)
Critical Effect: Gastrointestinal effects in women
Reference: Pizarro et al. 1999
Point of Departure: $BMDL_{10}$ of 0.05 mg/kg/day
Uncertainty Factor: 3
LSE Graph Key: 6
Species: Humans

MRL Summary: A provisional acute-duration oral MRL of 0.02 mg Cu/kg/day was derived for copper based on gastrointestinal effects of abdominal pain, vomiting, and nausea in female adults ingesting copper sulfate in drinking water for 2 weeks (Pizarro et al. 1999). The MRL is based on a $BMDL_{10}$ of 0.05 mg/kg/day which was divided by a total uncertainty factor of 3 for human variability; a partial uncertainty factor was applied because toxicokinetic differences among individuals should not affect the sensitivity of this direct contact effect. The acute oral MRL may be a more conservative estimate as some evidence indicates that gastrointestinal symptoms may be due in part to a direct contact effect on the gastric mucosa, influenced by the amount of copper in the stomach at a given time, especially at high exposure concentrations.

Selection of the Critical Effect: Numerous experimental studies and case reports support the identification of the gastrointestinal tract as a sensitive endpoint of toxicity in humans acutely exposed to copper in drinking water or in contaminated beverages (Araya et al. 2001, 2003a, 2003c; Chuttani et al. 1965; Gotteland et al. 2001; Knobeloch et al. 1994; Olivares et al. 2001; Pizarro et al. 1999, 2001; Spitalny et al. 1984). Pizarro et al. (1999) identified a dose-response relationship with ingestion of copper in drinking water and gastrointestinal symptoms of abdominal pain, nausea and/or vomiting among participants exposed for 2 weeks. The incidence of these symptoms was significantly higher among subjects exposed to doses ≥0.07 mg/kg/day (3 mg Cu/L; total GI symptoms in 10/60 subjects) compared to incidence reported among subjects exposed to doses ≤0.03 mg/kg/day (≤1 mg Cu/L; total GI symptoms in ≤3/60 subjects). Pizarro et al. (1999) reported no association between copper levels in drinking water and diarrhea, with or without other gastrointestinal symptoms present, as 4 cases of diarrhea occurred at each level of added copper. Eight of the twelve cases of diarrhea had presented within the first 2 weeks of the study and number of cases declined afterwards in all exposure groups. Pizarro et al. (2001) made a similar observation seven of the ten cases of diarrhea occurred during the first half of the study, unlike other symptoms which were uniformly distributed throughout the study period. The same study reported increased abdominal pain, vomiting, and nausea in women exposed to doses of 0.095 mg Cu/kg/day, equating to 5 mg Cu/L added to drinking water (Pizarro et al. 2001). Several studies report noted that nausea is the most reported gastrointestinal symptoms among adults exposed to copper in drinking water, with effects observed at doses ranging from 0.01 to 0.09 (Araya et al. 2001, 2003a, 2003c; Gotteland et al. 2001; Olivares et al. 2001). Olivares et al. (2001) reported increased incidence of nausea at the lowest copper concentrations of 4 mg Cu/L in drinking water. Araya et al. (2003a) observed a delay in gastric emptying induced by copper exposure, and Gotteland et al. (2000) reported a 36.5% increase in gastric permeability to sucrose following ingestion of 10 mg Cu/L. Both Araya et al. (2003a) and Gotteland et al.

(2000) reported that these observations were independent of gastrointestinal symptoms. It should also be noted these studies in humans tested subjects among the general population, therefore, the resulting MRL would not be applicable to sensitive populations, such as individuals with Wilson's disease.

Animal studies have also identified the gastrointestinal tract as the most sensitive target of toxicity following acute-duration oral exposure to copper. In mice exposed to 4 mg Cu/kg/day for 7 days, the intestine showed focal ulceration (Kadammattil et al. 2018). Fifteen episodes of emesis (vomiting) were observed in 4 shrews exposed to 31 mg Cu/kg in feed once (Yamamoto et al. 2004). Hepatic and renal effects have also been observed in animals following acute oral exposure to copper. At LOAELs of ranging from 4 to198 mg/kg/day, hepatic effects observed in rats and mice include inflammation, altered liver enzymes, reduced liver weight, hepatocyte necrosis, and liver hemorrhage (Alhusaini et al. 2018a, 2018b; Kadammattil et al. 2018; Khushboo et al. 2018). Renal effects observed in rats include altered kidney enzymes and histopathological abnormalities following acute oral exposure to copper doses of 10 to 119 mg/kg/day (Alharbi et al. 2019; Khushboo et al. 2018). Additionally, altered body weight gain and spleen effects have been observed in mice at doses as low as 3.4 to 4 mg/kg/day (Franke et al. 2006; Kadammattil et al. 2018). See Table A-1 for a summary of all relevant NOAEL and LOAEL values for gastrointestinal effects which were considered for MRL development.

It is suspected that acute copper toxicity may be a direct contact effect associated with the concentration of copper in the stomach at a specific time rather than just dosage throughout the day (Donohue 1997). In dogs exposed intragastrically to single doses of copper sulfate, the neural pathways of some dogs, either the vagus nerve, sympathetic nerve, or both, were severed (Wang and Borison 1951). When the neural pathway was severed, dogs showed increased response threshold and response latency, with the greatest effects on dogs with both pathways severed. This led the study authors to hypothesize a biphasic mechanism of copper sulfate induced emesis, with copper sulfate having a direct contact effect on the peripheral nervous system followed by a systemic effect on the central nervous system associated with absorbed copper intake (Wang and Borison 1951). A study in shrews injected with copper sulfate supports this hypothesis, as vagotomized (i.e., having underwent a surgical procedure to sever the vagus nerve that controls the digestive system) shrews had a significant decrease of emetic episodes and longer latency to the first episode, compared to exposed non-vagotomized controls (Horn et al. 2014). This study indicates that low doses of copper activate the vagal afferent pathways to produce emesis, while higher doses generated emesis independent of an intact vagus nerve (Horn et al. 2014). Several other studies in dogs and ferrets demonstrate that copper sulfate induced emesis results from contact in the stomach mediated by the vagus nerve (Makale and King 1992), and also show that 5-HT_4 receptors and abdominal vagal afferents are closely associated and play a role inducing vomiting (Bhandari and Andrews 1991; Fukui et al. 1994).

Table A-1. Summary of Repeat Exposure Gastrointestinal NOAEL and LOAEL Values of Acute-Duration Oral Exposure to Copper

Species (sex)	Frequency/ Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Human (F)	2 weeks Daily	0.03	0.07	Abdominal pain, nausea, and/or vomiting	Pizarro et al. 1999
Human (F)	1 week Daily		0.1	Nausea, vomiting, and/or abdominal pain	Pizarro et al. 2001
Swiss albino mice (NS)	7 days Daily	2	4	Intestine showing focal ulceration	Kadammattil et al. 2018

F=female; LOAEL = lowest-observed adverse-effect level; NOAEL = no-observed-adverse-effect level; NS=not specified; SLOAEL = serious lowest-observed-effect levels

Selection of the Principal Study: The Pizarro et al. (1999) study identified the lowest LOAEL and accompanying NOAEL among acute repeat exposure studies for gastrointestinal symptoms. In addition, the Pizarro et al. (1999) study is a longer-duration study and the exposure scenario closely mimics that of a population drinking copper-contaminated water and as such was selected as the critical study.

Summary of the Principal Study:

Pizarro, F., Olivares, M., Uauy, R., et al. 1999. Acute gastrointestinal effects of graded levels of copper in drinking water. Environmental Health Perspectives 107:117-121.

A group of 60 healthy women in Chile were divided into four exposure sequence groups, with mean ages within each group ranging from 32.9 to 36.3 years. The mean body weight of the participants was 64 kg. Each group consumed water containing 0, 1, 3, or 5 mg/L ionic copper as copper sulfate pentahydrate (0.0006, 0.0272, 0.0731, and 0.124 mg Cu/kg/day, respectively) for a 2-week period followed by a 1-week rest, followed by the next dose of copper in the sequence. Each group of women was assigned to a different order of copper concentrations to consume over an 11-week period. For example, the first group was assigned to consume the control group drinking water for 2 weeks followed by a 1-week rest period, then drank the water containing 1 mg Cu/L for 2 weeks followed by a 1-week rest. This process continued in the same group with the water containing 3 mg Cu/L and 5 mg Cu/L. Ultimately, each dose was tested in all 60 women, therefore, n=60 for each dose group, and each woman served as their own control. Each week, the subjects received a bottle containing copper sulfate solution and were asked to mix the contents of the bottle with 3 L of their drinking water. The subjects recorded daily water consumption, and any reported any symptoms during each 2-week exposure period. If a participant presented diarrhea, abdominal pain, or vomiting, they were told not to ingest copper-containing water for the next 2 days and consumption began once symptoms disappeared. Blood samples were collected 1 week before the study, at the end of the first 2-week exposure period, and at the end of the study; the blood was analyzed for levels of serum copper, aspartate aminotransferase, alanine aminotransferase, and GGT activities, and hemoglobin. The average dietary intake of copper in study participants, based on a 24-hour dietary recall, was 1.7 mg Cu/day (0.0266 mg Cu/kg/day using the study-reported average body weight of 64 kg).

Daily doses of copper from drinking water were calculated using reported daily copper intakes (0.04, 1.74, 4.68, and 7.94 mg) and the average of the mean reported body weights (64 kg). Daily doses were 0.0006, 0.0272, 0.0731, 0.124 mg Cu/kg/day for 0, 1, 3 and 5 mg/L, respectively. No significant alterations in levels of serum copper, ceruloplasmin, hemoglobin, or liver enzymes were observed. Twenty-one subjects reported gastrointestinal symptoms, predominantly nausea, at some point during the study period. Nine of those subjects reported 12 episodes diarrhea with or without abdominal pain, and study authors reported no association between copper concentration in water and diarrhea. Eight of these episodes of diarrhea occurred during the 2 weeks of the study independent of copper concentration. Twelve subjects reported abdominal pain, nausea, and/or vomiting; the incidences were 3/60, 1/60, 10/60, and 9/60 in the 0, 0.0272, 0.0731, and 0.124 mg Cu/kg/day groups, respectively (see Table A-2). There was a significant difference between in the incidences at concentrations of ≤1 mg/L (0.0272 mg/kg/day) versus ≥3 mg/L (0.0731 mg/kg/day). No other differences between groups were found.

Table A-1. Incidence of Gastrointestinal Symptoms in Women Exposed to Copper in Drinking Water for 2-Week Periods (Pizarro et al. 1999)

Symptoms	Drinking water doses in mg Cu/kg/day[1]			
	0.0006 (control)	0.03	0.07	0.1
Abdominal pain only	2/60	1/60	3/60	2/60
Vomiting only	0/60	0/60	1/60	2/60
Nausea only	1/60	0/60	6/60	5/60
Total symptoms	3/60	1/60	10/60	9/60

[1]Doses of 0.0006, 0.0272, 0.0731, 0.124 mg Cu/kg/day equates to 0, 1, 3 and 5 mg Cu/L, respectively

Selection of the Point of Departure for the MRL: The $BMDL_{10}$ of 0.05 mg/kg/day for gastrointestinal symptoms of abdominal pain, nausea, and vomiting in females was selected as the basis for the provisional acute-duration oral MRL.

Incidence data for total gastrointestinal symptoms (abdominal pain, vomiting and nausea, see Table A-2) were fit to all dichotomous models in EPA's BMDS (version 3.1.2) using a BMR of 10% extra risk. The data did not require an adjustment for intermittent exposure. Adequate model fit was judged by four criteria: chi-square goodness-of-fit p-values ($p \geq 0.1$), visual inspection of the dose-response curve, BMDL <10 times the lowest non-zero dose, and scaled residual (>-2 and <+2) at the data point (except the control) closest to the predefined BMR. The dichotomous Hill model was the viable, recommended model but was not selected as generally the number of dose groups in the data should be at least one more than the number of parameters in a model. In this case, the dichotomous Hill model uses 4 parameters and the incidence data has 4 dose groups. The Multistage Degree 1 was the only viable alternative and the BMDL from this model was selected as the POD. Table A-3 presents the BMD/BMDL values considered for MRL derivation, and Figure A-1 presents the curve from the chosen model.

While the study NOAEL is lower than the $BMDL_{10}$, the results from BMD modeling were preferred as the data on the full dose-response curve can be used to inform the MRL with benchmark dose modeling and the $BMDL_{10}$ is lower than the LOAEL.

Pizarro et al. (1999) considers other sources of dietary copper intake based on dietary habits reported by participants. The MRL is based off of concentrations in excess of those that a person would normally be exposed from diet. For further information on dietary copper intake, see Section 5.6. Since Pizarro et al. (1999) uses individuals from the general population, the MRL is not protective of individuals with Wilson's disease, which causes excess accumulation of copper in the body. This condition is further is discussed in Section 2.9.

Figure A-1. Fit of Frequentist Multistage Degree 1 Model to Data on Copper for Gastrointestinal Illness in Female Adults, Daily for 2 Weeks (Pizarro et al. 1999)

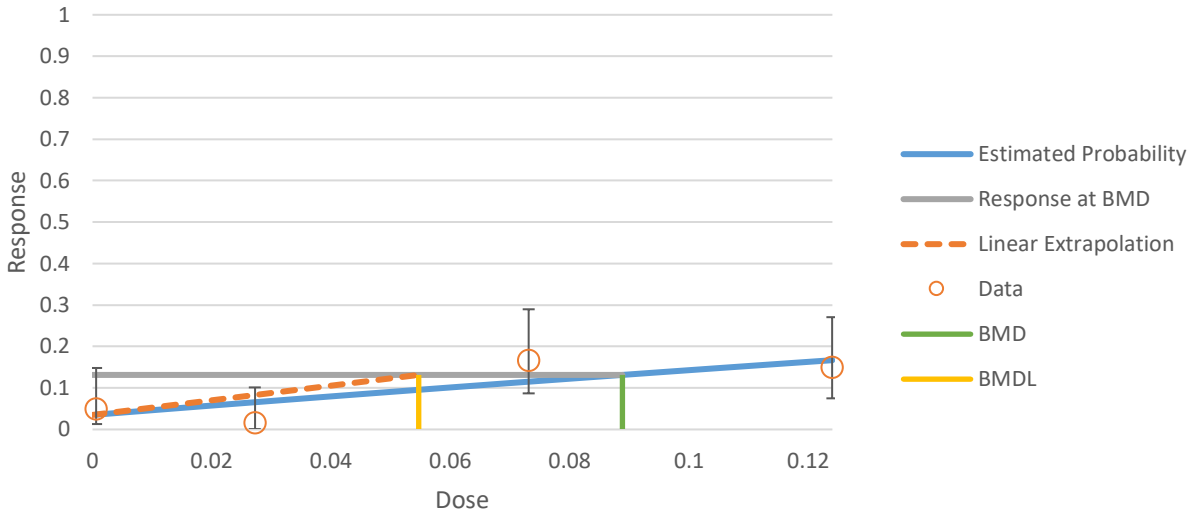


Table A-3. Results from BMD Analysis of Incidence of Gastrointestinal Illness in Women Following Exposure to Copper in Drinking Water Daily for 2 Weeks (Pizarro et al. 1999)

Model	BMD_{10}[a] (mg/kg/day)	$BMDL_{10}$[a] (mg/kg/day)	P-Value[b]	AIC	Scaled residuals[c]	
					Dose below BMD	Dose above BMD
Dichotomous Hill	0.05	0.03	0.29	145.93	0.18	0.72
Gamma[d]	0.09	0.06	0.04	149.56	1.43	0.52
Log-Logistic[e]	0.09	0.05	0.04	149.54	1.4	0.54
Log-Probit[f]	0.09	0.07	0.09	147.62	1.60	0.42
Multistage Degree 3[g]	0.09	0.05	0.03	149.87	1.39	0.55
Multistage Degree 2	0.09	0.05	0.03	149.87	1.39	0.55
Multistage Degree 1	**0.09**	**0.05**	**0.11**	**147.92**	**1.25**	**0.6**
Weibull[d]	0.09	0.06	0.04	149.62	1.42	0.53
Logistic	0.1	0.08	0.08	148.26	-0.58	0.26
Log-Probit	0.09	0.05	0.04	149.16	1.35	0.56
Probit	0.1	0.07	0.09	148.1	1.58	0.36

[a]BMDLs <10 times the lowest non-zero dose and their corresponding BMDs are not included in this table.
[b]Values <0.1 fail to meet conventional χ^2 goodness-of-fit criteria.
[c]Scaled residuals at doses immediately below and above the BMD; also, the largest residual at any dose.
[d]Power restricted to ≥1.
[e]Slope restricted to ≥1.
[f]Betas restricted to ≥0.
[g]Selected model. The Multistage Degree 1 and Dichotomous Hill models provide adequate fit to the data. The dichotomous Hill model had the same number of parameters as the number of dose levels in the data; therefore, the viable, alternative model was selected (Multistage Degree 1).

AIC = Akaike Information Criterion; BMD = benchmark dose (maximum likelihood estimate of the dose associated with the selected benchmark response); $BMDL_{10}$ = 95% lower confidence limit on the BMD (subscripts denote benchmark response: i.e., 10 = dose associated with 10% extra risk); DF = degree of freedom

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty Factor: The $BMDL_{10}$ is divided by a total uncertainty factor of 3:

- 3 for human variability; a partial uncertainty factor was applied because toxicokinetic differences among individuals should not affect the sensitivity of this direct contact effect

$$Provisional\ MRL = \frac{BMDL_{10}}{UF} = \frac{0.05\ mg/kg/day}{3}$$

$$= 0.01667\ mg/kg/day\ (Rounded\ to\ 0.02\ mg/kg/day)$$

Agency Contact (Chemical Managers): Breanna Alman, MPH

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Oral
Duration: Intermediate
MRL: 0.02 mg/kg/day (provisional); adopted acute-duration.

MRL Summary: The provisional acute-duration oral MRL of 0.02 mg Cu/kg/day was adopted as the provisional intermediate-duration oral MRL. The intermediate-duration database was assessed for suitability for MRL derivation. The resulting MRL based on the lowest point-of-departure derived from an intermediate-duration study in humans is higher than the acute-duration oral MRL. Additionally, the critical effect of gastrointestinal symptoms may result in part from a direct contact effect dependent on the amount of copper present at a given time in the stomach. Therefore, the exposure duration may not be driving the gastrointestinal symptoms, and an acute-duration MRL is expected to be applicable to an intermediate-duration exposure scenario.

Selection of the Critical Effect: Gastrointestinal effects have been recorded in humans following intermediate-duration exposure to copper in drinking water. In Araya et al. (2003b), for groups given 0, 2, 4, or 6 mg Cu/day in drinking water, corresponding copper intakes were 0, 2.7, 5.9 and 11.3 mg/day, respectively. To calculate the dose, a reference body weight of 65 kg for all adults was used, resulting in doses of 0, 0.042, 0.091, and 0.17 mg Cu/kg/day. A significant number of subjects (19.7%) exposed to 0.17 mg Cu/kg/day reported at least one gastrointestinal symptom (nausea, vomiting, diarrhea, or abdominal pain). At doses of 0, 0.042 and 0.091 mg Cu/kg/day, symptoms were reported in 11.7, 15.3 and 18.3% of subjects, respectively (Araya et al. 2003b). These gastrointestinal symptoms were also reported in a similar 2-month study among females exposed to ≥0.106 mg Cu/kg/day and males exposed to ≥0.169 mg Cu/kg/day (Araya et al. 2004). A study in infants observed no increase in the reporting of gastrointestinal symptoms following exposure a daily 9-month exposure to doses of 0.123 to 0.158 mg Cu/kg/day (Olivares et al. 1998). Histological effects have been observed in experimental animal studies. At 2.36 mg Cu/kg/day, female mice showed effects including increased thickness of outer muscularis and smooth muscle fiber, widened submucosa, severe atrophy of central lacteal and changes in rectal microbial gut bacteria composition (Cheng et al. 2020). Squamous mucosa hyperplasia of forestomach was observed in higher doses of 44-46 mg Cu/kg/day for 15 days and 33-34 mg Cu/lg/day for 13 weeks (NTP 1993). No changes in hepatic enzyme levels were observed in human adults or infants (Araya et al. 2003b; Olivares et al. 1998). A study in adults men found that exposure to 0.1 mg Cu/kg/day in feed for 18 days resulted in significantly reduced antibody titer against an influenza strain after immunization, compared to controls (Turnlund et al. 2004). Controls had a 47-fold increase in antibody, while the exposure group only had a 14-fold increase. When exposed to 0.02 mg Cu/kg/day for 18 days (same participants), no effects on immune parameters were observed; the immunization testing was not done during this period (Turnlund et al. 2004).

Animal studies have reported altered hepatic enzymes levels in rats and mice (Epstein et al. 1982; Hashish and Elgaml 2016; Kumar et al. 2015; Kumar and Sharma 1987; NTP 1993; Sakhaee et al. 2012, 2014; Suttle and Mills 1966a; Tian et al. 2019). At 1.59 mg Cu/kg/day, female albino rats exposed for 30 days exhibited acute swelling of hepatocytes, including coagulative necrosis with presence of faintly stained nuclei, and a thickened wall of the bile duct among (Hashish and Elgaml 2016). The severity of hepatic histological effects increased with dose in mice exposed to concentrations ranging from 3.19 to

5.6 mg Cu/kg/day. (Kvietkauskatte et al. 2004; Wu et al. 2020). At 16 mg Cu/kg/day, male rats exhibited chronic active inflammation with focal necrosis and altered liver cholesterol (NTP 1993; Shen et al. 2005). At higher doses ranging from 39.81to 199 mg/kg/day, rats displayed altered liver function including hepatocellular necrosis and degeneration, hepatic lesions with tubular necrosis, reduced liver weight, hemorrhage, and indications of oxidative stress (Abe et al. 2008; Kumar et al. 2016a, 2016b; Rana and Kumar 1980; Sakhaee et al. 2012; Seven et al. 2018; Tian et al. 2019).

Neurological, reproductive, and renal effects have been reported in multiple animal studies. Impaired motor function, reduced cognitive function, neurochemical changes, memory impairment, neuronal loss, depleted myelin, and increased hippocampal mitochondria lipid peroxidation have been seen at doses ≥3.98 mg Cu/kg/day in mice, rats, and rabbits (Behzadfar et al. 2017; Kalita et al. 2020; Kumar et al. 2015, 2019; Kumar et al. 2016a, 2016b). Reproductive effects of reduced fertility in male rodents, altered sperm parameters and malformation, decreased testicular weight, decreased hormone activity and depletion of seminiferous epithelium and tubules was seen at >39.81 mg Cu/kg/day (Arafa et al. 2019; Babaei and Abshenas 2013; Kheirandish et al. 2014; Liu et al. 2016; Sakhaee et al. 2012, 2014; Sakhaee et al. 2016a, 2016b). Female mice exposed to 39.81 mg Cu/kg/day for 35 days, showed significant decreases in number of ovarian follicles and ovarian cell damage (Babaei et al. 2012). Renal effects of altered enzymes levels, altered kidney function, lesions, tubular cell necrosis and degeneration, kidney hemorrhage, and necrosis of epithelial lining of the cortical tubules have been seen at doses ranging from 1.6 to 199 mg Cu/kg/day (Hashish and Elgaml 2016; Kumar et al. 2015; Kumar et al. 2016a, 2016b; Kumar and Sharma 1987; NTP 1993; Rana and Kumar 1980; Sakhaee et al. 2012; Seven et al. 2018). Other effects recorded in animals at various doses ranging from 5.6 – 199 mg/kg/day include hematological, cardiovascular, respiratory, immunological, and endocrine (Arafa et al. 2019; Kumar et al. 2015; Kumar and Sharma 1987; Kvietkauskatte et al. 2004; NTP 1993; Rana and Kumar 1980; Seven et al. 2018; Suttle and Mills 1966a; Tian et al. 2019). The values considered for MRL derivation in the intermediate-duration database are presented in Table A-4.

Table A-2. Summary of Lowest LOAEL Values for Health Effects Following Intermediate-Duration Oral Exposure to Copper

Species (sex)	Frequency/ Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Body weight effects					
Pigs (NS)	88 days Ad libitum	1.7	2.3	17% reduction in body weight gain	Kline et al. 1971
Musculoskeletal effects					
Rat (M)	16 weeks Daily		4	Impaired muscle strength	Kumar et al. 2019
Gastrointestinal effects					
Human (Adults)	2 months Daily	0.091	0.17	Incidence of gastrointestinal symptoms	Araya et al. 2003b
Human (Infants)	9 months	0.319		Gastrointestinal symptoms	Olivares et al. 1998

Table A-2. Summary of Lowest LOAEL Values for Health Effects Following Intermediate-Duration Oral Exposure to Copper

Species (sex)	Frequency/ Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Mice (F)	90 days Daily		2.4 (SLOAEL)	Increased histological lesions and cecum and rectum including increased thickness of outer muscularis, widened submucosa, and severe atrophy of central lacteal; changes in rectal microbial gut bacteria composition	Cheng et al. 2020
Rabbit (F)	GD 7-28 Daily		6	Diarrhea in 5/22 rabbits	Munley 2003a
Rabbit (F)	GD 7-28 Daily		7.5	Diarrhea in 1/8 rabbits	Munley 2003a
Hepatic effects					
Human (Adults)	2 months Daily	0.17		Enzyme levels	Araya et al. 2003b
Human (Infants)	9 months	0.319		Liver function test	Olivares et al. 1998
Rats (F)	30 days Daily		1.59	Acute cell swelling of hepatocytes and karyolysis of nuclei, mild hyperplasia of portal area lining epithelium of bile ducts	Hashish and Elgaml 2016
Mice (NS)	42 days Daily		4 (SLOAEL)	Increased incidence of granular and vacuolar degeneration in hepatocytes and increased rate of hepatic apoptosis	Wu et al. 2020
Mice (M)	19 weeks Ad libitum		5.6	13.6% decrease in total liver protein	Kvietkauskaite et al. 2004
Renal effects					
Rats (F)	30 days Daily		1.6	Degeneration of renal tissues and degeneration in lining epithelium of some renal tubules; decreased total protein by >32%, increased urea and creatinine by 50% and >65%	Hashish and Elgaml 2016
Immunological effects					
Human (Adults)	18 days Daily	0.02			Turnlund et al. 2004
Human (Adults)	18 days Daily		0.1	Significantly reduced antibody titer against influenza strain compared to controls	Turnlund et al. 2004

Table A-2. Summary of Lowest LOAEL Values for Health Effects Following Intermediate-Duration Oral Exposure to Copper

Species (sex)	Frequency/ Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Mice (M)	19 weeks Ad libitum		5.6	Changed percent of natural killer (CD4*CD8) and suppressor (CD8*CD4) cells and altered immunoregulatory index	Kvietkauskaite et al. 2004
Neurological effects					
Rat (M)	16 weeks Daily		4 (SLOAEL)	Decreased locomotor activity and neuromuscular coordination, reduced catalase, and SOD activity in brain tissues, decreased passive avoidance response, less exploration time	Kumar et al. 2019

F=females; GD = gestation day; LOAEL = lowest-observed-adverse-effect level; M=males; NOAEL = no-observed-adverse-effect level; NS = not specified; SOD = superoxide dismutase

The incidence of total gastrointestinal symptoms of nausea, vomiting, diarrhea, and abdominal pain reported in adults was considered as the basis for an intermediate-duration MRL for copper. Araya et al. (2003b) identified the lowest NOAEL and corresponding LOAEL for adverse health outcomes. For groups given 0, 2, 4, or 6 mg Cu/day in drinking water, corresponding daily copper intakes provided by study authors were 0, 2.7, 5.9 and 11.3 mg/day, respectively. To calculate the dose, a reference body weight of 65 kg for all adults was used, resulting in doses of 0, 0.042, 0.091, and 0.17 mg Cu/kg/day. The Araya et al. (2003b) study examined a Chilean population similar to that examined by Pizarro et al. (1999), which determined a body weight of 64 kg, and both studies were conducted during similar time periods. Additionally, the EPA had recommended using 65 kg as a reference body weight at the time the study was published, until that recommendation was increased to 80 kg in 2011.

BMD modeling was applied to the incidence data to estimate the potential point of departure (POD) for gastrointestinal symptoms observed by Araya et al. (2003b). The data were fit to all available dichotomous models in EPA's BMDS (version 3.2.1) using the extra risk option. Adequate model fit was judged by four criteria: chi-square goodness-of-fit p-values ($p \geq 0.1$), visual inspection of the dose-response curve, BMDL <10 times the lowest non-zero dose, and scaled residual (>-2 and <+2) at the data point (except the control) closest to the predefined BMR. Among the recommended, viable models providing adequate fit to the data, the BMDL from the model with the lowest AIC was selected as the POD. The results of the BMD modeling for incidence of gastrointestinal symptoms in adults are presented in Table A-5.

Table A-5. Results from BMD Analysis of Incidence of Gastrointestinal Illness in Adults Following Exposure to Copper in Drinking Water Daily for 2 Months (Araya et al. 2003b)

Model	BMD_{10}[a] (mg/kg/day)	$BMDL_{10}$[a] (mg/kg/day)	P-Value[b]	AIC	Scaled residuals[c]	
					Dose below BMD	Dose above BMD
Dichotomous Hill	65535	0	NA	1210.48	-9999	-0.00014
Gamma[d]	0.18	0.12	0.61	1207.24	-0.49	-0.48
Log-Logistic[e]	**0.18**	**0.11**	**0.64**	**1207.15**	**-0.49**	**-0.43**
Log-Probit[f]	0.18	0.12	0.62	1207.24	-0.49	-0.48
Multistage Degree 3[g]	0.18	0.12	0.61	1207.24	-0.49	-0.48
Multistage Degree 2	0.18	0.12	0.61	1207.24	-0.49	-0.48
Multistage Degree 1	0.18	0.12	0.61	1207.24	-0.49	-0.48
Weibull[d]	0.19	0.13	0.48	1207.73	-0.47	-0.7
Logistic	0.19	0.00042	0.72	1208.39	-0.13	0.01
Log-Probit	0.18	0.13	0.5	1207.65	-0.48	-0.66
Probit	65535	0	NA	1210.48	-9999	-0.00014

[a]BMDLs <10 times the lowest non-zero dose and their corresponding BMDs are not included in this table.
[b]Values <0.1 fail to meet conventional χ^2 goodness-of-fit criteria.
[c]Scaled residuals at doses immediately below and above the BMD; also, the largest residual at any dose.
[d]Power restricted to ≥1.
[e]Slope restricted to ≥1.
[f]Betas restricted to ≥0.
[g]Selected model. Among viable models, the model with the lowest AIC was selected (Log-Logistic).

AIC = Akaike Information Criterion; BMD = benchmark dose (maximum likelihood estimate of the dose associated with the selected benchmark response); $BMDL_{10}$ = 95% lower confidence limit on the BMD (subscripts denote benchmark response: i.e., 10 = dose associated with 10% extra risk); DF = degree of freedom

The BMDL of 0.11 mg Cu/kg/day from the Log-Logistic model was selected as the POD. The BMDL was divided by an uncertainty factor of 3 for human variability (a partial uncertainty factor was applied because toxicokinetic differences among individuals should not affect the sensitivity of this direct contact effect) resulting in an MRL of 0.03667 mg Cu/kg/day (rounded to 0.04 mg Cu/kg/day). However, this MRL is higher than the acute-duration oral MRL, and ATSDR adopted the acute-duration oral MRL of 0.02 mg Cu/kg/day for intermediate-duration exposure. As noted previously, the critical effect of gastrointestinal symptoms may result from a direct contact effect dependent on the amount of copper present at a given time in the gastrointestinal system. Therefore, the exposure duration may not be driving the gastrointestinal symptoms, and an acute-duration MRL is expected to be applicable to an intermediate-duration exposure scenario.

Agency Contact (Chemical Managers): Breanna Alman, MPH

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** Copper and compounds
CAS Numbers: 7440-50-8
Date: April 2022
Profile Status: Draft for Public Comment
Route: Oral
Duration: Chronic

MRL Summary: There are insufficient data for derivation of chronic-duration oral MRL due to lack of toxicity studies. No chronic human studies report sufficient dose information and animal studies are limited to three studies.

Rationale for Not Deriving an MRL: No human studies provided sufficient dose information to examine the chronic-duration oral toxicity of copper. Animal studies were limited, as one study in mice exposed for 850 days only reported decreased lifespan and found no effects on body weight (Massie and Aiello 1984). A study in monkeys conducted two experiments exposing them to copper in feed daily for 3 years and identified LOAELs of 0.77 and 1.05 mg/kg/day for hepatic effects (Araya et al. 2012). However, these studies tested a small number of animals, 4 monkeys (2 males and 2 females) per dose group, therefore, are insufficient in meeting the level of data quality required to inform an MRL.

Agency Contact (Chemical Managers): Breanna Alman, MPH

APPENDIX B. LITERATURE SEARCH FRAMEWORK FOR COPPER

The objective of the toxicological profile is to evaluate the potential for human exposure and the potential health hazards associated with inhalation, oral, or dermal/ocular exposure to copper.

B.1 LITERATURE SEARCH AND SCREEN

A literature search and screen was conducted to identify studies examining health effects, toxicokinetics, mechanisms of action, susceptible populations, biomarkers, chemical interactions, physical and chemical properties, production, use, environmental fate, environmental releases, and environmental and biological monitoring data for copper. ATSDR primarily focused on peer-reviewed articles without publication date or language restrictions. Non-peer-reviewed studies that were considered relevant to the assessment of the health effects of copper have undergone peer review by at least three ATSDR-selected experts who have been screened for conflict of interest. The inclusion criteria used to identify relevant studies examining the health effects of copper are presented in Table B-1.

Table B-1. Inclusion Criteria for the Literature Search and Screen

Health Effects
Species
Human
Laboratory mammals Drosophila (for genotoxicity studies) *In vitro* assay (for genotoxicity and for supporting data for other endpoints)
Route of exposure
Inhalation
Oral
Dermal (or ocular)
Parenteral (these studies will be considered supporting data)
Health outcome
Death
Systemic effects
Respiratory effects
Cardiovascular effects
Gastrointestinal effects
Hematological effects
Musculoskeletal effects
Hepatic effects
Renal effects
Endocrine effects
Dermal effects
Ocular effects
Body weight effects
Metabolic effects
Other systemic effects
Immunological effects

Table B-1. Inclusion Criteria for the Literature Search and Screen

	Neurological effects
	Reproductive effects
	Developmental effects
	Genotoxicity
	Cancer

B.1.1 Literature Search

The current literature search was intended to update the existing toxicological profile for copper (ATSDR 2004), thus, the literature search was restricted to studies published between 2002 to 2020.

The following main databases were searched in April and May 2020:

- PubMed
- MEDLINE
- Science Direct
- Scopus

The search strategy used the chemical names, Chemical Abstracts Service (CAS) numbers, synonyms, Medical Subject Headings (MeSH) headings, and keywords for copper and copper sulfate. The query strings used for the literature search are presented in Table B-2.

Additional databases were searched in the creation of various tables and figures, such as the TRI Explorer, the Substance Priority List (SPL) resource page, and other items as needed. Regulations applicable to copper were identified by searching international and U.S. agency websites and documents. The EPA's Office of Pesticide Programs Chemical Search and ChemView were searched for studies relevant to copper toxicity in mammals.

Review articles were identified and used for the purpose of providing background information and identifying additional references. ATSDR also identified reports from the grey literature, which included unpublished research reports, technical reports from government agencies, conference proceedings and abstracts, and theses and dissertations.

Table B-2. Database Query Strings

Database Search date	Query string
MEDLINE 04/24/20	((MH "Copper") OR AB ("Cuprum" OR "Cobre" OR "Cuivre" OR "Cuprum metallicum" OR "Rame" OR "Gold Bronze" OR "Bronze powder" OR "CDX" OR "CI Pigment Metal 2" OR "Cu" OR "CE 7" OR "Kupfer" OR "Cutox 6010" OR "Cutox 6030" OR "C100" OR "E115" OR "1721 Gold" OR "CU M3" OR "OFHC Cu" OR "Tatum-T" OR "Paragard T 38a") OR RN (7440-50-8)) AND ((MH Death OR "Body Weight" OR "respiratory system" OR "cardiovascular diseases" OR "gastrointestinal diseases" OR "hematologic diseases" OR "musculoskeletal diseases" OR "hepatic infraction" OR "renal insufficiency" OR dermatology OR "endocrine system" OR neurology OR "reproductive health" OR "developmental

Table B-2. Database Query Strings

Database Search date	Query string
	disabilities" OR "psychology, developmental" OR Neoplasms OR "DNA Damage") OR AB (Death OR "Body weight" OR respiratory OR cardiovascular OR gastrointestinal OR hematological OR musculoskeletal OR hepatic OR Renal OR dermal OR ocular OR endocrine OR immunological OR neurological OR reproductive OR developmental OR Cancer OR genotoxicity OR noncancer OR "health effects")) Limiters: Date of Publication: 20020101-20200424
04/27/20	((MH "Copper") OR AB ("Cuprum" OR "Cobre" OR "Cuivre" OR "Cuprum metallicum" OR "Rame" OR "Gold Bronze" OR "Bronze powder" OR "CDX" OR "CI Pigment Metal 2" OR "Cu" OR "CE 7" OR "Kupfer" OR "Cutox 6010" OR "Cutox 6030" OR "C100" OR "E115" OR "1721 Gold" OR "CU M3" OR "OFHC Cu" OR "Tatum-T" OR "Paragard T 38a") OR RN (7440-50-8)) AND ((MH Death OR "Body Weight" OR "respiratory system" OR "cardiovascular diseases" OR "gastrointestinal diseases" OR "hematologic diseases" OR "musculoskeletal diseases" OR "hepatic infraction" OR "renal insufficiency" OR dermatology OR "endocrine system" OR neurology OR "reproductive health" OR "developmental disabilities" OR "psychology, developmental" OR Neoplasms OR "DNA Damage") OR (Death OR mortality OR lethal OR "lethal dose" OR "Lethal concentration" OR fatal OR fatality OR necrosis) OR ("body weight" OR "weight loss" OR "weight gain" OR "weight change") OR (Respiratory OR "respiratory tract" OR "respiratory organ" OR "respiratory System" OR "respiratory volume" OR "respiratory function" OR "respiratory effect" OR "respiratory organ" OR "respiratory toxicity" OR "pulmonary edema" OR "pulmonary effect" OR "pulmonary system" OR "pulmonary function" OR "pulmonary organ" OR "pulmonary toxicity" OR airway OR Trachea OR tracheobronchial OR "lung function" OR "lung change*" OR "Lung congestion" OR nose OR nasal OR nasopharyngeal OR larynx OR pharynx OR Bronchial or bronchi OR bronchioles OR bronchitis OR hemothorax OR alveolar OR alveoli OR irritation or irritant OR "vineyard sprayer's lung" OR cilia OR mucocilliary) OR (CVD OR Cardio OR Vascular OR "Cardiovascular system" OR "cardiovascular function" OR "cardiovascular effect" OR "cardiovascular organ" OR "cardiovascular toxicity" OR "Circulatory system" OR "circulatory function" OR "circulatory effect" OR "circulatory organ" OR "circulatory toxicity" OR "Cardiac arrest" OR "cardiac palpitation" OR "cardiac arrhythmia" OR "cardiac edema" OR "Heart rate" OR "heart failure" OR "heart attack" OR "heart muscle" OR "heart beat" OR "Blood pressure" OR "blood flow" OR "Myocardial infarction" OR "Chest pain" OR Artery OR arteries OR veins OR venules) OR (Gastrointestinal OR "Gastrointestinal system" OR "gastrointestinal function" OR "gastrointestinal effect" OR "gastrointestinal organ" OR "Digestive system" OR "digestive function" OR "digestive effect" OR "digestive organ" OR "Intestinal system" OR "intestinal function" OR "intestinal microbiota" OR "intestinal effect" OR "intestinal organ" OR "GI tract" OR "GI disorder" OR Abdominal OR Esophagus OR Stomach OR Intestine OR Pancreas OR Pancreatic OR Diarrhea OR Nausea OR Vomit OR Ulcer OR Constipation OR Emesis OR "Gut microbes" OR "gut flora" OR "gut microflora OR anorexia) OR

Table B-2. Database Query Strings

Database Search date	Query string
	(Hematological OR Hematology OR "hematology system" OR "hematology function" OR "hematology effect" OR "hematology toxicity" OR Hemato OR Haemato OR Blood OR "blood chemistry" OR "blood disease" OR Anemia OR Cyanosis OR Erythrocytopenia OR Leukopenia OR Thrombocytopenia OR Hemoglobin OR Erythrocyte OR Hematocrit OR "Bone marrow" OR "bone marrow decrease" OR "bone marrow hyper" OR "bone marrow hypoplasia" OR Reticulocyte OR Methemoglobin OR "Red blood cell") OR (Musculoskeletal OR Skeletal OR "skeletal system" OR "skeletal function" OR "skeletal effect" OR Muscle OR "muscle loss" OR "muscle strength" OR "muscle structure" OR Muscular OR "muscular rigidity" OR "muscular atrophy" OR "muscular structure" OR "muscular system" OR Arthritis OR "Altered bone" OR "joint pain" OR "joint ache" OR "limb pain" OR "limb ache") OR (Hepatic OR "hepatic system" OR "hepatic function" OR "hepatic effect" OR "hepatic organ" OR "hepatic response" OR "hepatic necrosis" OR "hepatic biochemical changes" OR "hepatic toxicity" OR "liver system" OR "liver function" OR "liver effect" OR "liver organ" OR "Liver enzyme" OR "liver weight" OR "liver congestion" OR "liver changes" OR "liver biochemical changes" OR "liver toxicity" OR "gallbladder system" OR "gallbladder function" OR "gallbladder organ" OR "gallbladder effect" OR "gallbladder toxicity" OR Hepatocytes OR Cirrhosis OR Jaundice OR "Hepatocellular degeneration" OR "hepatocellular hypertrophy" OR Hepatomegaly) OR ("Renal system" OR "renal function" OR "renal effect" OR "renal organ" OR "renal tubular" OR "renal toxicity" OR "kidney system" OR "kidney function" OR "Kidney effect" OR "kidney toxicity" OR "urinary system" OR "urinary function" OR "urinary effect" OR "Urinary toxicity" OR "bladder system" OR "bladder effect" OR "bladder function" OR "bladder toxicity" OR "Urine volume" OR "blood urea nitrogen" OR BUN OR nephropathy) OR ("Dermal system" OR "dermal function" OR "dermal effect" OR "dermal irritation" OR "dermal toxicity" OR "skin rash" OR "skin itch" OR "skin irritation" OR "skin redness" OR "skin effect" OR "skin Necrosis" OR "skin acanthosis" OR dermatitis OR edema OR ulceration OR acne) OR ("Ocular system" OR "ocular function" OR "ocular effect" OR "ocular irritation" OR "ocular toxicity" OR "eye function" OR "eye effect" OR "eye irritation" OR "eye drainage" OR "eye tearing" OR Blindness OR Myopia OR Cataracts) OR ("Endocrine system" OR "endocrine function" OR "endocrine effect" OR "endocrine gland" OR "endocrine toxicity" OR "hormone changes" OR "hormone excess" OR "hormone deficiency" OR "hormone gland" OR "hormone secretion" OR "hormone toxicity" OR Pancreas OR "Pancreatic system" OR "pancreatic function" OR "pancreatic effect" OR "pancreatic toxicity" OR "sella turcica" OR thyroid OR adrenal OR pituitary) OR ("Immunological system" OR "immunological function" OR "immunological effect" OR "immunological toxicity" OR Immune OR "immunologic system" OR "immunologic function" OR "immunologic effect" OR "immunologic response" OR "immunologic tissue" OR "immunologic toxicity" OR "Lymphoreticular changes" OR "Lymphoreticular effects" OR "Lymphoreticular function" OR "Lymphoreticular tissue" OR "Lymph node" OR Spleen OR Thymus OR Macrophage OR "white blood cell")

Table B-2. Database Query Strings

Database Search date	Query string
	OR (Neurological OR neurologic OR "Neuro system" OR "neuro function" OR "neuro effect" OR "neuro toxicity" OR "Nervous system" OR "Brain function" OR "brain effect" OR "brain toxicity" OR Neurotoxicant OR Neurochemistry OR Neurophysiology OR Neuropathology OR "Motor activity" OR "motor change" OR (Changes AND (behavior OR behavioral OR sensory OR cognitive)) OR Vertigo OR Drowsiness OR Headache OR Ataxia) OR (Reproductive OR "reproduction system" OR "reproduction function" OR "reproduction effect" OR "reproduction toxicity" OR Fertility OR "Maternal toxicity") OR (Developmental OR "development system" OR "developmental effect" OR "developmental toxicity" OR "developmental function" OR "developmental delay" OR "developmental abnormality" OR "developmental defect" OR (Offspring AND ("child effect" OR toxicity)) OR "In utero") OR ("altered food consumption" OR "altered water consumption" OR "Metabolic effect" OR "metabolic toxicity" OR fever) OR (Cancer OR Cancerous OR Tumor OR Carcinoma OR Carcinogen OR Mutation) OR (Genotoxicity OR "Genotoxic in vivo" OR "genotoxic in vitro" OR Mutagenicity OR Mutagenic) OR ("Mechanism of action" OR "mechanism of absorption" OR "mechanism of distribution" OR "mechanism of excretion" OR "Mechanism of metabolism" OR "Mechanism of toxic effect") OR (Human or Animal OR "Occupational workers" OR "Adverse effects" OR Poisoning OR Morbidity OR Inflammation OR antagonist OR inhibitor OR metabolism OR "environmental exposure" OR toxicokinetics OR pharmacokinetics OR "gene expression" OR epidemiology OR epidemiological OR "Population health" OR Inhalation OR "oral intake" OR "oral feed" OR "oral ingestion") Limiters: Date of Publication: 20020101-20200427
04/09/20	(MH "Copper Sulfate" OR "Blue Vitriol" OR "Cupric Sulfate" OR "Sulfate, Copper" OR "Sulfate, Cupric" OR "Vitriol, Blue") OR "Copper sulphate" OR "Cupric sulfate" OR "Cupric sulfate anhydrous" OR "Copper monosulfate" OR "Cupric sulphate" OR "copper (II) sulfate" OR "hylinec" OR "trinagle" OR "delcup" OR "incracide E 51" OR "incracide 10A" OR "blue stone" OR "Roman vitriol" OR "Salzburg vitriol" OR "blue copperas" OR "sulfuric acid copper (2+) salt (1:1)" OR "sulfate de cuivre" OR RN (7758-98-7) Limiters: Date of Publication: 20020101-20200410
PubMed 04/24/20	(("copper"[MeSH Terms]) OR (TI/AB ("Cuprum" OR "Cobre" OR "Cuivre" OR "Cuprum metallicum" OR "Rame" OR "Gold Bronze" OR "Bronze powder" OR "CDX" OR "CI Pigment Metal 2" OR "Cu" OR "CE 7" OR "Kupfer" OR "Cutox 6010" OR "Cutox 6030" OR "C100" OR "E115" OR "1721 Gold" OR "CU M3" OR "OFHC Cu" OR "Tatum-T" OR "Paragard T 38a") OR "7440-50-8"[EC/RN Number]) AND (MeSH Terms ("Death"[MeSH Terms] OR "Body Weight"[MeSH Terms] OR "respiratory system"[MeSH Terms] OR "cardiovascular diseases"[MeSH Terms] OR "gastrointestinal diseases" [MeSH Terms] OR "hematologic diseases" [MeSH Terms] OR "musculoskeletal diseases" [MeSH Terms] OR "hepatic infraction" [MeSH Terms] OR "renal insufficiency" [MeSH Terms] OR dermatology [MeSH Terms] OR "endocrine system" [MeSH Terms] OR neurology[MeSH Terms] OR "reproductive health" [MeSH Terms] OR "developmental disabilities" [MeSH Terms] OR "psychology, developmental" [MeSH Terms] OR Neoplasms[MeSH Terms] OR "DNA Damage" [MeSH Terms]) OR

Table B-2. Database Query Strings

Database Search date	Query string
	TI/AB (Death OR “Body weight” OR respiratory OR cardiovascular OR gastrointestinal OR hematological OR musculoskeletal OR hepatic OR Renal OR dermal OR ocular OR endocrine OR immunological OR neurological OR reproductive OR developmental OR Cancer OR genotoxicity OR noncancer OR “health effects”)) Limited 2002 – present
04/27/20	((“copper”[MeSH Terms]) OR (TI/AB (“Cuprum” OR “Cobre” OR “Cuivre” OR “Cuprum metallicum” OR “Rame” OR “Gold Bronze” OR “Bronze powder” OR “CDX” OR “CI Pigment Metal 2” OR “Cu” OR “CE 7” OR “Kupfer” OR “Cutox 6010” OR “Cutox 6030” OR “C100” OR “E115” OR “1721 Gold” OR “CU M3” OR “OFHC Cu” OR “Tatum-T” OR “Paragard T 38a”) OR “7440-50-8”[EC/RN Number]) AND (MeSH Terms (“Death”[MeSH Terms] OR “Body Weight”[MeSH Terms] OR “respiratory system”[MeSH Terms] OR “cardiovascular diseases”[MeSH Terms] OR “gastrointestinal diseases” [MeSH Terms] OR “hematologic diseases” [MeSH Terms] OR “musculoskeletal diseases” [MeSH Terms] OR “hepatic infraction” [MeSH Terms] OR “renal insufficiency” [MeSH Terms] OR dermatology [MeSH Terms] OR “endocrine system” [MeSH Terms] OR neurology[MeSH Terms] OR “reproductive health” [MeSH Terms] OR “developmental disabilities” [MeSH Terms] OR “psychology, developmental” [MeSH Terms] OR Neoplasms[MeSH Terms] OR “DNA Damage” [MeSH Terms]) OR Death OR mortality OR lethal OR “lethal dose” OR “Lethal concentration” OR fatal OR fatality OR necrosis OR “body weight” OR “weight loss” OR “weight gain” OR “weight change” OR Respiratory OR “respiratory tract” OR “respiratory organ” OR “respiratory System” OR “respiratory volume” OR “respiratory function” OR “respiratory effect” OR “respiratory organ” OR “respiratory toxicity” OR “pulmonary edema” OR “pulmonary effect” OR “pulmonary system” OR “pulmonary function” OR “pulmonary organ” OR “pulmonary toxicity” OR airway OR Trachea OR tracheobronchial OR “lung function” OR “lung change*” OR “Lung congestion” OR nose OR nasal OR nasopharyngeal OR larynx OR pharynx OR Bronchial or bronchi OR bronchioles OR bronchitis OR hemothorax OR alveolar OR alveoli OR irritation or irritant OR “vineyard sprayer’s lung” OR cilia OR mucocilliary OR CVD OR Cardio OR Vascular OR “Cardiovascular system” OR “cardiovascular function” OR “cardiovascular effect” OR “cardiovascular organ” OR “cardiovascular toxicity” OR “Circulatory system” OR “circulatory function” OR “circulatory effect” OR “circulatory organ” OR “circulatory toxicity” OR “Cardiac arrest” OR “cardiac palpitation” OR “cardiac arrhythmia” OR “cardiac edema” OR “Heart rate” OR “heart failure” OR “heart attack” OR “heart muscle” OR “heart beat” OR “Blood pressure” OR “blood flow” OR “Myocardial infarction” OR “Chest pain” OR Artery OR arteries OR veins OR venules OR Gastrointestinal OR “Gastrointestinal system” OR “gastrointestinal function” OR “gastrointestinal effect” OR “gastrointestinal organ” OR “Digestive system” OR “digestive function” OR “digestive effect” OR “digestive organ” OR “Intestinal system” OR “intestinal function” OR “intestinal microbiota” OR “intestinal effect” OR “intestinal organ” OR “GI tract” OR “GI disorder” OR Abdominal OR Esophagus OR Stomach OR Intestine OR Pancreas OR Pancreatic OR Diarrhea OR Nausea OR Vomit OR Ulcer OR Constipation OR Emesis OR “Gut microbes” OR “gut flora” OR “gut microflora OR anorexia OR Hematological OR Hematology OR “hematology system” OR “hematology function” OR “hematology effect” OR “hematology toxicity” OR Hemato OR Haemato OR Blood OR “blood chemistry” OR “blood disease” OR Anemia OR Cyanosis OR Erythrocytopenia OR Leukopenia OR Thrombocytopenia OR Hemoglobin OR Erythrocyte OR Hematocrit OR “Bone marrow”

Table B-2. Database Query Strings

Database Search date	Query string
	OR “bone marrow decrease” OR “bone marrow hyper” OR “bone marrow hypoplasia” OR Reticulocyte OR Methemoglobin OR “Red blood cell” OR musculoskeletal OR Skeletal OR “skeletal system” OR “skeletal function” OR “skeletal effect” OR Muscle OR “muscle loss” OR “muscle strength” OR “muscle structure” OR Muscular OR “muscular rigidity” OR “muscular atrophy” OR “muscular structure” OR “muscular system” OR Arthritis OR “Altered bone” OR “joint pain” OR “joint ache” OR “limb pain” OR “limb ache” OR Hepatic OR “hepatic system” OR “hepatic function” OR “hepatic effect” OR “hepatic organ” OR “hepatic response” OR “hepatic necrosis” OR “hepatic biochemical changes” OR “hepatic toxicity” OR “liver system” OR “liver function” OR “liver effect” OR “liver organ” OR “Liver enzyme” OR “liver weight” OR “liver congestion” OR “liver changes” OR “liver biochemical changes” OR “liver toxicity” OR “gallbladder system” OR “gallbladder function” OR “gallbladder organ” OR “gallbladder effect” OR “gallbladder toxicity” OR Hepatocytes OR Cirrhosis OR Jaundice OR “Hepatocellular degeneration” OR “hepatocellular hypertrophy” OR Hepatomegaly OR “Renal system” OR “renal function” OR “renal effect” OR “renal organ” OR “renal tubular” OR “renal toxicity” OR “kidney system” OR “kidney function” OR “Kidney effect” OR “kidney toxicity” OR “urinary system” OR “urinary function” OR “urinary effect” OR “Urinary toxicity” OR “bladder system” OR “bladder effect” OR “bladder function” OR “bladder toxicity” OR “Urine volume” OR “blood urea nitrogen” OR BUN OR nephropathy OR “Dermal system” OR “dermal function” OR “dermal effect” OR “dermal irritation” OR “dermal toxicity” OR “skin rash” OR “skin itch” OR “skin irritation” OR “skin redness” OR “skin effect” OR “skin Necrosis” OR “skin acanthosis” OR dermatitis OR edema OR ulceration OR acne OR “Ocular system” OR “ocular function” OR “ocular effect” OR “ocular irritation” OR “ocular toxicity” OR “eye function” OR “eye effect” OR “eye irritation” OR “eye drainage” OR “eye tearing” OR Blindness OR Myopia OR Cataracts OR “Endocrine system” OR “endocrine function” OR “endocrine effect” OR “endocrine gland” OR “endocrine toxicity” OR “hormone changes” OR “hormone excess” OR “hormone deficiency” OR “hormone gland” OR “hormone secretion” OR “hormone toxicity” OR Pancreas OR “Pancreatic system” OR “pancreatic function” OR “pancreatic effect” OR “pancreatic toxicity” OR “sella turcica” OR thyroid OR adrenal OR pituitary OR “Immunological system” OR “immunological function” OR “immunological effect” OR “immunological toxicity” OR Immune OR “immunologic system” OR “immunologic function” OR “immunologic effect” OR “immunologic response” OR “immunologic tissue” OR “immunologic toxicity” OR “Lymphoreticular changes” OR “Lymphoreticular effects” OR “Lymphoreticular function” OR “Lymphoreticular tissue” OR “Lymph node” OR Spleen OR Thymus OR Macrophage OR “white blood cell” OR Neurological OR neurologic OR “Neuro system” OR “neuro function” OR “neuro effect” OR “neuro toxicity” OR “Nervous system” OR “Brain function” OR “brain effect” OR “brain toxicity” OR Neurotoxicant OR Neurochemistry OR Neurophysiology OR Neuropathology OR “Motor activity” OR “motor change” OR behavior change OR behavioral change OR sensory change OR cognitive change OR Vertigo OR Drowsiness OR Headache OR Ataxia OR Reproductive OR “reproduction system” OR “reproduction function” OR “reproduction effect” OR “reproduction toxicity” OR Fertility OR “Maternal toxicity” OR Developmental OR “development system” OR “developmental effect” OR “developmental toxicity” OR “developmental function” OR “developmental delay” OR “developmental abnormality” OR “developmental defect” OR “offspring toxicity” OR “In utero” OR “altered food consumption” OR “altered water consumption” OR “Metabolic effect” OR “metabolic toxicity” OR fever OR Cancer OR Cancerous OR Tumor OR Carcinoma OR Carcinogen OR Mutation OR Genotoxicity OR “Genotoxic in vivo” OR “genotoxic in vitro” OR Mutagenicity OR Mutagenic OR “Mechanism of action” OR “mechanism of absorption” OR “mechanism of distribution”

Table B-2. Database Query Strings

Database Search date	Query string
	OR “mechanism of excretion” OR “Mechanism of metabolism” OR “Mechanism of toxic effect” OR Human or Animal OR “Occupational workers” OR “Adverse effects” OR Poisoning OR Morbidity OR Inflammation OR antagonist OR inhibitor OR metabolism OR “environmental exposure” OR toxicokinetics OR pharmacokinetics OR “gene expression” OR epidemiology OR epidemiological OR “Population health” OR Inhalation OR “oral intake” OR “oral feed” OR “oral ingestion” Limited 2002 – present
04/09/20	((“Copper sulfate”[MeSH Terms]) OR (“Blue vitriol”[MeSH Terms]) OR (“Cupric sulfate”[MeSH Terms]) OR (“Sulfate, copper”[MeSH Terms]) OR (“Sulfate, cupric”[MeSH Terms]) OR (“Vitriol, blue”[MeSH Terms]) OR (TI/AB (“Copper sulphate” OR “Cupric sulfate” OR “Cupric sulfate anhydrous” OR “Copper monosulfate” OR “Cupric sulphate” OR “copper (II) sulfate” OR “hylinec” OR “trinagle” OR “delcup” OR “incracide E 51” OR “incracide 10A” OR “blue stone” OR “Roman vitriol” OR “Salzburg vitriol” OR “blue copperas” OR “sulfuric acid copper (2+) salt (1:1)” OR “sulfate de cuivre”))) OR “7758-98-7”[EC/RN Number] Limited 2002 – present
Science Direct 04/24/20	“Copper” OR “Cuprum” OR “Cobre” OR “Cuivre” OR “Cuprum metallicum” OR “Rame” OR “Gold bronze” OR “Bronze powder” OR “7440-50-8” AND ((MH Death OR “Body Weight” OR “respiratory system” OR “cardiovascular diseases” OR “gastrointestinal diseases” OR “hematologic diseases” OR “musculoskeletal diseases” OR “hepatic infraction” OR “renal insufficiency” OR dermatology OR “endocrine system” OR neurology OR “reproductive health” OR “developmental disabilities” OR “psychology, developmental” OR Neoplasms OR “DNA Damage”) OR AB (Death OR “Body weight” OR respiratory OR cardiovascular OR gastrointestinal OR hematological OR musculoskeletal OR hepatic OR Renal OR dermal OR ocular OR endocrine OR immunological OR neurological OR reproductive OR developmental OR Cancer OR genotoxicity OR noncancer OR “health effects”)) Limited 2002 - present
04/27/20	“Copper” OR “Cuprum” OR “Cobre” OR “Cuivre” OR “Cuprum metallicum” OR “Rame” OR “Gold bronze” OR “Bronze powder” OR “7440-50-8” AND ((MH Death OR “Body Weight” OR “respiratory system” OR “cardiovascular diseases” OR “gastrointestinal diseases” OR “hematologic diseases” OR “musculoskeletal diseases” OR “hepatic infraction” OR “renal insufficiency” OR dermatology OR “endocrine system” OR neurology OR “reproductive health” OR “developmental disabilities” OR “psychology, developmental” OR Neoplasms OR “DNA Damage”) OR ((Death OR mortality OR lethal OR “lethal dose” OR “Lethal concentration” OR fatal OR fatality OR necrosis) OR (“body weight” OR “weight loss” OR “weight gain” OR “weight change”) OR (Respiratory OR “respiratory tract” OR “respiratory organ” OR “respiratory System” OR “respiratory volume” OR “respiratory function” OR “respiratory effect” OR “respiratory organ” OR “respiratory toxicity” OR “pulmonary edema” OR “pulmonary effect” OR

Table B-2. Database Query Strings

Database Search date	Query string
	"pulmonary system" OR "pulmonary function" OR "pulmonary organ" OR "pulmonary toxicity" OR airway OR Trachea OR tracheobronchial OR "lung function" OR "lung change*" OR "Lung congestion" OR nose OR nasal OR nasopharyngeal OR larynx OR pharynx OR Bronchial or bronchi OR bronchioles OR bronchitis OR hemothorax OR alveolar OR alveoli OR irritation or irritant OR "vineyard sprayer's lung" OR cilia OR mucocilliary) OR (CVD OR Cardio OR Vascular OR "Cardiovascular system" OR "cardiovascular function" OR "cardiovascular effect" OR "cardiovascular organ" OR "cardiovascular toxicity" OR "Circulatory system" OR "circulatory function" OR "circulatory effect" OR "circulatory organ" OR "circulatory toxicity" OR "Cardiac arrest" OR "cardiac palpitation" OR "cardiac arrhythmia" OR "cardiac edema" OR "Heart rate" OR "heart failure" OR "heart attack" OR "heart muscle" OR "heart beat" OR "Blood pressure" OR "blood flow" OR "Myocardial infarction" OR "Chest pain" OR Artery OR arteries OR veins OR venules) OR (Gastrointestinal OR "Gastrointestinal system" OR "gastrointestinal function" OR "gastrointestinal effect" OR "gastrointestinal organ" OR "Digestive system" OR "digestive function" OR "digestive effect" OR "digestive organ" OR "Intestinal system" OR "intestinal function" OR "intestinal microbiota" OR "intestinal effect" OR "intestinal organ" OR "GI tract" OR "GI disorder" OR Abdominal OR Esophagus OR Stomach OR Intestine OR Pancreas OR Pancreatic OR Diarrhea OR Nausea OR Vomit OR Ulcer OR Constipation OR Emesis OR "Gut microbes" OR "gut flora" OR "gut microflora OR anorexia) OR (Hematological OR Hematology OR "hematology system" OR "hematology function" OR "hematology effect" OR "hematology toxicity" OR Hemato OR Haemato OR Blood OR "blood chemistry" OR "blood disease" OR Anemia OR Cyanosis OR Erythrocytopenia OR Leukopenia OR Thrombocytopenia OR Hemoglobin OR Erythrocyte OR Hematocrit OR "Bone marrow" OR "bone marrow decrease" OR "bone marrow hyper" OR "bone marrow hypoplasia" OR Reticulocyte OR Methemoglobin OR "Red blood cell") OR (Musculoskeletal OR Skeletal OR "skeletal system" OR "skeletal function" OR "skeletal effect" OR Muscle OR "muscle loss" OR "muscle strength" OR "muscle structure" OR Muscular OR "muscular rigidity" OR "muscular atrophy" OR "muscular structure" OR "muscular system" OR Arthritis OR "Altered bone" OR "joint pain" OR "joint ache" OR "limb pain" OR "limb ache") OR (Hepatic OR "hepatic system" OR "hepatic function" OR "hepatic effect" OR "hepatic organ" OR "hepatic response" OR "hepatic necrosis" OR "hepatic biochemical changes" OR "hepatic toxicity" OR "liver system" OR "liver function" OR "liver effect" OR "liver organ" OR "Liver enzyme" OR "liver weight" OR "liver congestion" OR "liver changes" OR "liver biochemical changes" OR "liver toxicity" OR "gallbladder system" OR "gallbladder function" OR "gallbladder organ" OR "gallbladder effect" OR "gallbladder toxicity" OR Hepatocytes OR Cirrhosis OR Jaundice OR "Hepatocellular degeneration" OR "hepatocellular hypertrophy" OR Hepatomegaly) OR ("Renal system" OR "renal function" OR "renal effect" OR "renal organ" OR "renal tubular" OR "renal toxicity" OR "kidney system" OR "kidney function" OR "Kidney effect" OR "kidney toxicity" OR "urinary system" OR "urinary function" OR "urinary effect" OR

Table B-2. Database Query Strings

Database Search date	Query string
	"Urinary toxicity" OR "bladder system" OR "bladder effect" OR "bladder function" OR "bladder toxicity" OR "Urine volume" OR "blood urea nitrogen" OR BUN OR nephropathy) OR ("Dermal system" OR "dermal function" OR "dermal effect" OR "dermal irritation" OR "dermal toxicity" OR "skin rash" OR "skin itch" OR "skin irritation" OR "skin redness" OR "skin effect" OR "skin Necrosis" OR "skin acanthosis" OR dermatitis OR edema OR ulceration OR acne) OR ("Ocular system" OR "ocular function" OR "ocular effect" OR "ocular irritation" OR "ocular toxicity" OR "eye function" OR "eye effect" OR "eye irritation" OR "eye drainage" OR "eye tearing" OR Blindness OR Myopia OR Cataracts) OR ("Endocrine system" OR "endocrine function" OR "endocrine effect" OR "endocrine gland" OR "endocrine toxicity" OR "hormone changes" OR "hormone excess" OR "hormone deficiency" OR "hormone gland" OR "hormone secretion" OR "hormone toxicity" OR Pancreas OR "Pancreatic system" OR "pancreatic function" OR "pancreatic effect" OR "pancreatic toxicity" OR "sella turcica" OR thyroid OR adrenal OR pituitary) OR ("Immunological system" OR "immunological function" OR "immunological effect" OR "immunological toxicity" OR Immune OR "immunologic system" OR "immunologic function" OR "immunologic effect" OR "immunologic response" OR "immunologic tissue" OR "immunologic toxicity" OR "Lymphoreticular changes" OR "Lymphoreticular effects" OR "Lymphoreticular function" OR "Lymphoreticular tissue" OR "Lymph node" OR Spleen OR Thymus OR Macrophage OR "white blood cell") OR (Neurological OR neurologic OR "Neuro system" OR "neuro function" OR "neuro effect" OR "neuro toxicity" OR "Nervous system" OR "Brain function" OR "brain effect" OR "brain toxicity" OR Neurotoxicant OR Neurochemistry OR Neurophysiology OR Neuropathology OR "Motor activity" OR "motor change" OR (Changes AND (behavior OR behavioral OR sensory OR cognitive)) OR Vertigo OR Drowsiness OR Headache OR Ataxia) OR (Reproductive OR "reproduction system" OR "reproduction function" OR "reproduction effect" OR "reproduction toxicity" OR Fertility OR "Maternal toxicity") OR (Developmental OR "development system" OR "developmental effect" OR "developmental toxicity" OR "developmental function" OR "developmental delay" OR "developmental abnormality" OR "developmental defect" OR (Offspring AND ("child effect" OR toxicity)) OR "In utero") OR ("altered food consumption" OR "altered water consumption" OR "Metabolic effect" OR "metabolic toxicity" OR fever) OR (Cancer OR Cancerous OR Tumor OR Carcinoma OR Carcinogen OR Mutation) OR (Genotoxicity OR "Genotoxic in vivo" OR "genotoxic in vitro" OR Mutagenicity OR Mutagenic) OR ("Mechanism of action" OR "mechanism of absorption" OR "mechanism of distribution" OR "mechanism of excretion" OR "Mechanism of metabolism" OR "Mechanism of toxic effect")

Table B-2. Database Query Strings

Database Search date	Query string
	OR (Human or Animal OR "Occupational workers" OR "Adverse effects" OR Poisoning OR Morbidity OR Inflammation OR antagonist OR inhibitor OR metabolism OR "environmental exposure" OR toxicokinetics OR pharmacokinetics OR "gene expression" OR epidemiology OR epidemiological OR "Population health" OR Inhalation OR "oral intake" OR "oral feed" OR "oral ingestion")) Limited 2002 – present
04/09/20	"Copper sulfate" OR "Blue Vitriol" OR "Cupric Sulfate" OR "Sulfate, Copper" OR "Sulfate, Cupric" OR "Vitriol, Blue" OR "Copper Sulphate" OR "Copper Sulphate anhydrous" OR "Copper Monosulfate" OR "Cupric Sulphate" OR "Copper (II) Sulfate" OR "7758-98-7" Limited 2002 - present
Scopus 05/04/20	AB ("Copper" OR "Cuprum" OR "Cobre" OR "Cuivre" OR "Cuprum metallicum" OR "Rame" OR "Gold Bronze" OR "Bronze powder" OR "CDX" OR "CI Pigment Metal 2" OR "Cu" OR "CE 7" OR "Kupfer" OR "Cutox 6010" OR "Cutox 6030" OR "C100" OR "E115" OR "1721 Gold" OR "CU M3" OR "OFHC Cu" OR "Tatum-T" OR "Paragard T 38a" OR 7440-50-8) AND AB (Death OR "Body weight" OR respiratory OR cardiovascular OR gastrointestinal OR hematological OR musculoskeletal OR hepatic OR Renal OR dermal OR ocular OR endocrine OR immunological OR neurological OR reproductive OR developmental) OR (Cancer OR genotoxicity OR noncancer OR "health effects") Time span 2002-present Limiters: Articles only
05/04/20	"Copper sulfate" OR "Blue Vitriol" OR "Cupric Sulfate" OR "Sulfate, Copper" OR "Sulfate, Cupric" OR "Vitriol, Blue" OR "Copper Sulphate" OR "Copper Sulphate anhydrous" OR "Copper Monosulfate" OR "Cupric Sulphate" OR "Copper (II) Sulfate" OR "7758-98-7" Time span 2002-2020 Limiters: Articles only

The April and May 2020 results were:

- Number of records identified from PubMed, Medline, Science Direct and Scopus (after duplicate removal): 28,002
- Number of records identified from other strategies: 29 (26 added September 2020)
- Total number of records to undergo literature screening: 28,031

B.1.2 Literature Screening

A two-step process was used to screen the literature search to identify relevant studies on copper:

- Title and abstract screen
- Full text screen

Title and Abstract Screen. Within the reference library, titles and abstracts were screened manually for relevance. Studies that were considered relevant (see Table B-1 for inclusion criteria) were moved to the second step of the literature screening process. Studies were excluded when the title and abstract clearly indicated that the study was not relevant to the toxicological profile.

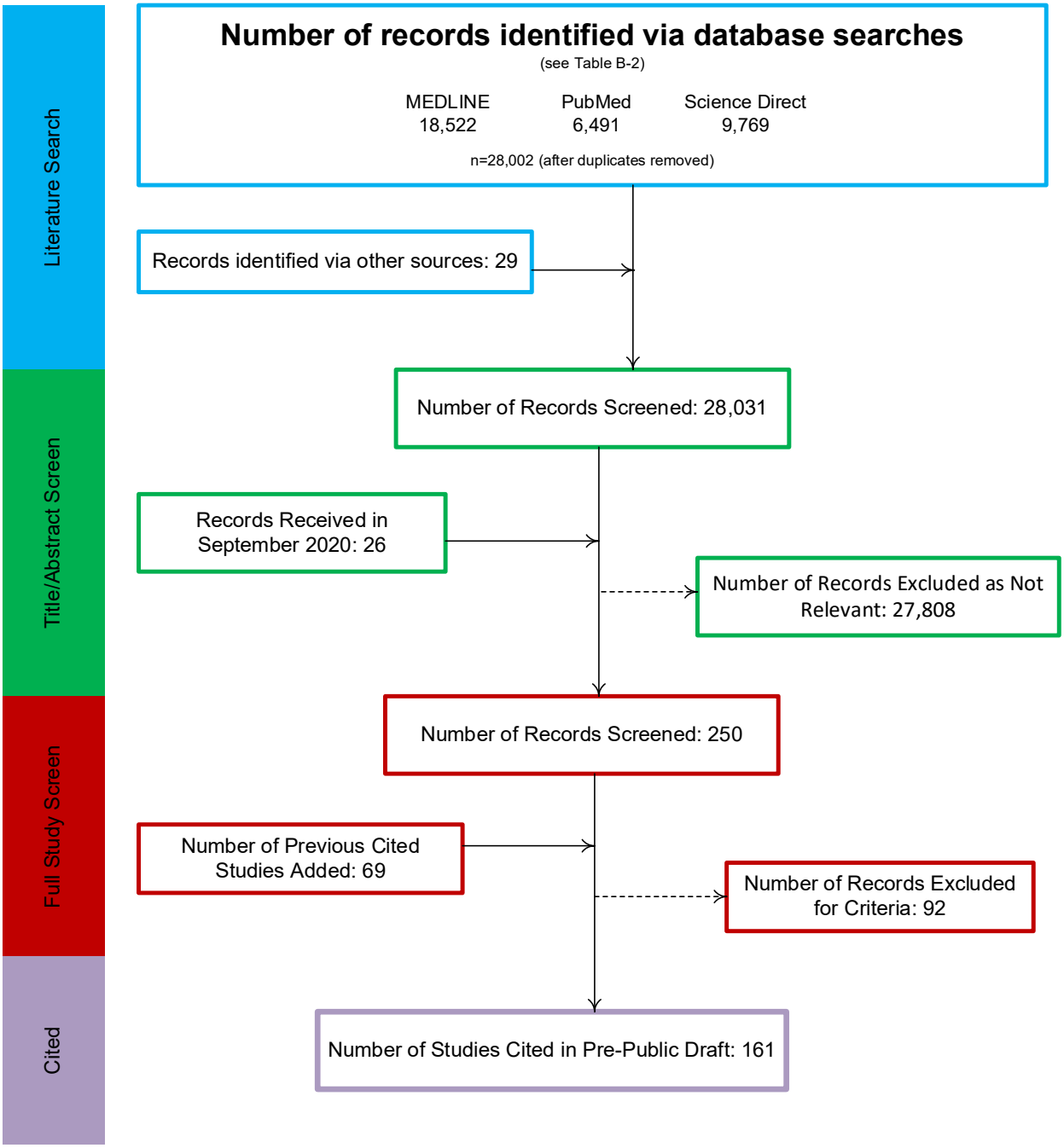
- Number of titles and abstracts screened: 28,031
- Number of studies considered relevant and moved to the next step: 223

Full Text Screen. The second step in the literature screening process was a full text review of individual studies considered relevant in the title and abstract screen step. Each study was reviewed to determine whether it was relevant for inclusion in the toxicological profile.

- Number of studies received September 2020: 26
- Number of studies undergoing full text review: 250
- Number of studies cited in the previous profile: 69
- Number of new studies cited in the profile: 92
- Total number of studies cited in the profile: 161

A summary of the results of the literature search and screening are presented in Figure B-1.

Figure B-1. April and May 2020 Literature Search Results and Screen for Copper



APPENDIX C. FRAMEWORK FOR ATSDR'S SYSTEMATIC REVIEW OF HEALTH EFFECTS DATA FOR COPPER

To increase the transparency of ATSDR's process of identifying, evaluating, synthesizing, and interpreting the scientific evidence on the health effects associated with exposure to copper, ATSDR utilized a slight modification of NTP's Office of Health Assessment and Translation (OHAT) systematic review methodology (NTP 2013, 2015; Rooney et al. 2014). ATSDR's framework is an eight-step process for systematic review with the goal of identifying the potential health hazards of exposure to copper:

- Step 1. Problem Formulation
- Step 2. Literature Search and Screen for Health Effects Studies
- Step 3. Extract Data from Health Effects Studies
- Step 4. Identify Potential Health Effect Outcomes of Concern
- Step 5. Assess the Risk of Bias for Individual Studies
- Step 6. Rate the Confidence in the Body of Evidence for Each Relevant Outcome
- Step 7. Translate Confidence Rating into Level of Evidence of Health Effects
- Step 8. Integrate Evidence to Develop Hazard Identification Conclusions

C.1 PROBLEM FORMULATION

The objective of the toxicological profile and this systematic review was to identify the potential health hazards associated with inhalation, oral, or dermal/ocular exposure to copper. The inclusion criteria used to identify relevant studies examining the health effects of copper are presented in Table C-1.

Data from human and laboratory animal studies were considered relevant for addressing this objective. Human studies were divided into two broad categories: observational epidemiology studies and controlled exposure studies. The observational epidemiology studies were further divided: cohort studies (retrospective and prospective studies), population studies (with individual data or aggregate data), and case-control studies.

C.2 LITERATURE SEARCH AND SCREEN FOR HEALTH EFFECTS STUDIES

A literature search and screen was conducted to identify studies examining the health effects of copper. The literature search framework for the toxicological profile is discussed in detail in Appendix B.

C.2.1 Literature Search

As noted in Appendix B, the literature search to update the existing toxicological profile for copper (ATSDR 2004) was restricted to studies published between 2002 to 2020. See Appendix B for the databases searched and the search strategy.

A total of 28,031 records relevant to all sections of the toxicological profile were identified (after duplicate removal).

C.2.2 Literature Screening

As described in Appendix B, a two-step process was used to screen the literature search to identify relevant studies examining the health effects of copper.

Title and Abstract Screen. In the Title and Abstract Screen step, 28,031 records were reviewed; 223 studies were considered to meet the health effects inclusion criteria in Table B-1 and were moved to the next step in the process.

Full Text Screen. In September 2020, ATSDR received 26 studies that were relevant to copper toxicity and included into the full text screening. In the second step in the literature screening process for the systematic review, a full text review of the 250 health effects studies identified in the Title and Abstract Screen was performed. Among these studies, 92 studies did not meet the inclusion criteria; some of the excluded studies were used as background information on toxicokinetics or mechanism of action or were relevant to other sections of the toxicological profile. Additionally, 69 health effects studies from the 2004 profile were included for review.

C.3 EXTRACT DATA FROM HEALTH EFFECTS STUDIES

Relevant data extracted from the individual studies selected for inclusion in the systematic review were collected in customized data forms. A summary of the type of data extracted from each study is presented in Table C-1. For references that included more than one experiment or species, data extraction records were created for each experiment or species.

A summary of the extracted data for each study is presented in the Supplemental Documents for copper and overviews of the results of the inhalation, oral and dermal exposure studies are presented in Sections 2.2 –2.19 of the profile and in the Levels of Significant Exposures tables in Section 2.1 of the profile (Table 2-1, Table 2-2, and Table 2-3, respectively).

Table C-1. Data Extracted From Individual Studies

Citation
Chemical form
Route of exposure (e.g., inhalation, oral, dermal)
Specific route (e.g., gavage in oil, drinking water)
Species
Strain
Exposure duration category (e.g., acute, intermediate, chronic)
Exposure duration
Frequency of exposure (e.g., 6 hours/day, 5 days/week)
Exposure length
Number of animals or subjects per sex per group
Dose/exposure levels
Parameters monitored
Description of the study design and method
Summary of calculations used to estimate doses (if applicable)
Summary of the study results
Reviewer's comments on the study
Outcome summary (one entry for each examined outcome)
No-observed-adverse-effect level (NOAEL) value
Lowest-observed-adverse-effect level (LOAEL) value
Effect observed at the LOAEL value

C.4 IDENTIFY POTENTIAL HEALTH EFFECT OUTCOMES OF CONCERN

Overviews of the potential health effect outcomes for copper identified in human and animal studies are presented in Table C-2 and Table C-3, respectively. The available human toxicity studies primarily evaluated the gastrointestinal endpoint including in controlled-exposure studies. Observational and controlled-exposure cohort studies and population level studies have examined a wide range of endpoints in humans. Animal studies examined all endpoints following oral exposure to copper. The hepatic endpoint was the most examined in animal studies. Animal studies have also examined body weight, renal, neurological, and reproductive effects in oral animal studies. A very limited number of animal studies examined toxicity following inhalation or dermal exposure. Gastrointestinal and hepatic effects were considered sensitive outcomes of copper exposure, as effects were observed at low doses, and are commonly reported in case studies. Studies examining these potential outcomes were carried through to Steps 4–8 of the systematic review.

Table C-2. Overview of the Health Outcomes for Copper Evaluated in Human Studies

	Body Weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological	Neurological	Reproductive	Developmental	Other Noncancer	Cancer
Inhalation Studies																	
Cohort		4 4	2 2	1 1	1 1		2 0		1 1		1 1	2 2	1 1	1 1	1 0		
Case Control																	
Population		2 2	5 5										1 1				4 4
Case Series																	
Oral Studies																	
Cohort	2 0			19 16	1 0		8 1					2 2	1 1				
Case Control							1 1								1 1		1 1
Population			1 1	1 1										2 2	1 1	1 1	
Case Series																	
Dermal Studies																	
Cohort										1 1							
Case Control																	
Population																	
Case Series																	

Number of studies examining endpoint	0	1	2	3	4	5-9	≥10
Number of studies reporting outcome	0	1	2	3	4	5-9	≥10

Table C-3. Overview of the Health Outcomes for Copper Evaluated in Experimental Animal Studies

	Body Weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological	Neurological	Reproductive	Developmental	Other Noncancer	Cancer
Inhalation Studies																	
Acute-duration		4										2					
		2										2					
Intermediate-duration		2										1					
		0										0					
Chronic-duration																	
Oral Studies																	
Acute-duration	1			3			3	2				1	1	1		2	
	1			2			3	1				1	0	1		1	
Intermediate-duration	28	8	8	10	10	2	28	19	1	1	7	7	17	18	2	5	1
	14	1	2	8	8	2	20	13	1	1	0	3	12	15	2	5	0
Chronic-duration	3				2		2									2	
	0				1		2									0	
Dermal Studies																	
Acute-duration										1							
										1							
Intermediate-duration	1	1	1	1	1	1	1	1	1	1	1	1	1	1		1	
	0	0	1	1	0	0	0	0	1	0	0	1	0	0		0	
Chronic-duration																	

Number of studies examining endpoint	0	1	2	3	4	5-9	≥10
Number of studies reporting outcome	0	1	2	3	4	5-9	≥10

C.5 ASSESS THE RISK OF BIAS FOR INDIVIDUAL STUDIES

C.5.1 Risk of Bias Assessment

The risk of bias of individual studies was assessed using OHAT's Risk of Bias Tool (NTP 2015). The risk of bias questions for observational epidemiology studies, human-controlled exposure studies, and animal experimental studies are presented in Table C-4, Table C-5, and Table C-6, respectively. Each risk of bias question was answered on a four-point scale:

- **Definitely low risk of bias (++)**
- **Probably low risk of bias (+)**
- **Probably high risk of bias (–)**
- **Definitely high risk of bias (– –)**

In general, "definitely low risk of bias" or "definitely high risk of bias" were used if the question could be answered with information explicitly stated in the study report. If the response to the question could be inferred, then "probably low risk of bias" or "probably high risk of bias" responses were typically used.

For the copper profile, the OHAT guidance on the question "Is there confidence in the exposure characterization?" was interpreted to only detract modestly from the rating in consideration of reporting of copper purity in studies. Studies were rated as probably low risk of bias (+) on this question if purity was not reported but the study does report that the test article was obtained from a commercial supplier of research chemicals, and if there is nothing in the study suggesting a risk of the test article decomposition during dosing or storage.

Table C-4. Risk of Bias Questionnaire for Observational Epidemiology Studies

Selection bias
Were the comparison groups appropriate?
Confounding bias
Did the study design or analysis account for important confounding and modifying variables?
Attrition/exclusion bias
Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?
Selective reporting bias
Were all measured outcomes reported?

Table C-5. Risk of Bias Questionnaire for Human-Controlled Exposure Studies

Selection bias
Was administered dose or exposure level adequately randomized?
Was the allocation to study groups adequately concealed?
Performance bias
Were the research personnel and human subjects blinded to the study group during the study?
Attrition/exclusion bias

Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?
Selective reporting bias
Were all measured outcomes reported?

Table C-6. Risk of Bias Questionnaire for Experimental Animal Studies

Selection bias
Was administered dose or exposure level adequately randomized?
Was the allocation to study groups adequately concealed?
Performance bias
Were experimental conditions identical across study groups?
Were the research personnel blinded to the study group during the study?
Attrition/exclusion bias
Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?
Selective reporting bias
Were all measured outcomes reported?

After the risk of bias questionnaires were completed for the health effects studies, the studies were assigned to one of three risk of bias tiers based on the responses to the key questions listed below and the responses to the remaining questions.

- Is there confidence in the exposure characterization? (only relevant for observational studies)
- Is there confidence in the outcome assessment?
- Does the study design or analysis account for important confounding and modifying variables? (only relevant for observational studies)

First Tier. Studies placed in the first tier received ratings of "definitely low" or "probably low" risk of bias on the key questions **AND** received a rating of "definitely low" or "probably low" risk of bias on the responses to at least 50% of the other applicable questions.

Second Tier. A study was placed in the second tier if it did not meet the criteria for the first or third tiers.

Third Tier. Studies placed in the third tier received ratings of "definitely high" or "probably high" risk of bias for the key questions **AND** received a rating of "definitely high" or "probably high" risk of bias on the response to at least 50% of the other applicable questions.

The results of the risk of bias assessment for the different types of copper health effects studies: observational epidemiology, controlled-exposure human studies and animal experimental studies are presented in Table C-7, Table C-8, and Table C-9, respectively.

Table C-7. Summary of Risk of Bias Assessment for Copper—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings: Selection bias — Were the comparison groups appropriate?	Confounding bias — Did the study design or analysis account for important confounding and modifying variables?*	Attrition / exclusion bias — Were outcome data complete without attrition or exclusion from analysis?	Detection bias — Is there confidence in the exposure characterization?*	Detection bias — Is there confidence in the outcome assessment?*	Selective reporting bias — Were all measured outcomes reported?	Risk of bias tier
Outcome: Gastrointestinal effects							
Cohort studies							
Buchanan et al. 1999	+	+	–	–	–	++	Second
Eife et al. 1999	–	–	+	–	+	++	Second
Pettersson et al. 2003	++	+	+	+	+	++	First
Pizarro et al. 2007	–	+	+	+	+	++	First
Suciu et al. 1981	+	–	–	–	+	+	Second
Case-control studies							
Buchanan et al. 1999	+	+	++	+	+	++	First
Cross-sectional studies							
Knobeloch et al. 1994, Study II	+	–	+	–	+	++	Second
Knobeloch et al. 1994, Study III	+	+	+	–	–	++	Second

Table C-7. Summary of Risk of Bias Assessment for Copper—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings: Selection bias — Were the comparison groups appropriate?	Confounding bias — Did the study design or analysis account for important confounding and modifying variables?*	Attrition / exclusion bias — Were outcome data complete without attrition or exclusion from analysis?	Detection bias — Is there confidence in the exposure characterization?*	Detection bias — Is there confidence in the outcome assessment?*	Selective reporting bias — Were all measured outcomes reported?	Risk of bias tier
Knobeloch et al. 1994, Study IV	+	–	–	–	+	++	Second
Knobeloch et al. 1994, Study V	+	+	+	+	–	++	Second
Knobeloch et al. 1998, Study 1	+	+	+	+	–	++	Second
Knobeloch et al. 1998, Study 2	+	–	–	–	–	++	Third
Outcome: Hepatic effects							
Cohort studies							
Eife et al. 1999	–	–	+	–	+	++	Second
Suciu et al. 1981	+	–	–	–	+	+	Second
Cross-sectional studies							
Zietz et al. 2003a	++	+	+	+	–	++	Second
Zietz et al. 2003b	++	+	+	+	–	++	Second

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable
*Key question used to assign risk of bias tier

Table C-8. Summary of Risk of Bias Assessment for Copper–Human-Controlled Exposure Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Risk of bias tier
Outcome: Gastrointestinal effects								
Oral acute exposure								
Araya et al. 2003a	++	++	++	++	+	+	++	First
Gotteland et al. 2001	++	++	++	++	–	+	++	First
Pizarro et al. 1999	++	++	++	++	+	+	++	First
Pizarro et al. 2001	++	++	++	++	–	+	++	First
Oral intermediate exposure								
Araya et al. 2001	++	++	++	+	+	+	++	First
Araya et al. 2003b	++	++	++	++	–	+	++	First
Araya et al. 2003c	++	++	++	+	+	+	++	First
Araya et al. 2004	++	++	++	++	–	–	++	Second
Olivares et al. 1998	–	–	+	+	–	–	++	Third

Table C-8. Summary of Risk of Bias Assessment for Copper–Human-Controlled Exposure Studies

Reference	Risk of bias criteria and ratings: Selection bias — Was administered dose or exposure level adequately randomized?	Selection bias — Was the allocation to study groups adequately concealed?	Performance bias — Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias — Were outcome data complete without attrition or exclusion from analysis?	Detection bias — Is there confidence in the exposure characterization?	Detection bias — Is there confidence in the outcome assessment?*	Selective reporting bias — Were all measured outcomes reported?	Risk of bias tier
Olivares et al. 2001	+	+	+	+	–	+	++	First
Outcome: Hepatic effects								
Oral acute exposure								
Pizarro et al. 1999	++	++	++	++	–	+	++	First
Pizarro et al. 2001	++	++	++	+	–	+	++	First
Oral intermediate exposure								
Araya et al. 2003b	++	++	++	++	–	+	++	First
O'Connor et al. 2003	++	++	++	++	–	+	++	First
Olivares et al. 1998	–	–	+	+	–	–	++	Third
Pratt et al. 1985	+	++	++	+	–	–	–	Second

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable
*Key question used to assign risk of bias tier

Table C-9. Summary of Risk Bias Assessment for Copper–Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were experimental conditions identical across study groups?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Risk of bias tier
Outcome: Gastrointestinal effects									
Oral acute exposure									
Kadammattil et al. 2018 (mice)	–	+	–	–	+	+	++	++	First
Yamamoto et al. 2004 (rats)	–	+	+	+	–	–	++	++	First
Yamamoto et al. 2004 (shrew)	–	+	+	+	–	–	++	++	First
Oral intermediate exposure									
Cheng et al. 2020 (mice)	+	+	++	–	++	–	+	++	First
Khushboo et al. 2018 (rats)	+	+	++	+	++	–	++	++	First
Munley 2003a (rabbits)	+	+	++	–	–	++	+	++	First
Munley 2003a (rabbits, fetus)	+	+	++	–	–	++	+	++	First
Munley 2003b (rabbits)	+	+	++	+	+	++	++	++	First

Table C-9. Summary of Risk Bias Assessment for Copper–Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were experimental conditions identical across study groups?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Risk of bias tier
Munley 2003b (rabbits, fetus)	+	+	++	+	+	++	++	++	First
NTP, 1993 (mice)	+	+	++	+	+	++	++	++	First
NTP, 1993 (rats)	+	+	++	+	+	++	++	++	First
NTP, 1993 (mice)	+	+	++	+	+	++	++	++	First
NTP, 1993 (rats)	+	+	++	+	+	++	++	++	First
Dermal intermediate exposure									
Hagemann 1992 (rats)	++	+	++	–	++	+	+	++	First
Outcome: Hepatic effects									
Oral acute exposure									
Alhusaini et al. 2018a (rats)	+	+	–	–	++	+	+	++	First
Alhusaini et al. 2018b (rats)	–	+	–	–	++	+	+	++	First

Table C-9. Summary of Risk Bias Assessment for Copper–Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were experimental conditions identical across study groups?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Risk of bias tier
Kadammattil et al. 2018 (mice)	–	+	–	–	+	+	++	++	First
Oral intermediate exposure									
Abe et al. 2008 (rats)	+	+	++	–	+	+	+	++	First
Epstein et al. 1982 (rats)	+	+	++	–	++	–	++	++	First
Hashish et al. 2016 (rats)	–	+	–	–	++	+	+	++	First
Khushboo et al. 2018 (rats)	+	+	++	+	++	–	++	++	First
Kumar et al. 2015 (rats)	–	+	++	+	+	+	++	++	First
Kumar et al. 2016a (rats)	+	+	++	–	+	+	++	++	First
Kumar et al. 2016b (rats)	+	+	++	+	+	+	++	++	First
Kumar and Sharma 1987 (rats)	+	+	–	+	– –	–	+	++	Second
Kvietkuaskaite et al. 2004 (mice)	+	+	++	+	+	–	+	++	First

Table C-9. Summary of Risk Bias Assessment for Copper–Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias — Was administered dose or exposure level adequately randomized?	Selection bias — Was the allocation to study groups adequately concealed?	Performance bias — Were experimental conditions identical across study groups?	Performance bias — Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias — Were outcome data complete without attrition or exclusion from analysis?	Detection bias — Is there confidence in the exposure characterization?	Detection bias — Is there confidence in the outcome assessment?*	Selective reporting bias — Were all measured outcomes reported?	Risk of bias tier
Munley 2003a (rabbits)	+	+	++	+	–	++	+	++	First
Munley 2003a (rabbits, fetus)	+	+	++	+	–	++	+	++	First
Munley 2003b (rabbits)	+	+	++	+	+	++	++	++	First
Munley 2003b (rabbits, fetus)	+	+	++	+	+	++	++	++	First
NTP, 1993 (mice)	+	+	++	+	+	++	++	++	First
NTP, 1993 (rats)	+	+	++	+	+	++	++	++	First
NTP, 1993 (mice)	+	+	++	+	+	++	++	++	First
NTP, 1993 (rats)	+	+	++	+	+	++	++	++	First
Rana and Kumar 1980 (rat)	– –	+	–	–	–	–	+	++	Third
Sakhaee et al. 2012 (rats)	+	+	++	–	+	–	+	++	First
Sakhaee et al. 2014 (mice)	+	+	++	+	+	–	+	++	First

Table C-9. Summary of Risk Bias Assessment for Copper–Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were experimental conditions identical across study groups?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Risk of bias tier
Seffner et al. 1997 (guinea pigs)	–	+	++	–	+	+	+	++	First
Seven et al. 2018 (rats)	+	+	–	–	+	+	+	++	First
Shen et al. 2005 (rabbits)	+	+	+	+	+	–	+	++	First
Suttle and Mills 1966 (pigs)	++	+	++	+	++	–	++	++	First
Tian et al. 2019 (rats)	+	+	++	+	+	–	++	++	First
Wu et al. 2020 (mice)	–	+	++	–	++	+	+	++	First
Oral chronic exposure									
Araya et al. 2012 (monkeys)	+	+	+	–	++	–	+	++	First
Dermal intermediate exposure									
Hagemann 1992 (rats)	++	+	++	-	++	+	+	++	First

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable
*Key question used to assign risk of bias tier

C.6 RATE THE CONFIDENCE IN THE BODY OF EVIDENCE FOR EACH RELEVANT OUTCOME

Confidences in the bodies of human and animal evidence were evaluated independently for each potential outcome. ATSDR did not evaluate the confidence in the body of evidence for carcinogenicity; rather, the Agency defaulted to the cancer weight-of-evidence assessment of other agencies including DHHS, EPA, and IARC. The confidence in the body of evidence for an association or no association between exposure to copper and a particular outcome was based on the strengths and weaknesses of individual studies. Four descriptors were used to describe the confidence in the body of evidence for effects or when no effect was found:

- **High confidence:** the true effect is highly likely to be reflected in the apparent relationship
- **Moderate confidence:** the true effect may be reflected in the apparent relationship
- **Low confidence:** the true effect may be different from the apparent relationship
- **Very low confidence:** the true effect is highly likely to be different from the apparent relationship

Confidence in the body of evidence for a particular outcome was rated for each type of study: case-control, case series, cohort, population, human-controlled exposure, and experimental animal. In the absence of data to the contrary, data for a particular outcome were collapsed across animal species, routes of exposure, and exposure durations. If species (or strain), route, or exposure duration differences were noted, then the data were treated as separate outcomes.

C.6.1 Initial Confidence Rating

In ATSDR's modification to the OHAT approach, the body of evidence for an association (or no association) between exposure to copper and a particular outcome was given an initial confidence rating based on the key features of the individual studies examining that outcome. The presence of these key features of study design was determined for individual studies using four "yes or no" questions in Distiller, which were customized for epidemiology, human controlled exposure, or experimental animal study designs. Separate questionnaires were completed for each outcome assessed in a study. The key features for observational epidemiology (cohort, population, and case-control) studies, human controlled exposure, and experimental animal studies are presented in Table C-10, Table C-11, and Table C-12, respectively. The initial confidence in the study was determined based on the number of key features present in the study design:

- **High Initial Confidence:** Studies in which the responses to the four questions were "yes".
- **Moderate Initial Confidence:** Studies in which the responses to only three of the questions were "yes".
- **Low Initial Confidence:** Studies in which the responses to only two of the questions were "yes".
- **Very Low Initial Confidence:** Studies in which the response to one or none of the questions was "yes".

Table C-10. Key Features of Study Design for Observational Epidemiology Studies

Exposure was experimentally controlled
Exposure occurred prior to the outcome
Outcome was assessed on individual level rather than at the population level
A comparison group was used

Table C-11. Key Features of Study Design for Human-Controlled Exposure Studies

A comparison group was used or the subjects served as their own control
A sufficient number of subjects were tested
Appropriate methods were used to measure outcomes (i.e., clinically-confirmed outcome versus self-reported)
Appropriate statistical analyses were performed and reported or the data were reported in such a way to allow independent statistical analysis

Table C-12. Key Features of Study Design for Experimental Animal Studies

A concurrent control group was used
A sufficient number of animals per group were tested
Appropriate parameters used to assess a potential adverse effect
Appropriate statistical analyses were performed and reported or the data were reported in such a way to allow independent statistical analysis

The presence or absence of the key features and the initial confidence levels for studies examining gastrointestinal and neurological health effects observed in the observational epidemiology, controlled-exposure human studies and animal experimental studies are presented in Table C-13, Table C-14, and Table C-15, respectively.

A summary of the initial confidence ratings for each outcome is presented in Table C-16. If individual studies for a particular outcome and study type had different study quality ratings, then the highest confidence rating for the group of studies was used to determine the initial confidence rating for the body of evidence.

Table C-13. Presence of Key Features of Study Design for Copper—Observational Epidemiology Studies

Reference	Key features				Initial study
	Controlled Exposure	Exposure prior	Outcome assess on individual level	Comparison	
Outcome: Gastrointestinal effects					
Cohort studies					
Buchanan et al. 1999	No	Yes	Yes	Yes	Moderate
Eife et al. 1999	No	Yes	Yes	No	Low
Pettersson et al. 2003	Yes	Yes	No	Yes	Moderate
Pizarro et al. 2007	No	Yes	Yes	Yes	Moderate
Suciu et al. 1981	No	Yes	Yes	Yes	Moderate
Case-control studies					
Buchanan et al. 1999	No	Yes	Yes	Yes	Moderate
Cross-sectional studies					
Knobeloch et al. 1994, Study II	No	Yes	No	No	Very Low
Knobeloch et al. 1994, Study III	No	Yes	Yes	No	Low
Knobeloch et al. 1994, Study IV	No	Yes	Yes	Yes	Moderate
Knobeloch et al. 1994, Study V	No	No	Yes	No	Very Low
Knobeloch et al. 1998, Study 1	No	No	Yes	No	Very Low
Knobeloch et al. 1998, Study 2	No	Yes	Yes	No	Low
Outcome: Hepatic effects					
Cohort studies					
Eife et al. 1999	No	Yes	Yes	No	Low
Suciu et al. 1981	No	Yes	Yes	Yes	Moderate
Cross-sectional studies					
Zietz et al. 2003a	No	Yes	Yes	No	Low
Zietz et al. 2003b	No	Yes	Yes	No	Low

Table C-14. Presence of Key Features of Study Design for Copper–Human-Controlled Exposure Studies

Reference	Key feature: Concurrent Control Group	Sufficient number of subjects per group	Appropriate parameters to assess potential effect	Adequate data for statistical analysis	Initial study confidence
Outcome: Gastrointestinal effects					
Oral acute exposure					
Araya et al. 2003a	Yes	Yes	Yes	Yes	High
Gotteland et al. 2001	Yes	Yes	Yes	Yes	High
Pizarro et al. 1999	Yes	Yes	No	Yes	Moderate
Pizarro et al. 2001	Yes	Yes	No	Yes	Moderate
Oral intermediate exposure					
Araya et al. 2001	Yes	Yes	No	Yes	Moderate
Araya et al. 2003b	Yes	Yes	No	Yes	Moderate
Araya et al. 2003c	Yes	Yes	No	Yes	Moderate
Araya et al. 2004	Yes	Yes	No	Yes	Moderate
Olivares et al. 1998	Yes	Yes	Yes	Yes	High
Olivares et al. 2001	Yes	Yes	Yes	Yes	High
Outcome: Hepatic effects					
Oral acute exposure					
Pizarro et al. 1999	Yes	Yes	No	Yes	Moderate
Pizarro et al. 2001	Yes	Yes	No	Yes	Moderate
Oral intermediate exposure					
Araya et al. 2003b	Yes	Yes	No	Yes	Moderate
O'Connor et al. 2003	Yes	Yes	Yes	Yes	High
Olivares et al. 1998	Yes	Yes	No	Yes	Moderate
Pratt et al. 1985	Yes	No	Yes	No	Low

Table C-15. Presence of Key Features of Study Design for Copper–Experimental Animal Studies

Reference	Key feature: Concurrent Control Group	Sufficient number of animals per group	Appropriate parameters to assess potential effect	Adequate data for statistical analysis	Initial study confidence
Outcome: Gastrointestinal effects					
Oral acute exposure					
Kadammattil et al. 2018 (mice)	No	No	Yes	Yes	Low
Yamamoto et al. 2004 (rats)	Yes	No	Yes	Yes	Moderate
Yamamoto et al. 2004 (shrew)	Yes	No	Yes	Yes	Moderate
Oral intermediate exposure					
Cheng et al. 2020 (mice)	Yes	Yes	Yes	Yes	High
Khushboo et al. 2018 (rats)	Yes	No	Yes	No	Low
Munley 2003a (rabbits)	Yes	No	Yes	Yes	Moderate
Munley 2003b (rabbits)	Yes	Yes	Yes	Yes	High
NTP 1993 (mice)	Yes	No	Yes	Yes	Moderate
NTP 1993 (mice)	Yes	Yes	Yes	Yes	High
NTP 1993 (rats)	Yes	No	Yes	Yes	Moderate
NTP 1993 (rats)	Yes	Yes	Yes	Yes	High
Dermal intermediate exposure					
Hagemann 1992 (rats)	Yes	Yes	Yes	Yes	High
Outcome: Hepatic effects					
Oral acute exposure					
Alhusaini et al. 2018a (rats)	Yes	Yes	Yes	Yes	High
Alhusaini et al. 2018b (rats)	Yes	Yes	Yes	Yes	High
Kadammattil et al. 2018 (mice)	Yes	No	Yes	Yes	Moderate
Oral intermediate exposure					
Abe et al. 2008 (rats)	Yes	No	Yes	Yes	Moderate
Epstein et al. 1982 (rats)	Yes	No	Yes	Yes	Moderate
Hashish and Elgaml 2016 (rats)	Yes	No	Yes	Yes	Moderate
Khushboo et al. 2018 (rats)	Yes	No	Yes	Yes	Moderate
Kumar et al. 2015 (rats)	Yes	Yes	Yes	Yes	High
Kumar et al. 2016a (rats)	Yes	No	Yes	Yes	Moderate
Kumar et al. 2016b (rats)	Yes	Yes	No	Yes	Moderate
Kumar and Sharma 1987 (rats)	Yes	Yes	No	No	Low

Table C-15. Presence of Key Features of Study Design for Copper–Experimental Animal Studies

Reference	Key feature				Initial study confidence
	Concurrent Control Group	Sufficient number of animals per group	Appropriate parameters to assess potential effect	Adequate data for statistical analysis	
Kvietkauskaite et al. 2004 (mice)	Yes	Yes	Yes	Yes	High
Munley 2003a (rabbits)	Yes	No	Yes	Yes	Moderate
Munley 2003b (rabbits)	Yes	Yes	Yes	Yes	High
NTP 1993 (mice)	Yes	No	Yes	Yes	Moderate
NTP 1993 (mice)	Yes	Yes	Yes	Yes	High
NTP 1993 (rats)	Yes	No	Yes	Yes	Moderate
NTP 1993 (rats)	Yes	Yes	Yes	Yes	High
Rana and Kumar 1980 (rats)	Yes	Yes	Yes	Yes	High
Sakhaee et al. 2012 (rats)	Yes	Yes	Yes	Yes	High
Sakhaee et al. 2014 (mice)	Yes	Yes	No	Yes	Moderate
Seffner et al. 1997 (guinea pigs)	Yes	No	Yes	Yes	Moderate
Seven et al. 2018 (rats)	Yes	No	Yes	Yes	Moderate
Shen et al. 2005 (rabbits)	Yes	No	Yes	Yes	Moderate
Suttle and Mills 1966 (pigs)	Yes	No	No	Yes	Low
Tian et al. 2019 (rats)	Yes	Yes	Yes	Yes	High
Wu et al. 2020 (mice)	Yes	Yes	Yes	Yes	High
Oral chronic exposure					
Araya et al. 2012 (monkeys)	Yes	No	Yes	Yes	Moderate
Dermal intermediate exposure					
Hagemann 1992 (rats)	Yes	Yes	Yes	Yes	High

Table C-16. Initial Confidence Rating for Copper Health Effects Studies

	Initial study confidence	Initial confidence rating
Outcome: Gastrointestinal effects		
Inhalation chronic exposure		
Human Studies		
Suciu et al. 1981	Moderate	Moderate
Oral acute exposure		
Animal Studies		
Kadammattil et al. 2018 (mice)	Low	Moderate
Yamamoto et al. 2004 (rats)	Moderate	
Yamamoto et al. 2004 (shrew)	Moderate	
Human Studies		
Araya et al. 2003a	High	High
Gotteland et al. 2001	High	
Pizarro et al. 1999	Moderate	
Pizarro et al. 2001	Moderate	
Oral intermediate exposure		
Animal Studies		
Cheng et al. 2020 (mice)	High	High
Khushboo et al. 2018 (rats)	Low	
Munley 2003a (rabbits)	Moderate	
Munley 2003b (rabbits)	High	
NTP 1993 (mice)	Moderate	
NTP 1993 (mice)	High	
NTP 1993 (rats)	Moderate	
NTP 1993 (rats)	High	
Human Studies		
Araya et al. 2001	Moderate	Moderate
Araya et al. 2003b	Moderate	
Araya et al. 2003c	Moderate	
Araya et al. 2004	Moderate	
Knobeloch et al. 1994, Study II	Very Low	
Knobeloch et al. 1994, Study III	Low	
Knobeloch et al. 1994, Study IV	Moderate	
Olivares et al. 1998	High	
Olivares et al. 2001	High	
Pettersson et al. 2003	Moderate	
Oral chronic exposure		
Human Studies		
Buchanan et al. 1999	Moderate	Moderate
Buchanan et al. 1999	Moderate	

Table C-16. Initial Confidence Rating for Copper Health Effects Studies

	Initial study confidence	Initial confidence rating
Eife et al. 1999	Low	
Knobeloch et al. 1994, Study V	Very Low	
Knobeloch et al. 1998, Study 1	Very Low	
Knobeloch et al. 1998, Study 2	Low	
Pizarro et al. 2007	Moderate	
Dermal intermediate exposure		
Animal Studies		
Hagemann 1992 (rats)	High	High
Outcome: Hepatic Effects		
Inhalation chronic exposure		
Human Studies		
Suciu et al. 1981	Moderate	Moderate
Oral acute exposure		
Animal Studies		
Alhusaini et al. 2018a (rats)	High	
Alhusaini et al. 2018b (rats)	High	High
Kadammattil et al. 2018 (mice)	Moderate	
Human Studies		
Pizarro et al. 1999	Moderate	Moderate
Pizarro et al. 2001	Moderate	
Oral intermediate exposure		
Animal Studies		
Abe et al. 2008 (rats)	Moderate	
Epstein et al. 1982 (rats)	Moderate	
Hashish and Elgaml 2016 (rats)	Moderate	
Khushboo et al. 2018 (rats)	Moderate	
Kumar et al. 2015 (rats)	High	
Kumar et al. 2016a (rats)	Moderate	
Kumar et al. 2016b (rats)	Moderate	
Kumar and Sharma 1987 (rats)	Low	
Kvietkauskaite et al. 2004 (mice)	High	
Munley 2003a (rabbits)	Moderate	
Munley 2003b (rabbits)	High	High
NTP 1993 (mice)	Moderate	
NTP 1993 (mice)	High	
NTP 1993 (rats)	Moderate	
NTP 1993 (rats)	High	
Rana and Kumar 1980 (rats)	High	

Table C-16. Initial Confidence Rating for Copper Health Effects Studies

	Initial study confidence	Initial confidence rating
Sakhaee et al. 2012 (rats)	High	
Sakhaee et al. 2014 (mice)	Moderate	
Seffner et al. 1997 (guinea pigs)	Moderate	
Seven et al. 2018 (rats)	Moderate	
Shen et al. 2005 (rabbits)	Moderate	
Suttle and Mills 1966 (pigs)	Low	
Tian et al. 2019 (rats)	High	
Wu et al. 2020 (mice)	High	
Human Studies		
Araya et al. 2003b	Moderate	Moderate
O'Connor et al. 2003	High	
Olivares et al. 1998	Moderate	
Pratt et al. 1985	Low	
Zietz et al. 2003a	Low	
Zietz et al. 2003b	Low	
Oral chronic exposure		
Animal Studies		
Araya et al. 2012 (monkey)	Moderate	Moderate
Human Studies		
Eife et al. 1999	Low	Low
Dermal intermediate exposure		
Animal Studies		
Hagemann 1992 (rats)	High	High

C.6.2 Adjustment of the Confidence Rating

The initial confidence rating was then downgraded or upgraded depending on whether there were substantial issues that would decrease or increase confidence in the body of evidence. The nine properties of the body of evidence that were considered are listed below. The summaries of the assessment of the confidence in the body of evidence for gastrointestinal and hepatic effects are presented in Table C-17. If the confidence ratings for a particular outcome were based on more than one type of human study, then the highest confidence rating was used for subsequent analyses. An overview of the confidence in the body of evidence for all health effects associated with copper exposure is presented in Table C-18.

Five properties of the body of evidence were considered to determine whether the confidence rating should be downgraded:

- **Risk of bias.** Evaluation of whether there is substantial risk of bias across most of the studies examining the outcome. This evaluation used the risk of bias tier groupings for individual studies examining a particular outcome (Table C-7, Table C-8, and Table C-9). Below are the criteria

used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for risk of bias:

- No downgrade if most studies are in the risk of bias first tier
- Downgrade one confidence level if most studies are in the risk of bias second tier
- Downgrade two confidence levels if most studies are in the risk of bias third tier

- **Unexplained inconsistency.** Evaluation of whether there is inconsistency or large variability in the magnitude or direction of estimates of effect across studies that cannot be explained. Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for unexplained inconsistency:
  - No downgrade if there is little inconsistency across studies or if only one study evaluated the outcome
  - Downgrade one confidence level if there is variability across studies in the magnitude or direction of the effect
  - Downgrade two confidence levels if there is substantial variability across studies in the magnitude or direct of the effect

- **Indirectness.** Evaluation of four factors that can affect the applicability, generalizability, and relevance of the studies:
  - Relevance of the animal model to human health—unless otherwise indicated, studies in rats, mice, and other mammalian species are considered relevant to humans
  - Directness of the endpoints to the primary health outcome—examples of secondary outcomes or nonspecific outcomes include organ weight in the absence of histopathology or clinical chemistry findings in the absence of target tissue effects
  - Nature of the exposure in human studies and route of administration in animal studies—inhalation, oral, and dermal exposure routes are considered relevant unless there are compelling data to the contrary
  - Duration of treatment in animal studies and length of time between exposure and outcome assessment in animal and prospective human studies—this should be considered on an outcome-specific basis

Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for indirectness:

- No downgrade if none of the factors are considered indirect
- Downgrade one confidence level if one of the factors is considered indirect
- Downgrade two confidence levels if two or more of the factors are considered indirect

- **Imprecision.** Evaluation of the narrowness of the effect size estimates and whether the studies have adequate statistical power. Data are considered imprecise when the ratio of the upper to lower 95% CIs for most studies is ≥10 for tests of ratio measures (e.g., odds ratios) and ≥100 for absolute measures (e.g., percent control response). Adequate statistical power is determined if the study can detect a potentially biologically meaningful difference between groups (20% change from control response for categorical data or risk ratio of 1.5 for continuous data). Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for imprecision:
  - No downgrade if there are no serious imprecisions
  - Downgrade one confidence level for serious imprecisions
  - Downgrade two confidence levels for very serious imprecisions

- **Publication bias.** Evaluation of the concern that studies with statistically significant results are more likely to be published than studies without statistically significant results.

- Downgrade one level of confidence for cases where there is serious concern with publication bias

Four properties of the body of evidence were considered to determine whether the confidence rating should be upgraded:

- **Large magnitude of effect.** Evaluation of whether the magnitude of effect is sufficiently large so that it is unlikely to have occurred as a result of bias from potential confounding factors.
  - Upgrade one confidence level if there is evidence of a large magnitude of effect in a few studies, provided that the studies have an overall low risk of bias and there is no serious unexplained inconsistency among the studies of similar dose or exposure levels; confidence can also be upgraded if there is one study examining the outcome, provided that the study has an overall low risk of bias

- **Dose response.** Evaluation of the dose-response relationships measured within a study and across studies. Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:
  - Upgrade one confidence level for evidence of a monotonic dose-response gradient
  - Upgrade one confidence level for evidence of a non-monotonic dose-response gradient where there is prior knowledge that supports a non-monotonic dose-response and a non-monotonic dose-response gradient is observed across studies

- **Plausible confounding or other residual biases.** This factor primarily applies to human studies and is an evaluation of unmeasured determinants of an outcome such as residual bias towards the null (e.g., "healthy worker" effect) or residual bias suggesting a spurious effect (e.g., recall bias). Below is the criterion used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:
  - Upgrade one confidence level for evidence that residual confounding or bias would underestimate an apparent association or treatment effect (i.e., bias toward the null) or suggest a spurious effect when results suggest no effect

- **Consistency in the body of evidence.** Evaluation of consistency across animal models and species, consistency across independent studies of different human populations and exposure scenarios, and consistency across human study types. Below is the criterion used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:
  - Upgrade one confidence level if there is a high degree of consistency in the database

The results of this assessment are presented in Table C-17, and the final confidence in the body of literature for the neurological endpoint is presented in Table C-18.

Table C-17. Adjustments to the Initial Confidence in the Body of Evidence

	Initial confidence	Adjustments to the initial confidence rating	Final confidence
Outcome: Gastrointestinal effects			
Human studies	Moderate	+1 Consistency in the body of evidence	High
Animal studies	Moderate	None	Moderate
Outcome: Hepatic effects			
Human studies	Moderate	-1 Indirectness: length of time between exposure and outcome assessment	Low
Animal studies	High	None	High

Table C-18. Confidence in the Body of Evidence for Copper

	Confidence in body of evidence	
Outcome	Human Studies	Animal Studies
Gastrointestinal effects	High	Moderate
Hepatic effects	Low	High

C.7 TRANSLATE CONFIDENCE RATING INTO LEVEL OF EVIDENCE OF HEALTH EFFECTS

In the seventh step of the systematic review of the health effects data for copper, the confidence in the body of evidence for specific outcomes was translated to a level of evidence rating. The level of evidence rating reflected the confidence in the body of evidence and the direction of the effect (i.e., toxicity or no toxicity); route-specific differences were noted. The level of evidence for health effects was rated on a five-point scale:

- **High level of evidence:** High confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Moderate level of evidence:** Moderate confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Low level of evidence:** Low confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Evidence of no health effect:** High confidence in the body of evidence that exposure to the substance is not associated with the health outcome
- **Inadequate evidence:** Low or moderate confidence in the body of evidence that exposure to the substance is not associated with the health outcome OR very low confidence in the body of evidence for an association between exposure to the substance and the health outcome

A summary of the level of evidence of health effects for copper, is presented in Table C-19.

Table C-19. Level of Evidence of Health Effects for Copper

Outcome	Confidence in body of evidence	Direction of health effect	Level of evidence for health effect
Human Studies			
Gastrointestinal effects	High	Health Effect	High
Hepatic effects	Low	No Health Effect	Inadequate
Animal Studies			
Gastrointestinal effects	Moderate	Health Effect	Moderate
Hepatic effects	High	Health Effect	High

C.8 INTEGRATE EVIDENCE TO DEVELOP HAZARD IDENTIFICATION CONCLUSIONS

The final step involved the integration of the evidence streams for the human studies and animal studies to allow for a determination of hazard identification conclusions. For health effects, there were four hazard identification conclusion categories:

- **Known** to be a hazard to humans
- **Presumed** to be a hazard to humans
- **Suspected** to be a hazard to humans
- **Not classifiable** as to the hazard to humans

The initial hazard identification was based on the highest level of evidence in the human studies and the level of evidence in the animal studies; if there were no data for one evidence stream (human or animal), then the hazard identification was based on the one data stream (equivalent to treating the missing evidence stream as having low level of evidence). The hazard identification scheme is presented in Figure C-1 and described below:

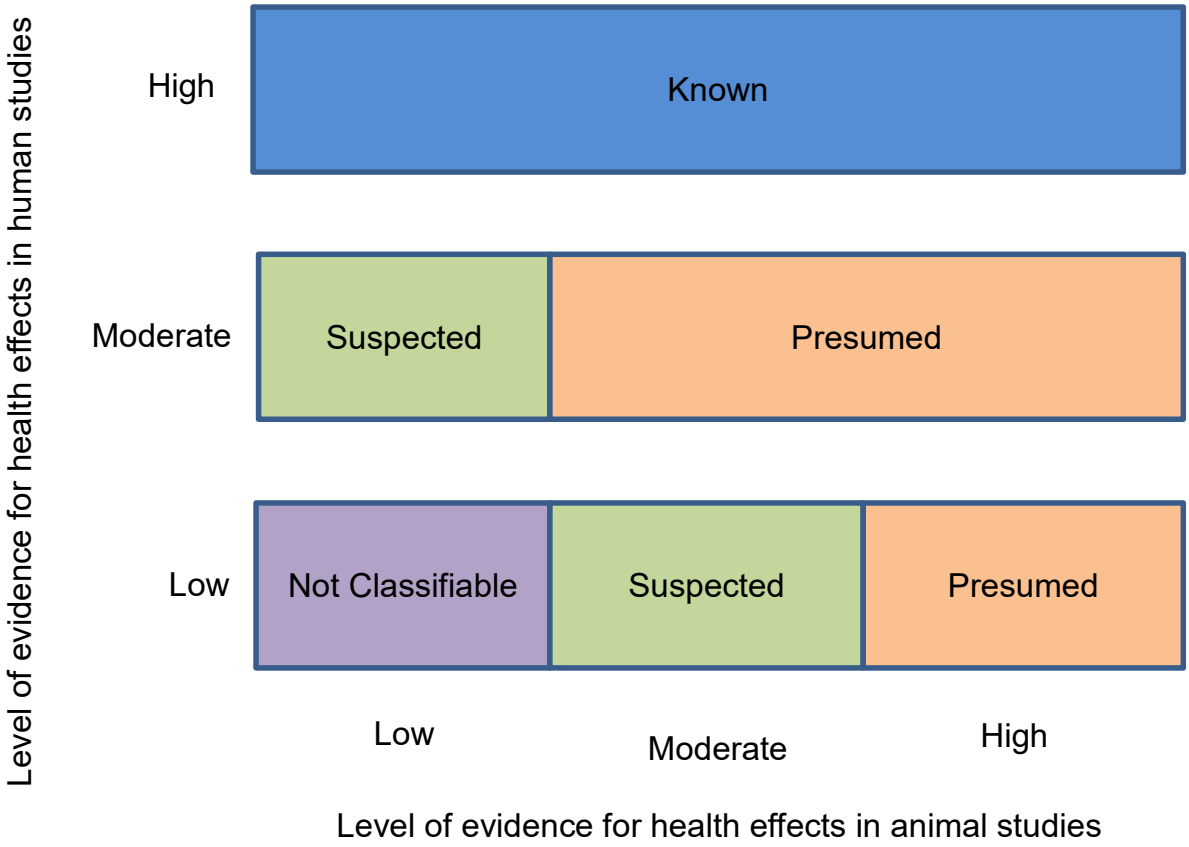
- **Known:** A health effect in this category would have:
  - High level of evidence for health effects in human studies **AND** a high, moderate, or low level of evidence in animal studies.

- **Presumed:** A health effect in this category would have:
  - Moderate level of evidence in human studies **AND** high or moderate level of evidence in animal studies **OR**
  - Low level of evidence in human studies **AND** high level of evidence in animal studies

- **Suspected:** A health effect in this category would have:
  - Moderate level of evidence in human studies **AND** low level of evidence in animal studies **OR**
  - Low level of evidence in human studies **AND** moderate level of evidence in animal studies
- **Not classifiable:** A health effect in this category would have:
  - Low level of evidence in human studies **AND** low level of evidence in animal studies

Other relevant data such as mechanistic or mode-of-action data were considered to raise or lower the level of the hazard identification conclusion by providing information that supported or opposed biological plausibility.

Figure C-1. Hazard Identification Scheme



Two hazard identification conclusion categories were used when the data indicated that there may be no health effect in humans:

- **Not identified** to be a hazard in humans
- **Inadequate** to determine hazard to humans

If the human level of evidence conclusion of no health effect was supported by the animal evidence of no health effect, then the hazard identification conclusion category of "not identified" was used. If the human or animal level of evidence was considered inadequate, then a hazard identification conclusion category of "inadequate" was used. As with the hazard identification for health effects, the impact of other relevant data was also considered for no health effect data.

The hazard identification conclusions for copper are listed below and summarized in Table C-20.

Presumed Health Effects

- High level of evidence in epidemiological studies where humans were orally exposed to copper in their drinking water (Eife et al. 1999; Gotteland et al. 2001; Knobeloch et al. 1994, 1998). One occupational study of workers who inhaled copper dust showed gastrointestinal symptoms (Suciu et al. 1981).
- High level of evidence in humans in controlled-exposure studies from acute exposure to copper sulfate in drinking water (Araya et al. 2001, 2003a; Pizarro et al. 1999, 2001) and

intermediate exposure to copper sulfate in drinking water or other juice (Araya et al. 2003b, 2003c, 2004; Olivares et al. 2001).
- Moderate level of evidence of effects in mice, rats and shrews from acute exposure to copper sulfate (Kadammattil et al. 2018; Yamamoto et al. 2004); and intermediate exposure to copper chloride (Cheng et al. 2020), copper sulfate (Khushboo et al. 2018; NTP 1993), copper hydroxide (Munley 2003a, 2003b).

Suspected Health Effects

- Hepatic effects
  - Low level of evidence in human studies as no changes in liver function measured by hepatic enzyme levels were observed from intermediate exposure to copper sulfate (Araya et al. 2003b; Olivares et al. 1998) and copper (O'Connor et al. 2003; Zietz et al. 2003a, 2003b). Evidence from inhalation exposure in one occupational study in workers who inhaled copper dust (Suciu et al. 1981).
  - High level evidence of effects in rats and mice from acute exposure to copper compounds (Alhusaini et al. 2018a, 2018b; Kadammattil et al. 2018) and intermediate exposure to copper compounds (Epstein et al. 1982; Hashish and Elgaml 2016; Khushboo et al. 2018; Kumar et al. 2015, 2016b; Kumar and Sharma 1987; Kvietkauskaite et al. 2004; NTP 1993; Sakhaee et al. 2012, 2014; Seven et al. 2018; Tian et al. 2019; Wu et al. 2020). Evidence of effects from intermediate exposure in pigs (Suttle and Mills 1966) and rabbits (Munley 2003a, 200b; Shen et al. 2005).

Table C-20. Hazard Identification Conclusions for Copper

Outcome	Hazard identification
Gastrointestinal effects	Presumed
Hepatic effects	Suspected

APPENDIX D. USER'S GUIDE

Chapter 1. Relevance to Public Health

This chapter provides an overview of U.S. exposures, a summary of health effects based on evaluations of existing toxicologic, epidemiologic, and toxicokinetic information, and an overview of the minimal risk levels. This is designed to present interpretive, weight-of-evidence discussions for human health endpoints by addressing the following questions:

1. What effects are known to occur in humans?

2. What effects observed in animals are likely to be of concern to humans?

3. What exposure conditions are likely to be of concern to humans, especially around hazardous waste sites?

Minimal Risk Levels (MRLs)

Where sufficient toxicologic information is available, ATSDR derives MRLs for inhalation and oral routes of entry at each duration of exposure (acute, intermediate, and chronic). These MRLs are not meant to support regulatory action, but to acquaint health professionals with exposure levels at which adverse health effects are not expected to occur in humans.

MRLs should help physicians and public health officials determine the safety of a community living near a hazardous substance emission, given the concentration of a contaminant in air or the estimated daily dose in water. MRLs are based largely on toxicological studies in animals and on reports of human occupational exposure.

MRL users should be familiar with the toxicologic information on which the number is based. Section 1.2, Summary of Health Effects, contains basic information known about the substance. Other sections, such as Section 3.2 Children and Other Populations that are Unusually Susceptible and Section 3.4 Interactions with Other Substances, provide important supplemental information.

MRL users should also understand the MRL derivation methodology. MRLs are derived using a modified version of the risk assessment methodology that the Environmental Protection Agency (EPA) provides (Barnes and Dourson 1988) to determine reference doses (RfDs) for lifetime exposure.

To derive an MRL, ATSDR generally selects the most sensitive endpoint which, in its best judgement, represents the most sensitive human health effect for a given exposure route and duration. ATSDR cannot make this judgement or derive an MRL unless information (quantitative or qualitative) is available for all potential systemic, neurological, and developmental effects. If this information and reliable quantitative data on the chosen endpoint are available, ATSDR derives an MRL using the most sensitive species (when information from multiple species is available) with the highest no-observed-adverse-effect level (NOAEL) that does not exceed any adverse effect levels. When a NOAEL is not available, a lowest-observed-adverse-effect level (LOAEL) can be used to derive an MRL, and an uncertainty factor of 10 must be employed. Additional uncertainty factors of 10 must be used both for human variability to protect sensitive subpopulations (people who are most susceptible to the health effects caused by the substance) and for interspecies variability (extrapolation from animals to humans). In deriving an MRL, these individual uncertainty factors are multiplied together. The product is then divided into the

inhalation concentration or oral dosage selected from the study. Uncertainty factors used in developing a substance-specific MRL are provided in the footnotes of the levels of significant exposure (LSE) tables that are provided in Chapter 2. Detailed discussions of the MRLs are presented in Appendix A.

Chapter 2. Health Effects

Tables and Figures for Levels of Significant Exposure (LSE)

Tables and figures are used to summarize health effects and illustrate graphically levels of exposure associated with those effects. These levels cover health effects observed at increasing dose concentrations and durations, differences in response by species and MRLs to humans for noncancer endpoints. The LSE tables and figures can be used for a quick review of the health effects and to locate data for a specific exposure scenario. The LSE tables and figures should always be used in conjunction with the text. All entries in these tables and figures represent studies that provide reliable, quantitative estimates of NOAELs, LOAELs, or Cancer Effect Levels (CELs).

The legends presented below demonstrate the application of these tables and figures. Representative examples of LSE tables and figures follow. The numbers in the left column of the legends correspond to the numbers in the example table and figure.

TABLE LEGEND

See Sample LSE Table (page D-5)

(1) Route of exposure. One of the first considerations when reviewing the toxicity of a substance using these tables and figures should be the relevant and appropriate route of exposure. Typically, when sufficient data exist, three LSE tables and two LSE figures are presented in the document. The three LSE tables present data on the three principal routes of exposure (i.e., inhalation, oral, and dermal). LSE figures are limited to the inhalation and oral routes. Not all substances will have data on each route of exposure and will not, therefore, have all five of the tables and figures. Profiles with more than one chemical may have more LSE tables and figures.

(2) Exposure period. Three exposure periods—acute (<15 days), intermediate (15–364 days), and chronic (≥365 days)—are presented within each relevant route of exposure. In this example, two oral studies of chronic-duration exposure are reported. For quick reference to health effects occurring from a known length of exposure, locate the applicable exposure period within the LSE table and figure.

(3) Figure key. Each key number in the LSE table links study information to one or more data points using the same key number in the corresponding LSE figure. In this example, the study represented by key number 51 identified NOAELs and less serious LOAELs (also see the three "51R" data points in sample LSE Figure 2-X).

(4) Species (strain) No./group. The test species (and strain), whether animal or human, are identified in this column. The column also contains information on the number of subjects and sex per group. Chapter 1, Relevance to Public Health, covers the relevance of animal data to human toxicity and Section 3.1, Toxicokinetics, contains any available information on comparative toxicokinetics. Although NOAELs and LOAELs are species specific, the levels are extrapolated to equivalent human doses to derive an MRL.

(5) Exposure parameters/doses. The duration of the study and exposure regimens are provided in these columns. This permits comparison of NOAELs and LOAELs from different studies. In this case (key number 51), rats were orally exposed to "Chemical X" via feed for 2 years. For a more complete review of the dosing regimen, refer to the appropriate sections of the text or the original reference paper (i.e., Aida et al. 1992).

(6) Parameters monitored. This column lists the parameters used to assess health effects. Parameters monitored could include serum (blood) chemistry (BC), biochemical changes (BI), body weight (BW), clinical signs (CS), developmental toxicity (DX), food intake (FI), gross necropsy (GN), hematology (HE), histopathology (HP), immune function (IX), lethality (LE), neurological function (NX), organ function (OF), ophthalmology (OP), organ weight (OW), reproductive function (RX), urinalysis (UR), and water intake (WI).

(7) Endpoint. This column lists the endpoint examined. The major categories of health endpoints included in LSE tables and figures are death, body weight, respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, dermal, ocular, endocrine, immunological, neurological, reproductive, developmental, other noncancer, and cancer. "Other noncancer" refers to any effect (e.g., alterations in blood glucose levels) not covered in these systems. In the example of key number 51, three endpoints (body weight, hematological, and hepatic) were investigated.

(8) NOAEL. A NOAEL is the highest exposure level at which no adverse effects were seen in the organ system studied. The body weight effect reported in key number 51 is a NOAEL at 25.5 mg/kg/day. NOAELs are not reported for cancer and death; with the exception of these two endpoints, this field is left blank if no NOAEL was identified in the study.

(9) LOAEL. A LOAEL is the lowest dose used in the study that caused an adverse health effect. LOAELs have been classified into "Less Serious" and "Serious" effects. These distinctions help readers identify the levels of exposure at which adverse health effects first appear and the gradation of effects with increasing dose. A brief description of the specific endpoint used to quantify the adverse effect accompanies the LOAEL. Key number 51 reports a less serious LOAEL of 6.1 mg/kg/day for the hepatic system, which was used to derive a chronic exposure, oral MRL of 0.008 mg/kg/day (see footnote "c"). MRLs are not derived from serious LOAELs. A cancer effect level (CEL) is the lowest exposure level associated with the onset of carcinogenesis in experimental or epidemiologic studies. CELs are always considered serious effects. The LSE tables and figures do not contain NOAELs for cancer, but the text may report doses not causing measurable cancer increases. If no LOAEL/CEL values were identified in the study, this field is left blank.

(10) Reference. The complete reference citation is provided in Chapter 8 of the profile.

(11) Footnotes. Explanations of abbreviations or reference notes for data in the LSE tables are found in the footnotes. For example, footnote "c" indicates that the LOAEL of 6.1 mg/kg/day in key number 51 was used to derive an oral MRL of 0.008 mg/kg/day.

FIGURE LEGEND

See Sample LSE Figure (page D-6)

LSE figures graphically illustrate the data presented in the corresponding LSE tables. Figures help the reader quickly compare health effects according to exposure concentrations for particular exposure periods.

(12) Exposure period. The same exposure periods appear as in the LSE table. In this example, health effects observed within the chronic exposure period are illustrated.

(13) Endpoint. These are the categories of health effects for which reliable quantitative data exist. The same health effect endpoints appear in the LSE table.

(14) Levels of exposure. Concentrations or doses for each health effect in the LSE tables are graphically displayed in the LSE figures. Exposure concentration or dose is measured on the log scale "y" axis. Inhalation exposure is reported in mg/m^3 or ppm and oral exposure is reported in mg/kg/day.

(15) LOAEL. In this example, the half-shaded circle that is designated 51R identifies a LOAEL critical endpoint in the rat upon which a chronic oral exposure MRL is based. The key number 51 corresponds to the entry in the LSE table. The dashed descending arrow indicates the extrapolation from the exposure level of 6.1 mg/kg/day (see entry 51 in the sample LSE table) to the MRL of 0.008 mg/kg/day (see footnote "c" in the sample LSE table).

(16) CEL. Key number 59R is one of studies for which CELs were derived. The diamond symbol refers to a CEL for the test species (rat). The number 59 corresponds to the entry in the LSE table.

(17) Key to LSE figure. The key provides the abbreviations and symbols used in the figure.

Table 2-X. Levels of Significant Exposure to [Chemical X] – Oral (1)

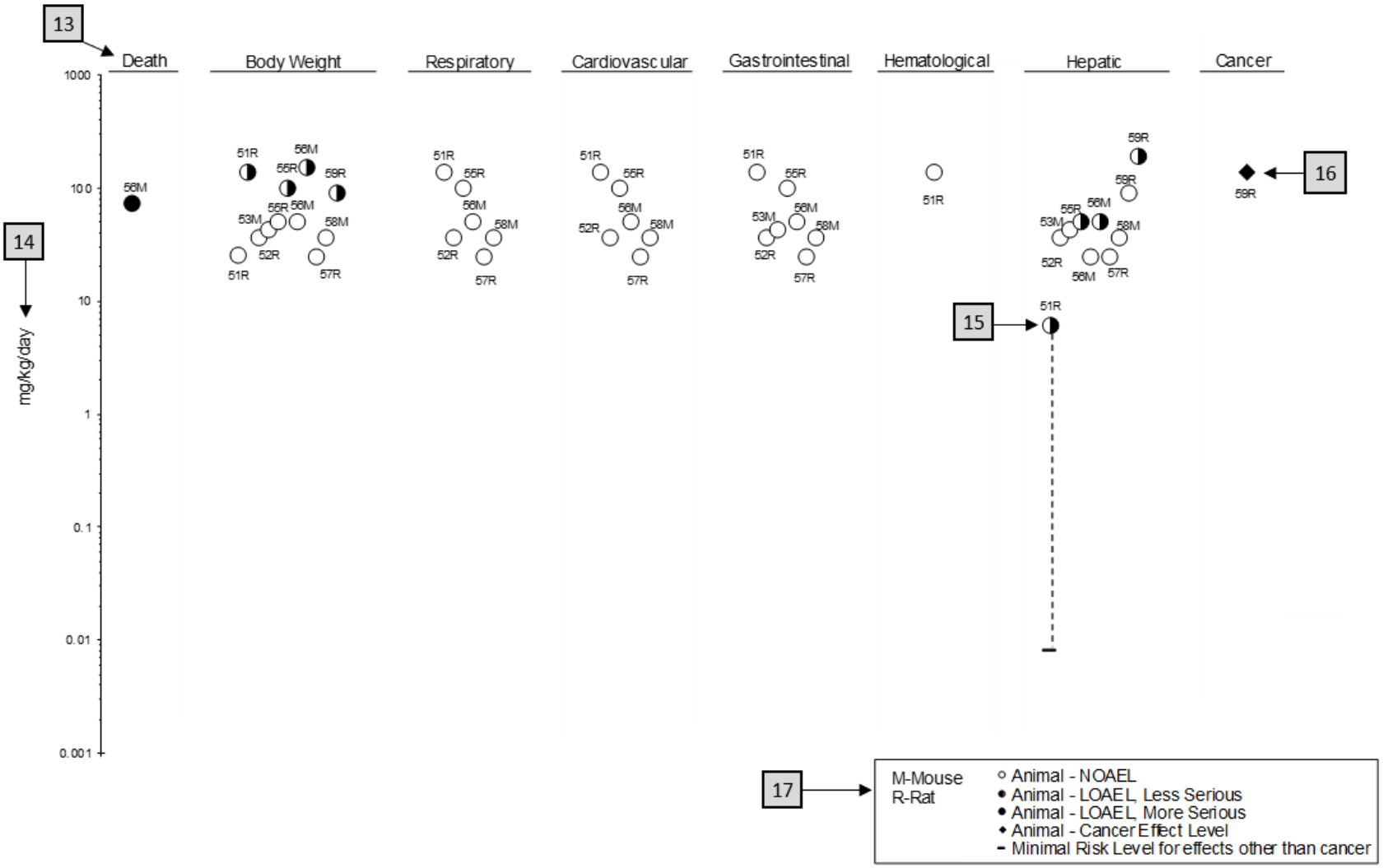
Figure key[a] (3)	Species (strain) No./group (4)	Exposure parameters (5)	Doses (mg/kg/day) (5)	Parameters monitored (6)	Endpoint (7)	NOAEL (mg/kg/day) (8)	Less serious LOAEL (mg/kg/day) (9)	Serious LOAEL (mg/kg/day) (9)	Effect
CHRONIC EXPOSURE (2)									
51	Rat (Wistar) 40 M, 40 F	2 years (F)	M: 0, 6.1, 25.5, 138.0 F: 0, 8.0, 31.7, 168.4	CS, WI, BW, OW, HE, BC, HP	Bd wt	25.5	138.0		Decreased body weight gain in males (23–25%) and females (31–39%)
					Hemato	138.0			
					Hepatic		6.1[c]		Increases in absolute and relative weights at ≥6.1/8.0 mg/kg/day after 12 months of exposure; fatty generation at ≥6.1 mg/kg/day in males and at ≥31.7 mg/kg/day in females, and granulomas in females at 31.7 and 168.4 mg/kg/day after 12, 18, or 24 months of exposure and in males at ≥6.1 mg/kg/day only after 24 months of exposure
Aida et al. 1992 (10)									
52	Rat (F344) 78 M	104 weeks (W)	0, 3.9, 20.6, 36.3	CS, BW, FI, BC, OW, HP	Hepatic	36.3			
					Renal	20.6	36.3		Increased incidence of renal tubular cell hyperplasia
					Endocr	36.3			
George et al. 2002									
59	Rat (Wistar) 58M, 58F	Lifetime (W)	M: 0, 90 F: 0, 190	BW, HP	Cancer		190 F		Increased incidence of hepatic neoplastic nodules in females only; no additional description of the tumors was provided
Tumasonis et al. 1985									

[a]The number corresponds to entries in Figure 2-x.

(11) [b]Used to derive an acute-duration oral minimal risk level (MRL) of 0.1 mg/kg/day based on the $BMDL_{05}$ of 10 mg/kg/day and an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

[c]Used to derive a chronic-duration oral MRL of 0.008 mg/kg/day based on the $BMDL_{10}$ of 0.78 mg/kg/day and an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

Figure 2-X. Levels of Significant Exposure to [Chemical X] - Oral



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APPENDIX E. QUICK REFERENCE FOR HEALTH CARE PROVIDERS

Toxicological Profiles are a unique compilation of toxicological information on a given hazardous substance. Each profile reflects a comprehensive and extensive evaluation, summary, and interpretation of available toxicologic and epidemiologic information on a substance. Health care providers treating patients potentially exposed to hazardous substances may find the following information helpful for fast answers to often-asked questions.

Primary Chapters/Sections of Interest

Chapter 1: Relevance to Public Health: The Relevance to Public Health Section provides an overview of exposure and health effects and evaluates, interprets, and assesses the significance of toxicity data to human health. A table listing minimal risk levels (MRLs) is also included in this chapter.

Chapter 2: Health Effects: Specific health effects identified in both human and animal studies are reported by type of health effect (e.g., death, hepatic, renal, immune, reproductive), route of exposure (e.g., inhalation, oral, dermal), and length of exposure (e.g., acute, intermediate, and chronic).
NOTE: Not all health effects reported in this section are necessarily observed in the clinical setting.

Pediatrics:

Section 3.2 Children and Other Populations that are Unusually Susceptible
Section 3.3 Biomarkers of Exposure and Effect

ATSDR Information Center

Phone: 1-800-CDC-INFO (800-232-4636) or 1-888-232-6348 (TTY)
Internet: http://www.atsdr.cdc.gov

ATSDR develops educational and informational materials for health care providers categorized by hazardous substance, clinical condition, and/or by susceptible population. The following additional materials are available online:

Physician Briefs discuss health effects and approaches to patient management in a brief/factsheet style. *Physician Overviews* are narrated PowerPoint presentations with Continuing Education credit available (see https://www.atsdr.cdc.gov/emes/health_professionals/index.html).

Managing Hazardous Materials Incidents is a set of recommendations for on-scene (prehospital) and hospital medical management of patients exposed during a hazardous materials incident (see https://www.atsdr.cdc.gov/MHMI/index.html).

Fact Sheets (ToxFAQs™) provide answers to frequently asked questions about toxic substances (see https://www.atsdr.cdc.gov/toxfaqs/Index.asp).

Other Agencies and Organizations

The National Center for Environmental Health (NCEH) focuses on preventing or controlling disease, injury, and disability related to the interactions between people and their environment outside the workplace. Contact: NCEH, Mailstop F-29, 4770 Buford Highway, NE, Atlanta, GA 30341-3724 • Phone: 770-488-7000 • FAX: 770-488-7015 • Web Page: https://www.cdc.gov/nceh/.

The National Institute for Occupational Safety and Health (NIOSH) conducts research on occupational diseases and injuries, responds to requests for assistance by investigating problems of health and safety in the workplace, recommends standards to the Occupational Safety and Health Administration (OSHA) and the Mine Safety and Health Administration (MSHA), and trains professionals in occupational safety and health. Contact: NIOSH, 395 E Street, S.W., Suite 9200, Patriots Plaza Building, Washington, DC 20201 • Phone: 202-245-0625 or 1-800-CDC-INFO (800-232-4636) • Web Page: https://www.cdc.gov/niosh/.

The National Institute of Environmental Health Sciences (NIEHS) is the principal federal agency for biomedical research on the effects of chemical, physical, and biologic environmental agents on human health and well-being. Contact: NIEHS, PO Box 12233, 104 T.W. Alexander Drive Research Triangle Park, NC 27709 • Phone: 919-541-3212 • Web Page: https://www.niehs.nih.gov/.

Clinical Resources (Publicly Available Information)

The Association of Occupational and Environmental Clinics (AOEC) has developed a network of clinics in the United States to provide expertise in occupational and environmental issues. Contact: AOEC, 1010 Vermont Avenue, NW, #513, Washington, DC 20005 • Phone: 202-347-4976 • FAX: 202-347-4950 • e-mail: AOEC@AOEC.ORG • Web Page: http://www.aoec.org/.

The American College of Occupational and Environmental Medicine (ACOEM) is an association of physicians and other health care providers specializing in the field of occupational and environmental medicine. Contact: ACOEM, 25 Northwest Point Boulevard, Suite 700, Elk Grove Village, IL 60007-1030 • Phone: 847-818-1800 • FAX: 847-818-9266 • Web Page: http://www.acoem.org/.

The American College of Medical Toxicology (ACMT) is a nonprofit association of physicians with recognized expertise in medical toxicology. Contact: ACMT, 10645 North Tatum Boulevard, Suite 200-111, Phoenix AZ 85028 • Phone: 844-226-8333 • FAX: 844-226-8333 • Web Page: http://www.acmt.net.

The Pediatric Environmental Health Specialty Units (PEHSUs) is an interconnected system of specialists who respond to questions from public health professionals, clinicians, policy makers, and the public about the impact of environmental factors on the health of children and reproductive-aged adults. Contact information for regional centers can be found at http://pehsu.net/findhelp.html.

The American Association of Poison Control Centers (AAPCC) provide support on the prevention and treatment of poison exposures. Contact: AAPCC, 515 King Street, Suite 510, Alexandria VA 22314 • Phone: 701-894-1858 • Poison Help Line: 1-800-222-1222 • Web Page: http://www.aapcc.org/

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APPENDIX F. GLOSSARY

Absorption—The process by which a substance crosses biological membranes and enters systemic circulation. Absorption can also refer to the taking up of liquids by solids, or of gases by solids or liquids.

Acute Exposure—Exposure to a chemical for a duration of ≤14 days, as specified in the Toxicological Profiles.

Adsorption—The adhesion in an extremely thin layer of molecules (as of gases, solutes, or liquids) to the surfaces of solid bodies or liquids with which they are in contact.

Adsorption Coefficient (Koc)—The ratio of the amount of a chemical adsorbed per unit weight of organic carbon in the soil or sediment to the concentration of the chemical in solution at equilibrium.

Adsorption Ratio (Kd)—The amount of a chemical adsorbed by sediment or soil (i.e., the solid phase) divided by the amount of chemical in the solution phase, which is in equilibrium with the solid phase, at a fixed solid/solution ratio. It is generally expressed in micrograms of chemical sorbed per gram of soil or sediment.

Benchmark Dose (BMD) or Benchmark Concentration (BMC)—is the dose/concentration corresponding to a specific response level estimate using a statistical dose-response model applied to either experimental toxicology or epidemiology data. For example, a BMD10 would be the dose corresponding to a 10% benchmark response (BMR). The BMD is determined by modeling the dose-response curve in the region of the dose-response relationship where biologically observable data are feasible. The BMDL or BMCL is the 95% lower confidence limit on the BMD or BMC.

Bioconcentration Factor (BCF)—The quotient of the concentration of a chemical in aquatic organisms at a specific time or during a discrete time period of exposure divided by the concentration in the surrounding water at the same time or during the same period.

Biomarkers—Indicators signaling events in biologic systems or samples, typically classified as markers of exposure, effect, and susceptibility.

Cancer Effect Level (CEL)—The lowest dose of a chemical in a study, or group of studies, that produces significant increases in the incidence of cancer (or tumors) between the exposed population and its appropriate control.

Carcinogen—A chemical capable of inducing cancer.

Case-Control Study—A type of epidemiological study that examines the relationship between a particular outcome (disease or condition) and a variety of potential causative agents (such as toxic chemicals). In a case-control study, a group of people with a specified and well-defined outcome is identified and compared to a similar group of people without the outcome.

Case Report—A report that describes a single individual with a particular disease or exposure. These reports may suggest some potential topics for scientific research, but are not actual research studies.

Case Series—Reports that describe the experience of a small number of individuals with the same disease or exposure. These reports may suggest potential topics for scientific research, but are not actual research studies.

Ceiling Value—A concentration that must not be exceeded.

Chronic Exposure—Exposure to a chemical for ≥365 days, as specified in the Toxicological Profiles.

Clastogen—A substance that causes breaks in chromosomes resulting in addition, deletion, or rearrangement of parts of the chromosome.

Cohort Study—A type of epidemiological study of a specific group or groups of people who have had a common insult (e.g., exposure to an agent suspected of causing disease or a common disease) and are followed forward from exposure to outcome, and who are disease-free at start of follow-up. Often, at least one exposed group is compared to one unexposed group, while in other cohorts, exposure is a continuous variable and analyses are directed towards analyzing an exposure-response coefficient.

Cross-sectional Study—A type of epidemiological study of a group or groups of people that examines the relationship between exposure and outcome to a chemical or to chemicals at a specific point in time.

Data Needs—Substance-specific informational needs that, if met, would reduce the uncertainties of human health risk assessment.

Developmental Toxicity—The occurrence of adverse effects on the developing organism that may result from exposure to a chemical prior to conception (either parent), during prenatal development, or postnatally to the time of sexual maturation. Adverse developmental effects may be detected at any point in the life span of the organism.

Dose-Response Relationship—The quantitative relationship between the amount of exposure to a toxicant and the incidence of the response or amount of the response.

Embryotoxicity and Fetotoxicity—Any toxic effect on the conceptus as a result of prenatal exposure to a chemical; the distinguishing feature between the two terms is the stage of development during which the effect occurs. Effects include malformations and variations, altered growth, and *in utero* death.

Epidemiology—The investigation of factors that determine the frequency and distribution of disease or other health-related conditions within a defined human population during a specified period.

Excretion—The process by which metabolic waste products are removed from the body.

Genotoxicity—A specific adverse effect on the genome of living cells that, upon the duplication of affected cells, can be expressed as a mutagenic, clastogenic, or carcinogenic event because of specific alteration of the molecular structure of the genome.

Half-life—A measure of rate for the time required to eliminate one-half of a quantity of a chemical from the body or environmental media.

Health Advisory—An estimate of acceptable drinking water levels for a chemical substance derived by EPA and based on health effects information. A health advisory is not a legally enforceable federal standard, but serves as technical guidance to assist federal, state, and local officials.

Immediately Dangerous to Life or Health (IDLH)—A condition that poses a threat of life or health, or conditions that pose an immediate threat of severe exposure to contaminants that are likely to have adverse cumulative or delayed effects on health.

Immunotoxicity—Adverse effect on the functioning of the immune system that may result from exposure to chemical substances.

Incidence—The ratio of new cases of individuals in a population who develop a specified condition to the total number of individuals in that population who could have developed that condition in a specified time period.

Intermediate Exposure—Exposure to a chemical for a duration of 15–364 days, as specified in the Toxicological Profiles.

***In Vitro*—**Isolated from the living organism and artificially maintained, as in a test tube.

***In Vivo*—**Occurring within the living organism.

Lethal Concentration(LO) (LCLO)—The lowest concentration of a chemical in air that has been reported to have caused death in humans or animals.

Lethal Concentration(50) (LC50)—A calculated concentration of a chemical in air to which exposure for a specific length of time is expected to cause death in 50% of a defined experimental animal population.

Lethal Dose(LO) (LDLO)—The lowest dose of a chemical introduced by a route other than inhalation that has been reported to have caused death in humans or animals.

Lethal Dose(50) (LD50)—The dose of a chemical that has been calculated to cause death in 50% of a defined experimental animal population.

Lethal Time(50) (LT50)—A calculated period of time within which a specific concentration of a chemical is expected to cause death in 50% of a defined experimental animal population.

Lowest-Observed-Adverse-Effect Level (LOAEL)—The lowest exposure level of chemical in a study, or group of studies, that produces statistically or biologically significant increases in frequency or severity of adverse effects between the exposed population and its appropriate control.

Lymphoreticular Effects—Represent morphological effects involving lymphatic tissues such as the lymph nodes, spleen, and thymus.

Malformations—Permanent structural changes that may adversely affect survival, development, or function.

Metabolism—Process in which chemical substances are biotransformed in the body that could result in less toxic and/or readily excreted compounds or produce a biologically active intermediate.

Minimal Risk Level (MRL)—An estimate of daily human exposure to a hazardous substance that is likely to be without an appreciable risk of adverse noncancer health effects over a specified route and duration of exposure.

Modifying Factor (MF)—A value (greater than zero) that is applied to the derivation of a Minimal Risk Level (MRL) to reflect additional concerns about the database that are not covered by the uncertainty factors. The default value for a MF is 1.

Morbidity—The state of being diseased; the morbidity rate is the incidence or prevalence of a disease in a specific population.

Mortality—Death; the mortality rate is a measure of the number of deaths in a population during a specified interval of time.

Mutagen—A substance that causes mutations, which are changes in the DNA sequence of a cell's DNA. Mutations can lead to birth defects, miscarriages, or cancer.

Necropsy—The gross examination of the organs and tissues of a dead body to determine the cause of death or pathological conditions.

Neurotoxicity—The occurrence of adverse effects on the nervous system following exposure to a hazardous substance.

No-Observed-Adverse-Effect Level (NOAEL)—The dose of a chemical at which there were no statistically or biologically significant increases in frequency or severity of adverse effects seen between the exposed population and its appropriate control. Although effects may be produced at this dose, they are not considered to be adverse.

Octanol-Water Partition Coefficient (Kow)—The equilibrium ratio of the concentrations of a chemical in *n*-octanol and water, in dilute solution.

Odds Ratio (OR)—A means of measuring the association between an exposure (such as toxic substances and a disease or condition) that represents the best estimate of relative risk (risk as a ratio of the incidence among subjects exposed to a particular risk factor divided by the incidence among subjects who were not exposed to the risk factor). An odds ratio that is greater than 1 is considered to indicate greater risk of disease in the exposed group compared to the unexposed group.

Permissible Exposure Limit (PEL)—An Occupational Safety and Health Administration (OSHA) regulatory limit on the amount or concentration of a substance not to be exceeded in workplace air averaged over any 8-hour work shift of a 40-hour workweek.

Pesticide—General classification of chemicals specifically developed and produced for use in the control of agricultural and public health pests (insects or other organisms harmful to cultivated plants or animals).

Pharmacokinetics—The dynamic behavior of a material in the body, used to predict the fate (disposition) of an exogenous substance in an organism. Utilizing computational techniques, it provides the means of studying the absorption, distribution, metabolism, and excretion of chemicals by the body.

Pharmacokinetic Model—A set of equations that can be used to describe the time course of a parent chemical or metabolite in an animal system. There are two types of pharmacokinetic models: data-based and physiologically-based. A data-based model divides the animal system into a series of compartments, which, in general, do not represent real, identifiable anatomic regions of the body, whereas the physiologically-based model compartments represent real anatomic regions of the body.

Physiologically Based Pharmacodynamic (PBPD) Model—A type of physiologically based dose-response model that quantitatively describes the relationship between target tissue dose and toxic endpoints. These models advance the importance of physiologically based models in that they clearly describe the biological effect (response) produced by the system following exposure to an exogenous substance.

Physiologically Based Pharmacokinetic (PBPK) Model—A type of physiologically based dose-response model that is comprised of a series of compartments representing organs or tissue groups with realistic weights and blood flows. These models require a variety of physiological information, including tissue volumes, blood flow rates to tissues, cardiac output, alveolar ventilation rates, and possibly membrane permeabilities. The models also utilize biochemical information, such as blood:air partition coefficients, and metabolic parameters. PBPK models are also called biologically based tissue dosimetry models.

Prevalence—The number of cases of a disease or condition in a population at one point in time.

Prospective Study—A type of cohort study in which a group is followed over time and the pertinent observations are made on events occurring after the start of the study.

Recommended Exposure Limit (REL)—A National Institute for Occupational Safety and Health (NIOSH) time-weighted average (TWA) concentration for up to a 10-hour workday during a 40-hour workweek.

Reference Concentration (RfC)—An estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious noncancer health effects during a lifetime. The inhalation RfC is expressed in units of mg/m^3 or ppm.

Reference Dose (RfD)—An estimate (with uncertainty spanning perhaps an order of magnitude) of the daily oral exposure of the human population to a potential hazard that is likely to be without risk of deleterious noncancer health effects during a lifetime. The oral RfD is expressed in units of mg/kg/day.

Reportable Quantity (RQ)—The quantity of a hazardous substance that is considered reportable under the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA). RQs are (1) ≥1 pound or (2) for selected substances, an amount established by regulation either under CERCLA or under Section 311 of the Clean Water Act. Quantities are measured over a 24-hour period.

Reproductive Toxicity—The occurrence of adverse effects on the reproductive system that may result from exposure to a hazardous substance. The toxicity may be directed to the reproductive organs and/or the related endocrine system. The manifestation of such toxicity may be noted as alterations in sexual behavior, fertility, pregnancy outcomes, or modifications in other functions that are dependent on the integrity of this system.

Retrospective Study—A type of cohort study based on a group of persons known to have been exposed at some time in the past. Data are collected from routinely recorded events, up to the time the study is undertaken. Retrospective studies are limited to causal factors that can be ascertained from existing records and/or examining survivors of the cohort.

Risk—The possibility or chance that some adverse effect will result from a given exposure to a hazardous substance.

Risk Factor—An aspect of personal behavior or lifestyle, an environmental exposure, existing health condition, or an inborn or inherited characteristic that is associated with an increased occurrence of disease or other health-related event or condition.

Risk Ratio/Relative Risk—The ratio of the risk among persons with specific risk factors compared to the risk among persons without risk factors. A risk ratio that is greater than 1 indicates greater risk of disease in the exposed group compared to the unexposed group.

Short-Term Exposure Limit (STEL)—A STEL is a 15-minute TWA exposure that should not be exceeded at any time during a workday.

Standardized Mortality Ratio (SMR)—A ratio of the observed number of deaths and the expected number of deaths in a specific standard population.

Target Organ Toxicity—This term covers a broad range of adverse effects on target organs or physiological systems (e.g., renal, cardiovascular) extending from those arising through a single limited exposure to those assumed over a lifetime of exposure to a chemical.

Teratogen—A chemical that causes structural defects that affect the development of an organism.

Threshold Limit Value (TLV)—An American Conference of Governmental Industrial Hygienists (ACGIH) concentration of a substance to which it is believed that nearly all workers may be repeatedly exposed, day after day, for a working lifetime without adverse effect. The TLV may be expressed as a Time-Weighted Average (TLV-TWA), as a Short-Term Exposure Limit (TLV-STEL), or as a ceiling limit (TLV-C).

Time-Weighted Average (TWA)—An average exposure within a given time period.

Toxicokinetic—The absorption, distribution, metabolism, and elimination of toxic compounds in the living organism.

Toxics Release Inventory (TRI)—The TRI is an EPA program that tracks toxic chemical releases and pollution prevention activities reported by industrial and federal facilities.

Uncertainty Factor (UF)—A factor used in operationally deriving the Minimal Risk Level (MRL), Reference Dose (RfD), or Reference Concentration (RfC) from experimental data. UFs are intended to account for (1) the variation in sensitivity among the members of the human population, (2) the uncertainty in extrapolating animal data to the case of human, (3) the uncertainty in extrapolating from data obtained in a study that is of less than lifetime exposure, and (4) the uncertainty in using lowest-observed-adverse-effect level (LOAEL) data rather than no-observed-adverse-effect level (NOAEL) data. A default for each individual UF is 10; if complete certainty in data exists, a value of 1 can be used; however, a reduced UF of 3 may be used on a case-by-case basis (3 being the approximate logarithmic average of 10 and 1).

Xenobiotic—Any substance that is foreign to the biological system.

APPENDIX G. ACRONYMS, ABBREVIATIONS, AND SYMBOLS

AAPCC	American Association of Poison Control Centers
ACGIH	American Conference of Governmental Industrial Hygienists
ACOEM	American College of Occupational and Environmental Medicine
ACMT	American College of Medical Toxicology
ADI	acceptable daily intake
ADME	absorption, distribution, metabolism, and excretion
AEGL	Acute Exposure Guideline Level
AIC	Akaike's information criterion
AIHA	American Industrial Hygiene Association
ALP	alkaline phosphatase
ALT	alanine aminotransferase
AOEC	Association of Occupational and Environmental Clinics
AST	aspartate aminotransferase
atm	atmosphere
ATSDR	Agency for Toxic Substances and Disease Registry
AWQC	Ambient Water Quality Criteria
BCF	bioconcentration factor
BMD/C	benchmark dose or benchmark concentration
BMDx	dose that produces a X% change in response rate of an adverse effect
BMDLx	95% lower confidence limit on the BMDx
BMDS	Benchmark Dose Software
BMR	benchmark response
BUN	blood urea nitrogen
C	centigrade
CAA	Clean Air Act
CAS	Chemical Abstract Services
CDC	Centers for Disease Control and Prevention
CEL	cancer effect level
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
CFR	Code of Federal Regulations
Ci	curie
CI	confidence interval
cm	centimeter
CPSC	Consumer Products Safety Commission
CuNP	Copper nanoparticles
CWA	Clean Water Act
DHHS	Department of Health and Human Services
DNA	deoxyribonucleic acid
DOD	Department of Defense
DOE	Department of Energy
DWEL	drinking water exposure level
EAFUS	Everything Added to Food in the United States
ECG/EKG	electrocardiogram
EEG	electroencephalogram
EPA	Environmental Protection Agency
ERPG	emergency response planning guidelines
F	Fahrenheit
F1	first-filial generation

FDA	Food and Drug Administration
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
FR	Federal Register
FSH	follicle stimulating hormone
g	gram
GC	gas chromatography
gd	gestational day
GFAP	glial fibrillary acidic protein
GGT	γ-glutamyl transferase
GRAS	generally recognized as safe
GSH	glutathione
HEC	human equivalent concentration
HED	human equivalent dose
HHS	Department of Health and Human Services
HPLC	high-performance liquid chromatography
HSDB	Hazardous Substance Data Bank
IARC	International Agency for Research on Cancer
ICC	Indian childhood cirrhosis
ICT	idiopathic copper toxicosis
IDLH	immediately dangerous to life and health
IRIS	Integrated Risk Information System
Kd	adsorption ratio
kg	kilogram
kkg	kilokilogram; 1 kilokilogram is equivalent to 1,000 kilograms and 1 metric ton
K_{oc}	organic carbon partition coefficient
K_{ow}	octanol-water partition coefficient
L	liter
LC	liquid chromatography
LC_{50}	lethal concentration, 50% kill
LC_{Lo}	lethal concentration, low
LD_{50}	lethal dose, 50% kill
LD_{Lo}	lethal dose, low
LDH	lactate dehydrogenase
LEC	Long-Evans Cinnamon
LH	luteinizing hormone
LOAEL	lowest-observed-adverse-effect level
LSE	Level of Significant Exposure
LT_{50}	lethal time, 50% kill
m	meter
mCi	millicurie
MCL	maximum contaminant level
MCLG	maximum contaminant level goal
MDA	malondialdehyde
MF	modifying factor
mg	milligram
mL	milliliter
mm	millimeter
mmHg	millimeters of mercury
mmol	millimole
MRL	Minimal Risk Level
MS	mass spectrometry

MSHA	Mine Safety and Health Administration
Mt	metric ton
NAAQS	National Ambient Air Quality Standard
NAS	National Academy of Science
NCEH	National Center for Environmental Health
ND	not detected
ng	nanogram
NHANES	National Health and Nutrition Examination Survey
NIEHS	National Institute of Environmental Health Sciences
NIOSH	National Institute for Occupational Safety and Health
NLM	National Library of Medicine
nm	nanometer
nmol	nanomole
NOAEL	no-observed-adverse-effect level
NPL	National Priorities List
NR	not reported
NRC	National Research Council
NS	not specified
NTP	National Toxicology Program
OR	odds ratio
OSHA	Occupational Safety and Health Administration
PAC	Protective Action Criteria
PAH	polycyclic aromatic hydrocarbon
PBPD	physiologically based pharmacodynamic
PBPK	physiologically based pharmacokinetic
PEHSU	Pediatric Environmental Health Specialty Unit
PEL	permissible exposure limit
PEL-C	permissible exposure limit-ceiling value
pg	picogram
PND	postnatal day
POD	point of departure
ppb	parts per billion
ppbv	parts per billion by volume
ppm	parts per million
ppt	parts per trillion
REL	recommended exposure level/limit
REL-C	recommended exposure level-ceiling value
RfC	reference concentration
RfD	reference dose
RNA	ribonucleic acid
ROS	reactive oxygen species
SARA	Superfund Amendments and Reauthorization Act
SCE	sister chromatid exchange
SD	standard deviation
SE	standard error
SGOT	serum glutamic oxaloacetic transaminase (same as aspartate aminotransferase or AST)
SGPT	serum glutamic pyruvic transaminase (same as alanine aminotransferase or ALT)
SIC	standard industrial classification
SMR	standardized mortality ratio
SOD	superoxide dismutase
sRBC	sheep red blood cell

STEL	short term exposure limit
TAC	total antioxidant capacity
TLV	threshold limit value
TLV-C	threshold limit value-ceiling value
TRI	Toxics Release Inventory
TSCA	Toxic Substances Control Act
TWA	time-weighted average
UF	uncertainty factor
U.S.	United States
USDA	United States Department of Agriculture
USGS	United States Geological Survey
USNRC	U.S. Nuclear Regulatory Commission
VOC	volatile organic compound
WBC	white blood cell
WHO	World Health Organization

>	greater than
≥	greater than or equal to
=	equal to
<	less than
≤	less than or equal to
%	percent
α	alpha
β	beta
γ	gamma
δ	delta
μm	micrometer
μg	microgram
q1*	cancer slope factor
–	negative
+	positive
(+)	weakly positive result
(–)	weakly negative result