Gastric Cancer (2013) 16:261–267
DOI 10.1007/s10120-012-0179-9

ORIGINAL ARTICLE

Prognosis of patients with advanced gastric cancer by HER2 status and trastuzumab treatment

Kohei Shitara · Yasushi Yatabe · Keitaro Matsuo · Masato Sugano · Chihiro Kondo · Daisuke Takahari · Takashi Ura · Masahiro Tajika · Seiji Ito · Kei Muro

Received: 18 May 2012 / Accepted: 28 June 2012 / Published online: 14 July 2012

Abstract
Background The purpose of this study was to evaluate the impact of human epidermal growth factor receptor 2 (HER2) status and trastuzumab treatment on the prognosis of patients with advanced gastric cancer (AGC).
Methods We retrospectively analyzed 364 AGC patients who received systemic chemotherapy. To evaluate the impact of trastuzumab exposure during any type of chemotherapy, our analysis used time-varying covariates to avoid a possible lead-time bias.
Results Among the 364 patients, 58 (15.9 %) were HER2-positive. The median overall survival of the HER2-positive patients treated with trastuzumab ($n = 43$) was significantly longer than that of the HER2-negative patients [$n = 306$; 24.7 vs. 13.9 months, with an adjusted hazard ratio (HR) of 0.58; 95 % confidence interval (CI), 0.36–0.95; $P = 0.03$]. Notably, 22 patients continued with trastuzumab beyond the date of progression. By contrast, the HER2-positive patients not treated with trastuzumab ($n = 15$) showed survival similar to that of the HER2-negative patients (13.5 vs. 13.9 months, with an adjusted HR of 1.04; 95 % CI, 0.52–2.11; $P = 0.91$). According to the multivariate analysis, exposure to trastuzumab was independently associated with a better prognosis (HR 0.56; 95 % CI; 0.33–0.93; $P = 0.026$).
Conclusions Recent HER2-positive AGC patients have a better prognosis than HER2-negative patients, particularly when treated with trastuzumab.

Keywords Chemotherapy · Gastric cancer · HER2 · Trastuzumab

K. Shitara · C. Kondo · D. Takahari · T. Ura · K. Muro
Department of Clinical Oncology, Aichi Cancer Center Hospital, Nagoya, Japan

K. Shitara (✉)
Department of Gastroenterology and Gastrointestinal Oncology, National Cancer Center Hospital East, 6-5-1, Kashiwanoha, Kashiwa, Chiba 277-8577, Japan
e-mail: kouheis0824@yahoo.co.jp

Y. Yatabe · M. Sugano
Department of Pathology and Molecular Diagnostics, Aichi Cancer Center Hospital, Nagoya, Japan

K. Matsuo
Division of Epidemiology and Prevention, Aichi Cancer Center Research Institute, Nagoya, Japan

M. Tajika
Department of Gastroenterology, Aichi Cancer Center Hospital, Nagoya, Japan

S. Ito
Department of Surgery, Aichi Cancer Center Hospital, Nagoya, Japan

Introduction

Gastric cancer is the fourth most common malignancy in the world (988,602 cases in 2008, 7.8 % of the total cancer cases) and the second leading cause of cancer deaths (737,419 deaths, 9.7 % of the total) [1]. Although the most effective treatment for localized disease is surgery, approximately half of all patients with advanced-stage disease experience a recurrence following a curative resection. The prognosis of patients with advanced or recurrent gastric cancer (AGC) remains poor, and commonly used combination chemotherapy regimens, consisting of a fluoropyrimidine plus a platinum agent with or without docetaxel or anthracyclines, lead to a median overall survival

(OS) of only 1 year [2–7]. Therefore, the development of novel anticancer agents or strategies for treating AGC is urgently required.

Human epidermal growth factor receptor 2 (HER2) is a growth factor receptor that is overexpressed in approximately 10–30 % of gastric cancers [8, 9]. The main function of HER2 is to mediate cell growth, differentiation, and survival [10, 11]. Therefore, HER2-positive tumors are expected to have more aggressive characteristics than HER2-negative tumors. Numerous studies have shown that HER2-positive breast cancer is associated with a poor prognosis when compared with HER2-negative breast cancer [11, 12]. Similarly, the prognostic value of HER2 status in gastric cancer has been evaluated in a number of reports. Although recent studies of gastric cancer have shown no impact of HER2 on survival [13, 14], the majority of the past studies have suggested that HER2-positive gastric cancer was associated with a poorer prognosis than HER2-negative gastric cancer [9, 15, 16].

Trastuzumab, a humanized monoclonal antibody that targets HER2, has recently been shown to improve the prognosis of HER2-positive AGC [17]. Combination chemotherapy consisting of 5-fluorouracil (5-FU) or capecitabine plus cisplatin with trastuzumab has shown a significantly higher response rate (47 vs. 35 %, $P = 0.0017$), a longer progression-free survival [PFS; 6.7 vs. 5.5 months; hazard ratio (HR) = 0.71, 95 % confidence interval (CI) 0.59–0.86], and a longer OS (13.8 vs. 11.1 months; HR = 0.74, 95 % CI 0.60–0.91) than chemotherapy alone [17]. This difference was more prominent in patients with immunohistochemical staining (IHC) of 3 + (IHC 3 +) or IHC 2 + plus positive gene amplification according to fluorescence in situ hybridization (FISH) (16.0 vs. 11.8 months; HR = 0.65, 95 % CI 0.51–0.83). The risk reduction achieved with trastuzumab is almost comparable to that found in a pivotal study of breast cancer [18]. According to these results, chemotherapy with trastuzumab is considered to be standard chemotherapy for HER2-positive AGC. However, the question of whether trastuzumab improves the prognosis of HER2-positive AGC patients to the levels observed in HER2-negative disease still remains. HER2-positive breast cancer patients who receive trastuzumab have been reported to have a better prognosis (HR of 0.56) than women with HER2-negative breast cancer [19].

The purpose of this study was to evaluate the impact of HER2 status and trastuzumab treatment on the prognosis of patients with AGC. We evaluated the impact of trastuzumab on OS using a time-varying covariate (TVC) analysis.

Patients and methods

Patients

This retrospective study was designed to compare the OS of HER2-positive and HER2-negative AGC patients who received systemic chemotherapy. In this analysis, HER2 positivity was defined as IHC 3 + or IHC 2 + plus FISH positivity, because these criteria were considered to be indications for using trastuzumab by a subset analysis of the ToGA trial [17, 20]. The following additional principal inclusion criteria were used: the presence of histologically proven, inoperable gastric cancer in patients who had received systemic chemotherapy; an Eastern Cooperative Oncology Group performance status (ECOG PS) of 0–2; and sufficient bone marrow, liver, and renal function.

Between April 2005 and August 2011, 807 consecutive patients with AGC received systemic chemotherapy at our institution, 650 of whom met the inclusion criteria. Among those 650 patients, 364 patients were evaluated for HER2 to screen them for the ToGA study or for trastuzumab treatment, and these 364 patients were analyzed in this study. Although trastuzumab had not yet been approved in Japan at the time of our study, using trastuzumab for HER2-positive patients was approved by our institution based on the results of the ToGA study. We note that fluoropyrimidine (5-FU or the oral fluoropyrimidine S-1), cisplatin, docetaxel, and paclitaxel were approved for use and were consistently available throughout this period. Written informed consent for the chemotherapy was obtained from all of the patients.

Statistical analysis

We compared the OS according to HER2 status and trastuzumab exposure by dividing the patients into three groups (HER2-negative vs. HER2-positive treated with trastuzumab vs. HER2-positive treated without trastuzumab). The OS was estimated from the date of systemic chemotherapy initiation to the date of death or last known date of survival. The vital status and disease status were confirmed by checking the medical records from the date of the last follow-up visit. In the cases that were lost to follow up, the vital status was confirmed by the census record, which is conducted annually at our institution. The median OS rate was estimated by the Kaplan–Meier method. The baseline patient characteristics and exposure to each class of chemotherapeutic agent, fluoropyrimidine (5-FU or oral fluoropyrimidine), platinum agents (cisplatin or oxaliplatin), taxanes (docetaxel or paclitaxel), and irinotecan and trastuzumab, were evaluated. Because other agents (mitomycin C or methotrexate) were also commonly used as

salvage therapy at our institution, we also evaluated exposure to these treatments. The disease progression associated with each line of chemotherapy was measured from the beginning of treatment to the date of disease progression, as evaluated by the attending physician.

The differences in the OS of each group were evaluated by univariate and multivariate analyses using a Cox proportional hazards model and presented as an HR and a 95 % CI. A subset analysis was stratified by the characteristics of the patients with significant differences in the three groups. We also evaluated the association between the exposure to each class of chemotherapeutic agent and the OS using multivariate analyses, because differences in exposure to agents other than trastuzumab may have contributed to survival differences. Because the length of exposure to each agent class varied over time (i.e., between first-, second-, and third-line treatments), the analyses may have been compromised by lead-time bias, which would result in false-positive or false-negative associations between a larger number of chemotherapeutic lines and longer survival. To minimize this potential bias, the exposure to each agent class was analyzed as a TVC. In addition, because disease progression was the primary reason for proceeding to the next line of chemotherapy, tumor progression during each line of chemotherapy was included in the TVCs. Each TVC was constructed as a step function initially set at 0 and increased by 1 U each time the corresponding event was observed. This method has been used in several reports that have evaluated the impact of drug exposure on survival [21–23].

Other variables considered in the multivariate analyses were ECOG PS (0 vs. $\geq$1), gender, histological type (diffuse vs. intestinal), age ($<$ 65 vs. $\geq$65 years), previous gastrectomy (no vs. yes), disease status (advanced vs. recurrent), prior adjuvant chemotherapy (no vs. yes), presence of liver metastasis (no vs. yes), presence of peritoneal metastasis (no vs. yes), number of metastatic sites (1 vs. $\geq$2), and first-line chemotherapy (monotherapy vs. combinations). The *P* values for testing differences in the baseline characteristics of each group were calculated with the χ^2 test for homogeneity or trend or with Fisher's exact test.

The statistical analyses were performed using STATA ver. 10 (Stata Corp LP, College Station, TX, USA). All of the tests were two-sided, and *P* values less than 0.05 were considered to be statistically significant.

Results

Patient characteristics

The patient characteristics according to HER2 status and trastuzumab use are shown in Table 1. Among the HER2-evaluated patients, 58 (15.9 %) were HER2-positive. HER2-positivity was more frequently associated with intestinal-type histology, liver metastasis, and lymph node metastasis. By contrast, peritoneal metastasis was more common in the HER2-negative patients than in the HER2-positive patients (Table 1). Among the HER2-positive patients, 43 received trastuzumab; 23 patients started it as first-line therapy (9 of these patients were included in the ToGA study) and 20 started it as second- or further-line therapy. Among the patients who were treated with trastuzumab who were not in the ToGA study, 22 patients continued with trastuzumab beyond the date of progression. The median duration of trastuzumab treatment in the 43 patients receiving this agent was 8.4 months (range 0.5 to 25 +), and 28 patients were still continuing with trastuzumab treatment at the time of this analysis. The frequency of exposure to platinum agents tended to be higher in HER2-positive disease, although no other significant differences were observed between the 3 groups (Table 1).

Survival and multivariate analysis

The median follow up at the time of the analysis was 38.9 months. The median OS of the HER2-positive patients ($n = 58$; 24.1 months; 95 % CI, 14.9–31.1 months) was significantly longer than that of the HER2-negative patients ($n = 304$; 13.9 months; 95 % CI, 12.7–16.1 months) by univariate analysis (HR 0.68; 95 % CI, 0.46–0.99; $P = 0.045$), although the difference was not significant in the multivariate analysis (HR 0.67; 95 % CI, 0.44–1.02; $P = 0.067$). The median OS of the HER2-positive patients treated with trastuzumab ($n = 43$; 24.7 months; 95 % CI, 15.6–35.1 months) was significantly longer than that of the HER2-negative patients (13.9 months), as verified by univariate analysis (HR 0.57; 95 % CI, 0.36–0.90; $P = 0.015$, Fig. 1) and multivariate analysis (adjusted HR of 0.58; 95 % CI, 0.36–0.95; $P = 0.03$. By contrast, the HER2-positive patients not treated with trastuzumab ($n = 15$) showed survival similar to that of the HER2-negative patients (13.5 months; 95 % CI, 3.6 to not reached), as demonstrated by univariate analysis (HR 1.11; 95 % CI, 0.58–2.08; $P = 0.76$) and multivariate analysis (HR 1.04; 95 % CI, 0.52–2.11; $P = 0.91$). The subset analysis according to the characteristics of selected patients demonstrated consistently higher rates of survival among HER2-positive patients treated with trastuzumab than among HER2-negative patients, with no significant heterogeneity (Table 2).

According to the multivariate TVC analysis, exposure to trastuzumab was independently associated with an improved prognosis (HR 0.56, 95 % CI, 0.33–0.93; $P = 0.026$, Table 3). Additionally, exposure to fluoropyrimidine, taxanes, and irinotecan was associated with a better prognosis, and exposure to platinum agents was

Table 1 Patient characteristics

Characteristics	HER2-positive patients treated with trastuzumab ($n = 43$, %)	HER2-positive patients not treated with trastuzumab ($n = 15$, %)	HER2-negative patients ($n = 306$, %)	*P* value
Age (years)				
Median (range)	63 (30–78)	68 (45–80)	64 (30–93)	0.33
Gender				0.45
Male	28 (65)	12 (80)	196 (64)	
Female	15 (35)	3 (20)	110 (36)	
ECOG PS				0.52
0	23 (53)	7 (47)	123 (40)	
1	17 (40)	6 (40)	149 (49)	
2	3 (7)	2 (13)	34 (11)	
Histological type				<0.001
Diffuse	12 (28)	4 (27)	217 (71)	
Intestinal	31 (72)	11 (73)	99 (29)	
Site of primary tumor				<0.001
EG junction	8 (19)	5 (33)	28 (9)	
Gastric	35 (81)	10 (67)	278 (91)	
Disease status				0.49
Advanced	33 (77)	9 (60)	203 (66)	
Recurrent	10 (23)	6 (40)	103 (34)	
Previous gastrectomy				0.59
No	26 (60)	8 (53)	160 (52)	
Yes	17 (40)	7 (47)	146 (48)	
Adjuvant chemotherapy				0.81
No	34 (79)	13 (87)	248 (81)	
Yes	9 (21)	2 (13)	58 (19)	
Site of metastasis				
Lymph node	27 (63)	12 (80)	145 (47)	0.01
Peritoneum	12 (28)	5 (33)	172 (56)	0.001
Liver	21 (49)	6 (40)	71 (23)	0.001
Number of metastatic organs				0.11
1	15 (35)	8 (53)	174 (57)	
2 or more	28 (65)	7 (47)	132 (43)	
First-line CTx				0.03
Combination	35 (81)	12 (80)	192 (63)	
Monotherapy	8 (19)	3 (20)	114 (37)	
Initiation of CTx				0.1
2005–2007	10 (23)	8 (53)	96 (31)	
2008–2011	33 (77)	7 (47)	210 (69)	
Exposure to agents				
Fluoropyrimidine	38 (88)	13 (87)	292 (95)	0.08
Platinum	42 (98)	11 (73)	228 (75)	0.003
Taxane	33 (77)	12 (80)	206 (67)	0.29
Irinotecan	22 (51)	6 (40)	137 (45)	0.67
Others	5 (12)	1 (7)	49 (16)	0.49

Underlined *P* values are significant

HER2 human epidermal growth factor receptor 2, *PS* performance status, *ECOG* Eastern Cooperative Oncology Group, *CTx* chemotherapy, *EG* esophagogastric

associated with improved survival, although the significance was borderline. By contrast, the other agents had no impact on survival.

Discussion

In this study, we compared the OS in AGC patients according to HER2 status and exposure to trastuzumab. The OS of the HER2-positive patients who were treated with trastuzumab was significantly longer than that of the HER2-negative patients, with an adjusted HR of 0.58. This result is similar to those observed in a large study of patients with breast cancer in which a similar degree of risk reduction was found [18]. In addition, the multivariate TVC analysis in our study showed that trastuzumab had a significant impact on OS. These results confirmed that trastuzumab therapy improves the prognosis of HER2-positive AGC patients beyond that of patients with HER2-negative AGC.

In this study, the HER2-positive patients not treated with trastuzumab showed survival rates similar to those of the HER2-negative patients. By contrast, the majority of previous studies have found poorer survival among patients with HER2-positive gastric cancer than among patients with HER2-negative tumors [9, 15, 16]. Most of these studies have been based on retrospective analyses in a single institution that combined patients with advanced-stage cancer and those with resected gastric cancer. Additionally, until the ToGA study, various definitions were used to evaluate HER2 status. Recently, Janjigian et al. [14] reported the results of a retrospective analysis of HER2 status among 381 patients who had enrolled in several clinical AGC studies. The HER2-positive patients (IHC 3 + or FISH +; $n = 78$) tended to have a better prognosis than the HER2-negative patients (13.9 vs. 11.4 months), although a multivariate analysis showed no impact of HER2 status on survival [14]. Additionally, an exploratory analysis of HER2 status in the AVAGAST study showed similar median OS in the HER2-negative and HER2-positive patients (10.5 vs. 9.8 months) who received capecitabine or 5-FU plus cisplatin [24]. Combined with our results, these findings suggest that HER2 status may

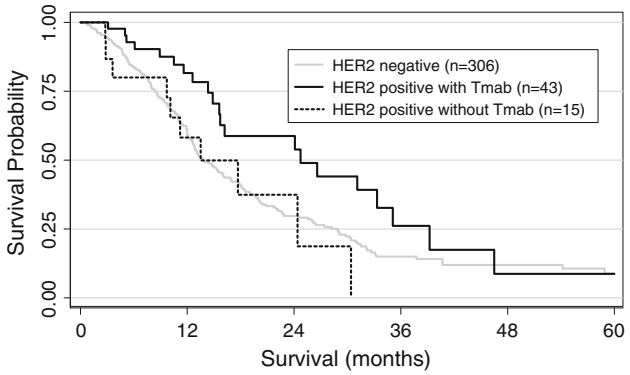

Fig. 1 The median OS of the HER2-positive patients treated with trastuzumab ($n = 43$; 24.7 months; 95 % CI, 15.6 to 35.1 months) was significantly longer than that of the HER2-negative patients $n = 306$; (13.9 months), as verified by univariate analysis (HR 0.57; 95 % CI, 0.36 to 0.90; $P = 0.015$) and multivariate analysis (adjusted HR of 0.58; 95 % CI, 0.36 to 0.95; $P = 0.03$). The HER2-positive patients not treated with trastuzumab ($n = 15$) showed survival similar to the HER2-negative patients (13.5 months; 95 % CI, 3.6 to not reached), as demonstrated by univariate analysis (HR 1.11; 95 % CI, 0.58 to 2.08; $P = 0.76$) and multivariate analysis (HR 1.04; 95 % CI, 0.52 to 2.11; $P = 0.91$)

Table 2 Impact of HER2 and trastuzumab use according to selected backgrounds

	HER2-negative	HER2-positive	
		Treated with trastuzumab HR (95 % CI)	Not treated with trastuzumab HR (95 % CI)
Histology			
Diffuse	1 (reference)	0.34 (0.12–0.99)	2.48 (0.67–8.90)
Intestinal	1 (reference)	0.53 (0.28–1.03)	0.90 (0.35–2.28)
p-heterogeneity		0.48	0.22
Site of metastasis			
Liver	1 (reference)	0.65 (0.28–1.51)	1.09 (0.24–4.74)
Lymph node	1 (reference)	0.68 (0.34–1.36)	0.78 (0.32–1.78)
Peritoneum	1 (reference)	0.76 (0.32–1.83)	1.00 (0.28–3.57)
p-heterogeneity		0.97	0.91
Initiation of chemotherapy			
Before 2008	1 (reference)	0.65 (0.27–1.59)	1.34 (0.56–3.24)
After 2008	1 (reference)	0.51 (0.25–1.06)	1.95 (0.25–15.27)
p-heterogeneity		0.69	0.87

Except for the stratifying factors, the HRs were adjusted for age, gender, ECOG PS, disease status, prior gastrectomy, adjuvant chemotherapy, and the number of metastatic sites

HR hazard ratio, *CI* confidence interval

Table 3 Results of the multivariate TVC analyses

Variable	Multivariate analysis with TVC[a]		
	HR	95 % CI	*P*
Exposure to agents			
Trastuzumab	0.56	0.33–0.93	0.026
Fluoropyrimidine	0.40	0.19–0.84	0.015
Platinum	0.63	0.38–1.03	0.06
Taxanes	0.37	0.25–0.57	<0.001
Irinotecan	0.47	0.33–0.67	<0.001
Other	0.85	0.55–1.38	0.39

Underlined *P* values are significant

TVC time-varying covariate, *HR* hazard ratio, *CI* confidence interval

[a] Adjusted by age, gender, ECOG PS, disease status, histology, prior gastrectomy, adjuvant chemotherapy, presence of liver metastasis, presence of peritoneal metastasis, number of metastatic sites, time of initiation of chemotherapy, and tumor progression during each line of chemotherapy

have a small impact on survival in AGC. In addition, recent data from a large randomized study of resected gastric cancer found that HER2 status was not associated with the OS or with relapse-free survival in either the group who received surgery only or the group who also received adjuvant chemotherapy [13].

Interestingly, the risk reduction or median survival with trastuzumab treatment in our analysis was relatively better than the results of a subset analysis among those patients with IHC 3 + or IHC2 + plus FISH + in the ToGA study [17]. Several possibilities may explain this difference. First, the median exposure to trastuzumab in the ToGA study was reported to be 4.9 months [17]. Although the reasons for discontinuing the treatment were not reported in detail, continuing with trastuzumab beyond progression was not allowed in the ToGA study. By contrast, the median duration of exposure to trastuzumab in our study was longer (8.4 months). A number of patients who were treated outside of the ToGA study continued with trastuzumab beyond progression because this strategy has been found to contribute to improved treatment outcomes in randomized studies of breast cancer patients [25, 26]. A prospective study that evaluates the importance of trastuzumab beyond progression is warranted, as is further evaluation of AGC patients in a randomized study. Second, owing to the retrospective nature of the present study, unblinded bias may have overestimated the treatment effect. Regardless, our results reconfirmed the results of the ToGA study, which demonstrated that a new effective agent with the right target improved the prognosis of gastric cancer. Additionally, other agents targeting HER2 (such as lapatinib, which has already been shown to be effective in HER2-positive breast cancer) are under investigation in AGC, and the results are awaited.

It is important to note the methodological limitations of the present study. First, not all of the patients in this study period were evaluated for their HER2 status. Thus, the analysis may have been subject to some selection bias, although no significant differences other than PS were observed between the patients in the analysis and the patients not evaluated for HER2 status during the same period (data not shown). The median OS of the patients not evaluated for HER2 was 13.0 months, which was similar to that of the HER2-negative patients or HER2-positive patients not treated with trastuzumab in this analysis. Second, the study was a retrospective, non-randomized comparison of trastuzumab treatment in patients evaluated for HER2 status. Therefore, the differing characteristics of the patients and the differing treatments (other than with trastuzumab) in each group may have affected the results. Because the use of trastuzumab and the exposure to platinum agents differed between the groups, we used the TVC analysis to adjust the exposure to agents and comprehensively evaluate the impact of each agent class, regardless of the treatment line. However, TVC analysis is not necessarily adequate under the conditions of our study because its validity may depend on the assumption of a strong association between treatment selection at the time of the events and the history leading up to the events [27]. Other potential confounders, such as PS and the metastatic site, were also considered in the multivariate analyses; owing to the retrospective, non-randomized nature of the study, however, residual confounding effects caused by non-included factors cannot be completely ruled out. Nevertheless, because our survival results for the HER2-negative patients and the HER2-positive patients not treated with trastuzumab were quite similar to those of the ToGA trial and the results of other randomized controlled trials, the effect of this selection bias may have been small. Third, the small sample size and single-center population are other major limitations of this study.

In conclusion, we found that introducing trastuzumab improved the prognosis of HER2-positive AGC patients beyond that of HER2-negative patients. Because other targeted therapies are currently under study, more improvement in the prognosis of HER2-positive AGC patients is expected. Importantly, because HER2-positive patients are a minority of AGC patients (with a prevalence of less than 20 %), further investigation of other key AGC markers is warranted.

Conflict of interest None of the authors have financial or personal conflicts of interest to disclose.

References

1. International Agency for Research on Cancer. GLOBOCAN 2008: http://www-dep.iarc.fr/CancerMondial.htm.
2. Van Cutsem E, Moiseyenko VM, Tjulandin S, Majlis A, Constenla M, Boni C, et al. Phase III study of docetaxel and cisplatin

plus fluorouracil compared with cisplatin and fluorouracil as first-line therapy for advanced gastric cancer: a report of the V325 Study Group. J Clin Oncol. 2006;24:4991–7.
3. Cunningham D, Starling N, Rao S, Iveson T, Nicolson M, Coxon F, et al. Capecitabine and oxaliplatin for advanced esophagogastric cancer. N Engl J Med. 2008;358:36–46.
4. Koizumi W, Narahara H, Hara T, Takagane A, Akiya T, Takagi M, et al. S-1 plus cisplatin versus S-1 alone for first-line treatment of advanced gastric cancer (SPIRITS trial): a phase III trial. Lancet Oncol. 2008;9:215–21.
5. Kang YK, Kang WK, Shin DB, Chen J, Xiong J, Wang J, et al. Capecitabine/cisplatin versus 5-fluorouracil/cisplatin as first-line therapy in patients with advanced gastric cancer: a randomised phase III noninferiority trial. Ann Oncol. 2009;20:666–73.
6. Ajani JA, Rodriguez W, Bodoky G, Moiseyenko V, Lichinitser M, Gorbunova V, et al. Multicenter phase III comparison of cisplatin/S-1 with cisplatin/infusional fluorouracil in advanced gastric or gastroesophageal adenocarcinoma study: the FLAGS trial. J Clin Oncol. 2010;28:1547–53.
7. Ohtsu A, Shah MA, Van Cutsem E, Rha SY, Sawaki A, Park SR, et al. Bevacizumab in combination with chemotherapy as first-line therapy in advanced gastric cancer: a randomized, double-blind, placebo-controlled phase III study. J Clin Oncol. 2011;29:3968–76.
8. Hofmann M, Stoss O, Shi D, Büttner R, van de Vijver M, Kim W, et al. Assessment of a HER2 scoring system for gastric cancer: results from a validation study. Histopathology. 2008;52:797–805.
9. Gravalos C, Jimeno A. HER2 in gastric cancer: a new prognostic factor and a novel therapeutic target. Ann Oncol. 2008;19:1523–9.
10. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. Science. 1987;235:177–82.
11. Slamon DJ, Godolphin W, Jones LA, Holt JA, Wong SG, Keith DE, et al. Studies of the HER-2/neu proto-oncogene in human breast and ovarian cancer. Science. 1989;244:707–12.
12. Seshadri R, Firgaira FA, Horsfall DJ, McCaul K, Setlur V, Kitchen P. Clinical significance of HER-2/neu oncogene amplification in primary breast cancer: the South Australian Breast Cancer Study Group. J Clin Oncol. 1993;11:1936–42.
13. Terashima M, Ochiai A, Kitada K, Ichikawa W, Kurahashi I, Sakuramoto S, et al. Impact of human epidermal growth factor receptor (EGFR) and ERBB2 (HER2) expressions on survival in patients with stage II/III gastric cancer, enrolled in the ACTS-GC study. J Clin Oncol. 2011;29 (suppl; abstr 4013).
14. Janjigian YY, Werner D, Pauligk C, Steinmetz K, Kelsen DP, Jäger E, et al. Prognosis of metastatic gastric and gastroesophageal junction cancer by HER2 status: a European and USA International collaborative analysis. Ann Oncol. 2012 [Epub ahead of print].
15. Begnami MD, Fukuda E, Fregnani JH, Nonogaki S, Montagnini AL, Da Costa WL Jr, et al. Prognostic implications of altered human epidermal growth factor receptors (HERs) in gastric carcinomas: HER2 and HER3 are predictors of poor outcome. J Clin Oncol. 2011;29:3030–6.
16. Kim KC, Koh YW, Chang HM, Kim TH, Yook JH, Kim BS, et al. Evaluation of HER2 protein expression in gastric carcinomas: comparative analysis of 1,414 cases of whole-tissue sections and 595 cases of tissue microarrays. Ann Surg Oncol. 2011;18:2833–40.
17. Bang YJ, Van Cutsem E, Feyereislova A, Chung HC, Shen L, Sawaki A, et al. Trastuzumab in combination with chemotherapy versus chemotherapy alone for treatment of HER2-positive advanced gastric or gastro-oesophageal junction cancer (ToGA): a phase 3, open-label, randomised controlled trial. Lancet. 2010;376:687–97.
18. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. N Engl J Med. 2001;344:783–92.
19. Dawood S, Broglio K, Buzdar AU, Hortobagyi GN, Giordano SH. Prognosis of women with metastatic breast cancer by HER2 status and trastuzumab treatment: an institutional-based review. J Clin Oncol. 2010;28:92–8.
20. National Comprehensive Cancer Network: NCCN clinical practice guidelines in Oncology, Gastric Cancer V.2.2011. http://www.nccn.org/professionals/physician_gls/f_guidelines.asp.
21. de Gramont A, Buyse M, Abrahantes JC, Burzykowski T, Quinaux E, Cervantes A, et al. Reintroduction of oxaliplatin is associated with improved survival in advanced colorectal cancer. J Clin Oncol. 2007;25:3224–9.
22. Grothey A, Sugrue MM, Purdie DM, Dong W, Sargent D, Hedrick E, et al. Bevacizumab beyond first progression is associated with prolonged overall survival in metastatic colorectal cancer: results from a large observational cohort study (BRiTE). J Clin Oncol. 2008;26:5326–34.
23. Shitara K, Matsuo K, Mizota A, Kondo C, Nomura M, Takahari D, et al. Association of fluoropyrimidines, platinum agents, taxanes, and irinotecan in any line of chemotherapy with survival in patients with advanced gastric cancer. Gastric Cancer. 2011;14:155–60.
24. Shah MA, Kang YK, Ohtsu A, Delmar P, Foernzler D, Langer B, et al. Tumour biomarker analyses in the AVAGAST phase III randomized study of first-line bevacizumab + capecitabine/cisplatin in patients with advanced gastric cancer. Ann Oncol. 2010;21 suppl 8:viii67–viii68 (abstr 174PD).
25. von Minckwitz G, du Bois A, Schmidt M, Maass N, Cufer T, de Jongh FE, et al. Trastuzumab beyond progression in human epidermal growth factor receptor 2-positive advanced breast cancer: a German Breast Group 26/Breast International Group 03–05 study. J Clin Oncol. 2009;27:1999–2006.
26. Blackwell KL, Burstein HJ, Storniolo AM, Rugo H, Sledge G, Koehler M, et al. Randomized study of Lapatinib alone or in combination with trastuzumab in women with ErbB2-positive, trastuzumab-refractory metastatic breast cancer. J Clin Oncol. 2010;28:1124–30.
27. Hernán MA, Brumback B, Robins JM. Marginal structural models to estimate the joint effect of non-randomized treatments. J Am Stat Assoc. 2001;96:440–8.