Effectiveness of Conservation Targets in Capturing Genetic Diversity

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Abstract: *Any conservation actions that preserve some populations and not others will have genetic consequences. We used empirical data from four rare plant taxa to assess these consequences in terms of how well allele numbers (all alleles and alleles occurring at a frequency of >0.05 in any population) and expected heterozygosity are represented when different numbers of populations are conserved. We determined sampling distributions for these three measures of genetic diversity using Monte Carlo methods. We assessed the proportion of alleles included in the number of populations considered adequate for conservation, needed to capture all alleles, and needed to meet an accepted standard of genetic-diversity conservation of having a 90–95% probability of including all common alleles. We also assessed the number of populations necessary to obtain values of heterozygosity within ±10% of the value obtained from all populations. Numbers of alleles were strongly affected by the number of populations sampled. Heterozygosity was only slightly less sensitive to numbers of populations than were alleles. On average, currently advocated conservation intensities represented 67–83% of all alleles and 85–93% of common alleles. The smallest number of populations to include all alleles ranged from 6 to 17 (42–57%), but <0.2% of 1000 samples of these numbers of populations included them all. It was necessary to conserve 16–29 (53–93%) of the sampled populations to meet the standard for common alleles. Between 20% and 64% of populations were needed to reliably represent species-level heterozygosity. Thus, higher percentages of populations are needed than are currently considered adequate to conserve genetic diversity if populations are selected without genetic data.*

Efectividad de los Objetivos de Conservación en la Captura de Diversidad Genética

Resumen: *Cualquier acción de conservación que preserve algunas poblaciones y no otras tendrá consecuencias genéticas. Utilizamos datos empíricos de cuatro taxones de plantas raras para evaluar estas consecuencias en términos de lo bien representados que están los números de alelos (todos los alelos ocurriendo a una frecuencia >0.05 en cualquier población) y la heterocigosidad esperada cuando se conservan diferentes números de poblaciones. Las distribuciones de muestreo de estas tres medidas de la diversidad genética fueron determinadas utilizando métodos Monte Carlo. Evaluamos la proporción de alelos incluida en números de poblaciones: consideradas adecuadas para la conservación; requeridas para capturar todos los alelos; y las requeridas para alcanzar un estándar de conservación de diversidad genética aceptable del 90–95% de probabilidad de incluir todos los alelos comunes. También evaluamos el número de poblaciones necesarias para obtener valores de heterocigosidad que caigan dentro de ±10% del valor obtenido de todas las poblaciones. Los números de alelos fueron afectados significativamente por el número de poblaciones muestreadas. La heterocigosidad solo fue ligeramente menos sensible a los números de poblaciones de lo que fueron los alelos. Las intensidades de conservación propugnadas actualmente representaron en promedio el 67–83% de todos los alelos y el 85–93% de los alelos comunes. El menor número de poblaciones para incluir a todos los alelos varió de 6 a 17 (42–57%), pero <0.2% de 1000 muestras de esos números de poblaciones los incluyó a todos. Fue necesario conservar de 16 a 29 (53–93%) de las poblaciones muestreadas para alcanzar el estándar para los alelos comunes. Se requirió entre 20% y 64% de las poblaciones para representar*

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la heterocigosidad a nivel de especie confiablemente. Por lo tanto, se requieren mayores porcentajes de poblaciones que los actualmente considerados adecuados para conservar la diversidad genética si las poblaciones son seleccionadas sin datos genéticos.

Introduction

A major conservation objective is to maintain biological diversity, including individual species of concern, general species diversity, and community diversity. Because habitat destruction and degradation are leading causes of the loss of diversity (e.g., Wilcove et al. 1998), attention has long focused on establishing reserves and reserve networks to slow rates of habitat loss and fragmentation (e.g., Soulé & Simberloff 1986; Simberloff 1988; Margules & Pressey 2000). Because of the economic and social costs of dedicating land to conservation purposes, much effort has been spent developing methods to determine the smallest number of sites needed to represent diversity (e.g., Margules et al. 1994; Csuti et al. 1997; Margules & Pressey 2000). When conservation efforts focus on individual rare species, one concern is to represent within-species genetic diversity, because its loss is thought to increase extinction risk and decrease the potential for future adaptation (Ellstrand & Elam 1993). Of course, maintaining populations of sufficient size and in particular configurations is also necessary to have an acceptably low probability of extinction (Schemske et al. 1994; Burgman et al. 2001). Typically, population sizes that are ecologically secure are also genetically secure (Lande 1988); as a result, reserve-selection decisions are most often not made on the basis of genetic diversity. Rather, they are based on ecological characteristics (e.g., Simberloff 1988; Prober 1996), demographic characteristics (e.g., Schemske et al. 1994; Burgman et al. 2001), or practical considerations including land availability (Prendergast et al. 1999). Although active management of genetic diversity is not necessary for the majority of rare species (Holsinger & Gottlieb 1991), it is important to represent within-species genetic diversity in reserve networks.

The purpose of our research was to assess how well genetic diversity, in terms of number of alleles and expected heterozygosity, is represented when different numbers of populations are conserved. We made this assessment using Monte Carlo methods to sample empirical allozyme data from four rare plant species. Our approach provides insight into the amount of diversity likely to be represented when conservation decisions are made in the absence of data on genetic diversity. Such an assessment is important for estimating how well genetic diversity will be maintained in the face of pressure to minimize the land area dedicated to conservation and when conservation decisions are made without detailed information on every species. Although it may be feasible to collect data on genetic diversity for a limited number of species of interest, the task becomes impractical for large-scale planning efforts involving hundreds of species. Further, because ecological factors tend to pose more immediate risks to species, funding is often focused on those factors rather than on genetic-diversity surveys.

Conservation goals for genetic diversity include maintaining variation affecting the current fitness of individuals, providing variation for adaptation to future environmental change, and providing for ongoing processes such as gene flow and natural selection while minimizing genetic drift (Namkoong 1993; Storfer 1996). We focus specifically on how well genetic diversity will be represented in reserves, rather than on maintenance of that diversity over time. Although both are important, if diversity is not included in the first place, maintaining it over time becomes moot. We used allozyme data rather than focusing directly on adaptive variation. Although adaptive traits are of primary interest, they are difficult to measure directly and data are not available for the majority of rare taxa. In lieu of such information, conservation of allele numbers and levels of heterozygosity, as detected by marker loci, is advocated and commonly practiced (Marshall 1989; Brown & Briggs 1991; Ceska et al. 1997; Petit et al. 1998). Maintaining identifiable marker alleles is considered a surrogate for indicating levels of variation throughout the genome (Marshall 1989; Namkoong 1993; Petit et al. 1998). For example, Schoen and Brown (1993) and Bataillon et al. (1996) demonstrated through simulations that marker diversity was indicative of diversity at nontargeted loci.

Genetic Diversity Standards

It is important to conserve both numbers of alleles and expected heterozygosity (interpreted in the context of gene diversity). In general, initial allelic composition determines the limit of response to selection over generations, whereas an immediate selection response is related to expected heterozygosity (Petit et al. 1998). Number of alleles has been advocated as a more appropriate measure than heterozygosity because it is more sensitive to differences in population size and number of populations and thus will be affected first as populations decline in size or as whole populations are extirpated (Allendorf 1986). In contrast, expected heterozygosity is influenced primarily by alleles of intermediate or higher

frequency and thus is relatively insensitive to changes in the frequency of rare alleles that are most likely to be lost from populations via drift or from a species as populations are extirpated. Thus, heterozygosity, although an important measure of overall genetic diversity, is considered more easily represented.

Regardless of the measure of diversity chosen, there is little specific guidance as to how much genetic diversity is enough to conserve. In terms of numbers of alleles, Petit et al. (1998) recommend conserving as many marker alleles as possible, regardless of their frequency. Marker alleles serve as correlates of diversity at loci directly affecting adaptive traits and account for the potential importance of low-frequency alleles in some contexts, including self-incompatibility, disease resistance, or adaptation to local environmental conditions. Because electrophoretically detectable allozyme alleles occurring below a frequency of 0.05 contribute little to heterozygosity and are typically lost in relatively few generations, Marshall and Brown (1975) consider them to be evolutionarily insignificant. For ex situ conservation, they argue for a standard of genetic-diversity conservation of a 90–95% probability of including all alleles occurring at a frequency of >0.05 in at least one population of a species (hereafter called common alleles) (Marshall & Brown 1975). The Center for Plant Conservation (1991) has adopted this standard for their ex situ collection program, and we used it as a benchmark.

Because expected heterozygosity is important for maintaining the potential for immediate adaptive response, the conservation objective for heterozygosity, in terms of the number of populations to include in reserve networks, is to maintain or increase observed levels. In the absence of knowledge of levels of heterozygosity, it is reasonable to seek to maintain existing levels with a certain level of confidence. Because no such level has been suggested previously, we adopted a standard of obtaining a mean value of H_e within $\pm 10\%$ of the mean value realized when all populations are included.

Conservation Targets

The number of populations necessary to conserve genetic diversity within a species depends on the measure of diversity one chooses, how that diversity is partitioned within and among populations, and how much of the total diversity one considers sufficient. In contrast to the extensive literature on the number of individuals needed to maintain genetic diversity within populations (e.g., Franklin 1980; Soulé 1980; Lande & Barrowclough 1987; Lande 1988), there has been little work on how many populations are needed to represent species-level diversity. Brown and Briggs (1991) determined that sampling from five populations of a rare species would be sufficient to have a 90–95% probability of capturing all common alleles for ex situ conservation, the standard suggested by Marshall and Brown (1975). The five-population target is based on typical allele-distribution patterns at a single locus and the fact that most plants for which ex situ germplasm collections are made occur in 20 or fewer populations (Brown & Briggs 1991). Falk (1991) argues that five populations are also sufficient for rare species having a larger number of populations and that the cost of additional sampling beyond five populations is too high for the marginal gain in new alleles. The Center for Plant Conservation (1991) has adopted the Marshall and Brown (1975) standard of genetic-diversity conservation and the Brown and Briggs (1991) five-population target for ex situ germplasm conservation collections. No targets for population number have been established specifically for heterozygosity.

Similarly, no targets for population number have been developed specifically for in situ genetic-diversity conservation, but if sampling five populations is sufficient to meet the standard for ex situ conservation, protecting five populations in the wild should also suffice. A number of general in situ conservation targets have been advocated. For example, several international organizations recommend protection of 10–12% of national or ecosystem land area (Noss 1996; Soulé & Sanjayan 1998). Although this target was developed for species diversity, it has been applied to conserving 10–12% of populations of rare plant species in an area (Duffy et al. 1999). Kiester et al. (1996) consider three populations, occupying at least 10,000 ha, sufficient to represent mid-size mammalian predators in Idaho. Additionally, methods to select reserves that represent all desired elements of biodiversity in the smallest area or number of sites (minimum-set or complementarity approaches) most often include only one occurrence of individual rare species (e.g., Csuti et al. 1997; Margules & Pressey 2000). Together, the five-population target, the target of conserving 10–12% of populations, and the minimum-set reserve-design approach represent a range of conservation intensities.

Methods

We used data from up to 30 individuals per population sampled from between 14 and 31 populations of each of four federally listed plant taxa: *Astragalus albens* E. Greene (Fabaceae), *Erigeron parishii* A. Gray (Asteraceae), *Eriogonum ovalifolium* var. *vineum* (Small) Jepson (Polygonaceae), and *Oxytheca parishii* var. *goodmaniana* B. Ertter (Polygonaceae). Sampling represented the ecological and geographic ranges of each taxon but did not include all populations (Neel 2000; Neel & Ellstrand 2001) (Table 1).

The four taxa are globally rare, completely or nearly endemic to restricted areas of limestone and dolomite substrates that occupy approximately 13,200 ha in the

Table 1. Genetic diversity characteristics of four federally listed plant taxa (Neel 2000; Neel & Ellstrand 2001).*

Taxon	Populations	Individuals	Loci	Total alleles (private alleles)		Gene diversity, H_e (SE)		Population differentiation, θ_P (95% CI)	
				all alleles	common alleles	all alleles	common alleles	all alleles	common alleles
Astragalus albens	30	879	12	69 (14)	36	0.14 (0.02)	0.12 (0.02)	0.01 (0.008–0.016)	0.01 (0.021–0.012)
Eriogonum ovalifolium var. *vineum*	31	929	11	60 (9)	48	0.17 (0.03)	0.17 (0.03)	0.07 (0.026–0.086)	0.07 (0.028–0.087)
Erigeron parishii	31	932	14	60 (9)	49	0.19 (0.03)	0.19 (0.03)	0.12 (0.089–0.155)	0.12 (0.090–0.157)
Oxytheca parishii var. *goodmaniana*	14	390	12	41 (9)	31	0.13 (0.04)	0.12 (0.04)	0.22 (0.036–0.460)	0.20 (0.040–0.447)

*Common alleles are those alleles that occur at >0.05 frequency in at least one population.

northeastern San Bernardino Mountains of southern California (U.S.A.), and threatened throughout most or all of their ranges, primarily by limestone mining operations (U. S. Fish and Wildlife Service 1994). One taxon, *Erigeron parishii*, is primarily restricted to limestone and dolomite substrates in this range but has one extant cluster of occurrences on quartz monzonite in the same mountain range and one historic occurrence on quartz monzonite in the nearby Little San Bernardino Mountains. As of July 1999, known populations of each of these taxa occupied the following areas: *A. albens,* 537 ha; *E. parishii*, 453 ha; *E. ovalifolium* var. *vineum*, 547 ha; and *O. parishii* var. *goodmaniana*, 221 ha (S. Redar, personal communication). Conservation planning efforts for these taxa are in progress and are intended to establish some protected areas and allow other areas to be mined.

We used the computer program GDA (Lewis and Zaykin 2001) to calculate total number of alleles, total number of common alleles (alleles occurring at a frequency of >0.05 in at least one population), and expected heterozygosity (H_e). Population genetic structure (Wright 1965) was described by θ_P ($\approx F_{ST}$), following methods of Weir (1996), as implemented in GDA (Lewis & Zaykin 2001). As an F_{ST} statistic analog, θ_P estimates the correlation among uniting gametes within populations relative to the whole species sample and thus estimates differentiation among populations. The 95% confidence intervals for θ_P were calculated from 1000 bootstrap replicates across loci. Estimates of population genetic parameters are summarized in Table 1, and all except number of common alleles are described in detail elsewhere (Neel 2000; Neel & Ellstrand 2001).

We used Monte Carlo methods to determine the amount of genetic diversity included in different numbers of populations of these taxa. For each species, we drew from 1 to $N - 1$ populations (without replacement) from the empirical data, where N was the total number of populations. We estimated H_e and the proportions of total alleles and total common alleles from populations of each species represented in each subsample. We repeated this procedure 1000 times for each number of populations for each species. From each of these replicate samples, we determined the mean, standard error, minimum, and maximum of the proportion of total and common alleles, as well as of the realized values of H_e.

We evaluated the proportion of alleles and levels of H_e for the numbers of populations included under three conservation intensities (one population, 10–12% of populations, and five populations). For these species, 10–12% of populations ranged from two to four, thus falling between the target of five populations (Center for Plant Conservation 1991) and the minimum set approach. We also determined the number of populations necessary to include all alleles and all common alleles. Al-

though we do not necessarily advocate capturing all alleles as a realistic conservation goal, it provides a convenient and unambiguous benchmark for comparison across species. Further, we assessed how well different numbers of populations met the conservation standard established for common alleles used by the Center for Plant Conservation (1991) and the standard we adopted for H_e. Specifically, we estimated the probability of obtaining all common alleles in each number of populations as the proportion of the 1000 samples that included them all. The number of populations required to meet the conservation standard was identified as the number at which 0.90–0.95 of the 1000 samples included all alleles. Similarly, we evaluated the number of populations required to meet the H_e standard by estimating the proportion of 1000 populations that fell within $\pm$10% of the H_e realized from all populations combined. The number of populations required was identified as the number at which 0.95 of the samples were within $\pm$10% of the species value.

Results

Numbers of Alleles

The proportion of total alleles represented increased with the number of populations sampled, and the number of new alleles included with each additional population decreased (Fig. 1). One randomly selected population included, on average, between 42% (*A. albens*) and 56% (*O. parishii* var. *goodmaniana*) of the total number of alleles, and variances and ranges in realized proportions were large (Fig. 1). For example, a single population included as little as 25% of all alleles (*A. albens*) or as much as 73% (*O. parishii* var. *goodmaniana*; Fig. 1). Five populations included, on average, 67–83% of all alleles (Fig. 1). In three species, the maximum percentage of alleles included in five populations was $\geq$90%, but all alleles were never included. At a minimum, five populations included 54% of the alleles in a species (Fig. 1). Although capturing all alleles was possible with a moderate number of populations, doing so reliably was difficult due to the presence of alleles that are restricted in distribution. For example, the smallest number of populations to include all alleles ranged from 6 (42%, *O. parishii* var. *goodmaniana*) to 17 (57%, *A. albens*), but only 1 or 2 of the 1000 samples (0.1–0.2%) of these numbers of populations included all alleles.

On average, 60% (*E. parishii* and *E. ovalifolium* var. *vineum*) to 70% (*A. albens* and *O. parishii* var. *goodmaniana*) of common alleles were included when only one population was selected (Fig. 1). One population could have included as little as 42% (*E. parishii*) of the common alleles, however, and never included all (Fig. 1). Five populations represented, on average, between 85% (*E. parishii*) and 93% (*A. albens*) of common alleles (Fig. 1), but this number of populations could have included as little as 69% (*E. parishii*). For three species (all but *E. parishii*), five populations could include all common alleles (Fig. 1), but they were included in only 0.5–4.0% of the 1000 samples (Fig. 2). For *E. parishii*, at least nine populations were required to include all common alleles, and only 0.1% of the samples of this number of populations included all. Fewer populations were required to include common alleles than all alleles because there were fewer of them and because common alleles occurred in more populations.

To meet the genetic-diversity conservation standard of a 90% probability of including all common alleles, between 16 (*A. albens*) and 30 (*E. parishii*), as well as all 14 *O. parishii* var. *goodmaniana* populations, were required (Fig. 2). Between 18 (*A. albens*) and all 31 (*E. parishii*) populations were needed for a 95% probability of including all common alleles (Fig. 2). Thus, between 53% and 100% of all populations were required to meet the established conservation standard. The proportion of populations required increased somewhat as differentiation among populations increased and as the total number of common alleles in a species increased (Table 1; Fig. 2), but a surprisingly high proportion of populations was required even in *A. albens*, which had a very low θ_P value (0.01).

Gene Diversity

As expected, mean estimates of H_e in subsamples were close to the species-level values, even for small numbers of populations (Fig. 3). Further, H_e was mostly unaffected by removal of rare alleles from the analysis (Table 1), so we present only the set of figures based on all alleles. As we observed with number of alleles, variance in realized values from small numbers of populations was substantial, and thus subsets of populations could differ greatly from the species values of H_e (Fig. 3). Differences from species-level H_e were equally likely to be above or below the mean value for any particular number of populations, and variance increased with θ_P of a species (Fig. 3).

Between 20% (6, *A. albens*) and 64% (9, *O. parishii* var. *goodmaniana*) of populations were required to meet our conservation standard of obtaining H_e values within $\pm$10% of estimates from all populations (Fig. 4). The proportion of populations of a species needed to meet this standard was dependent on θ_P, increasing more than three-fold over the range of θ_P values in these species (Fig 4).

Discussion

Any decisions to preserve some populations of a species and not others will have genetic consequences. Our

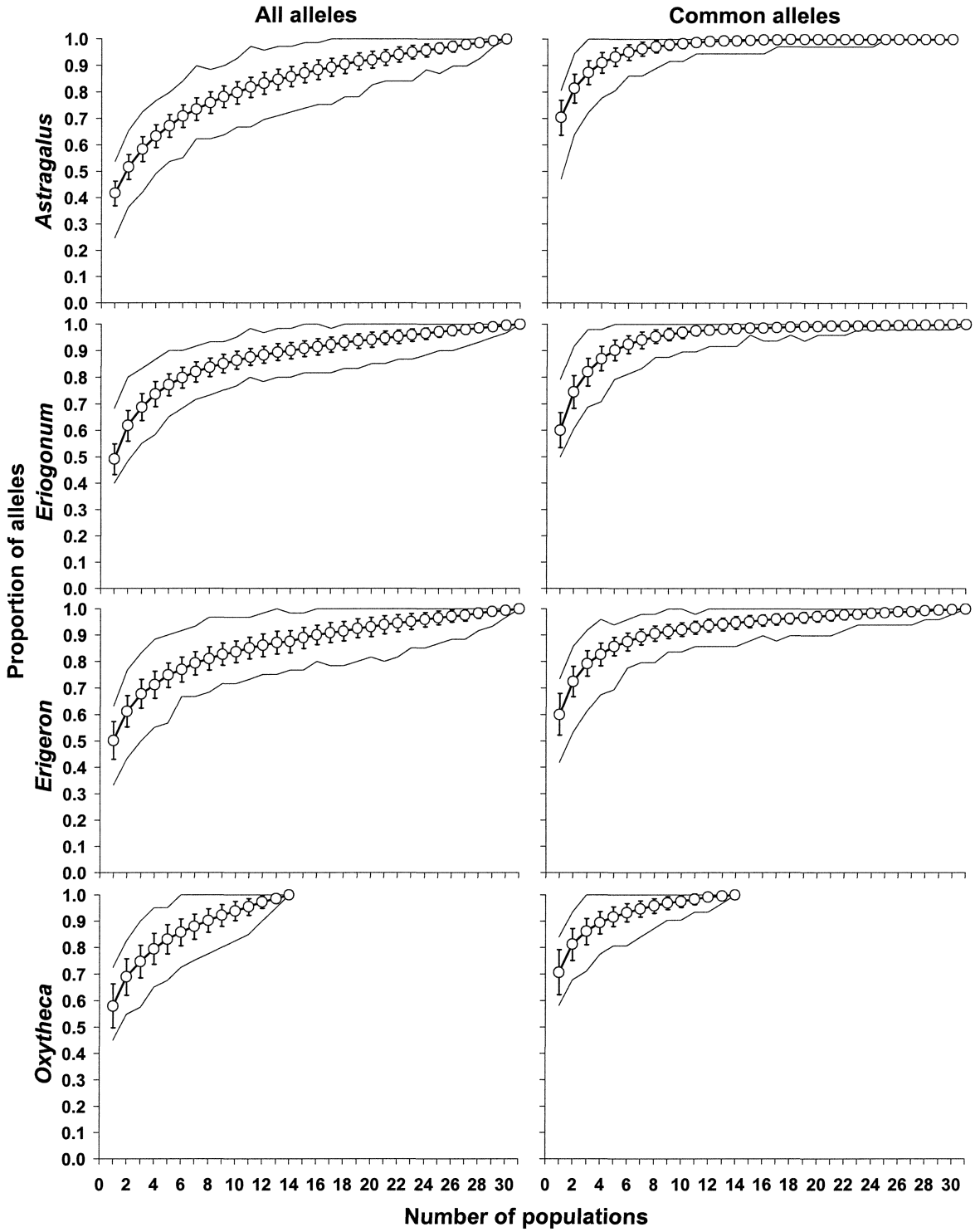

Figure 1. Proportion of all alleles and of all common alleles (alleles occurring at >0.05 frequency in at least one population) included in randomly chosen subsamples of populations of Astragalus albens*,* Erigeron parishii*,* Eriogonum ovalifolium *var.* vineum*, and* Oxytheca parishii *var.* goodmaniana*. Mean values and standard errors are indicated. Maximum and minimum values are denoted by solid lines.*

principal objective was to assess the genetic consequences of population loss in a general manner and in comparison with established conservation targets and intensities. When populations were chosen with no prior knowledge of genetic diversity, large numbers of populations were necessary to ensure that genetic diversity would be conserved. Our results are contrary to conventional wisdom and, more important, to generally

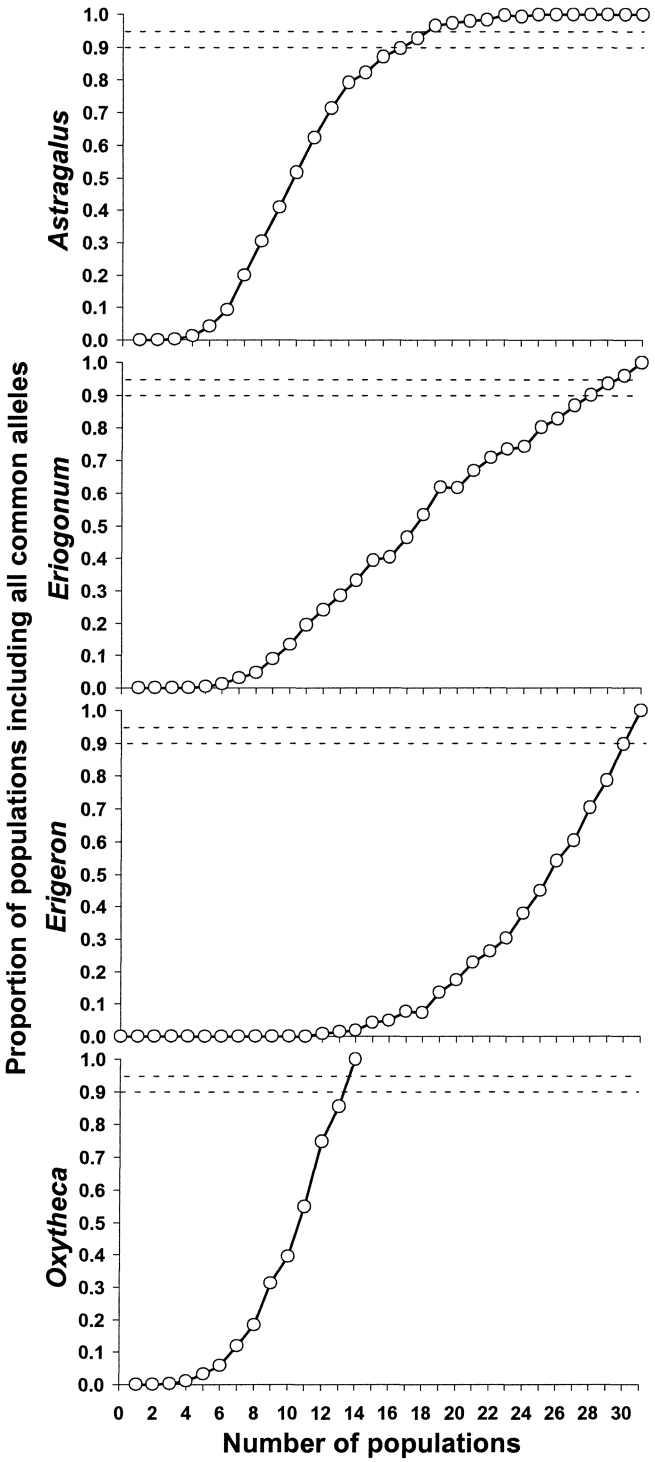

Figure 2. Proportion of 1000 samples that included all common alleles (alleles occurring at >0.05 in at least one population) for each number of populations of the four taxa. Levels corresponding to the 90% and 95% probabilities of all common alleles are denoted with dashed lines.

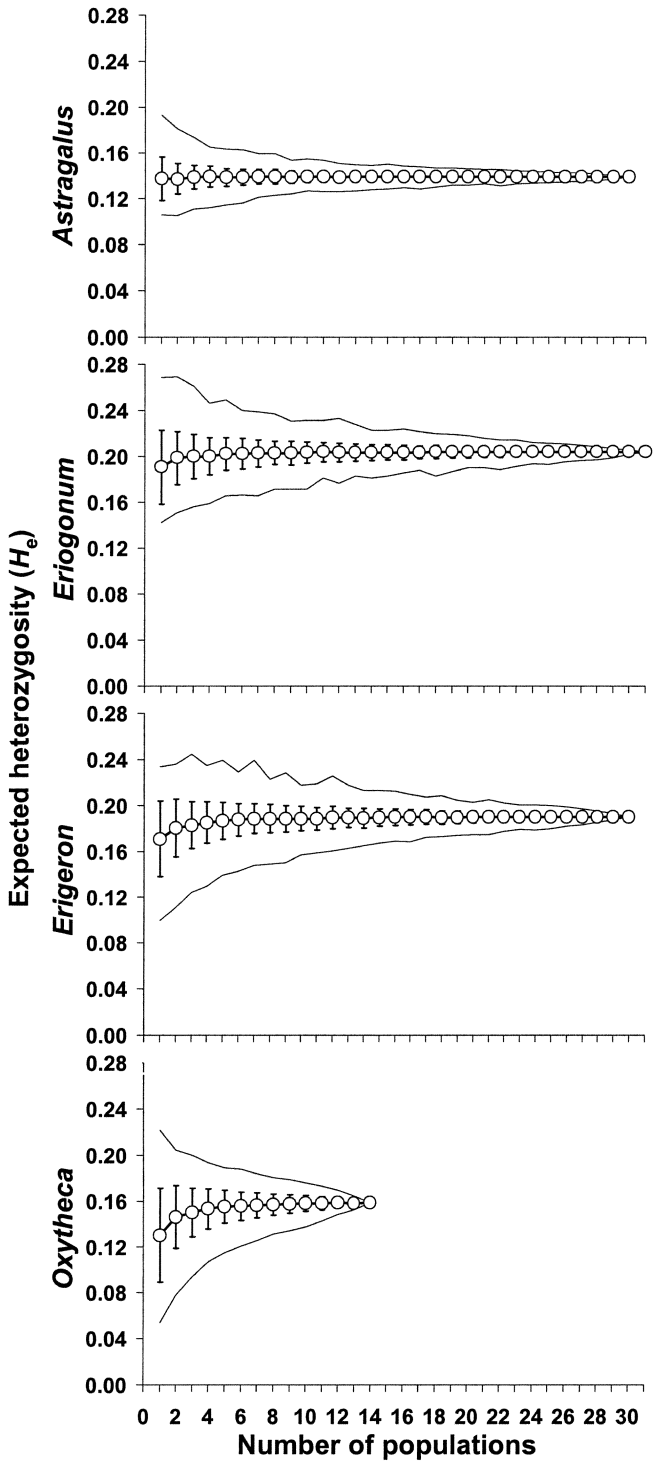

Figure 3. Levels of expected heterozygosity (H_e) included in randomly chosen subsamples of populations of Astragalus albens, Erigeron parishii, Eriogonum ovalifolium *var.* vineum, *and* Oxytheca parishii *var.* goodmaniana. *Mean values and standard errors are indicated. Maximum and minimum values are denoted by solid lines.*

applied and accepted conservation targets. The specific effects vary according to the measure of genetic diversity examined.

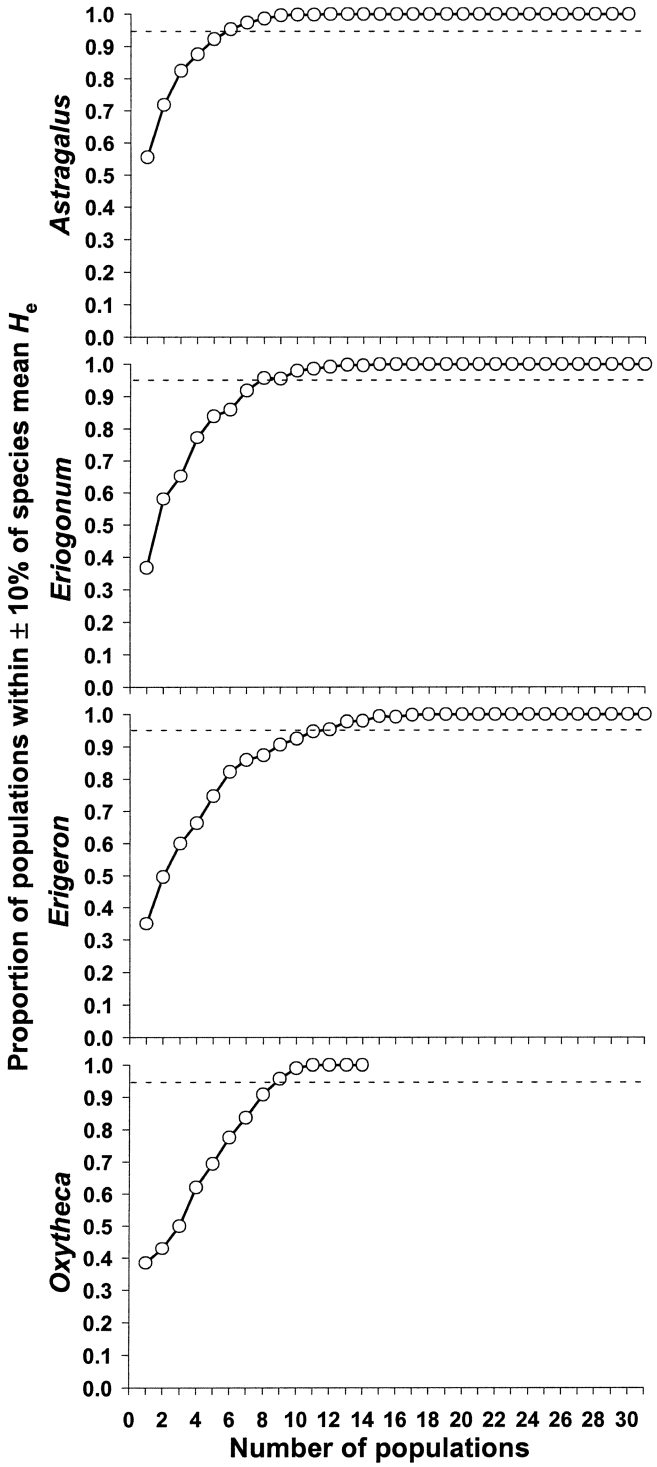

Figure 4. Proportion of 1000 samples for each number of populations of the four taxa that had values of H_e *within ±10% of the mean value realized when all populations are included. Dashed lines indicate the point at which 95% of samples were within ±10%.*

Numbers of Alleles

Minimum-set networks that include one occurrence of each taxon would include, on average, <56% of all alleles and fall short of the conservation standard for common alleles in all four species examined. Minimum-set approaches are also considered inadequate for ensuring the long-term persistence of taxa (e.g., Margules et al. 1994; Rodrigues et al. 2000*b*). The probability of species persistence increases if reserves include five populations of each species rather than one (Rodrigues et al. 2000*a*). Five populations is also the number recommended by the Center for Plant Conservation (1991) as sufficient to capture genetic diversity. Unfortunately, five populations of the species studied here never include all alleles (but would, on average, include 67–83% of them), would only rarely include all common alleles, and would not have a 90–95% probability of including all common alleles (Figs. 1 & 2). Conserving 10–12% of the populations of these species would include even fewer alleles than would conserving five.

Noss (1996) and Soulé and Sanjayan (1998) point out the insufficiency of conserving 10–12% of a land area for maintaining species diversity. Based on theoretical species-area relationships, they suggest that 50% of an area is required (Soulé & Sanjayan 1998). Conserving 50% of the populations of the taxa we studied would include, on average, 86–91% of all alleles (minimum 73%; *O. parishii* var. *goodmaniana*) (Fig. 1). The same target of 50% of populations would conserve, on average, between 95% (*O. parishii* var. *goodmaniana*) and 99% (*A. albens*) and a minimum of 84% of all common alleles (Fig. 1). Minimum values fell above 90% of all common alleles for all taxa except *O. parishii* var. *goodmaniana* (Fig. 1). Although substantial allelic diversity could still be lost, 50% of populations would capture substantially more allelic diversity than the other conservation targets we examined.

All alleles may be represented in fewer populations if one has thorough knowledge of diversity levels and population genetic structure. For example, Ceska et al. (1997) included 98% of the 49 allozyme alleles observed at 9 polymorphic and 28 monomorphic loci in only 2 of 10 known populations of the endangered plant *Baptisia arachnifera* (G_{ST} = 0.096; all alleles detected had frequencies of >0.05). Petit et al. (1998) represented all 38 alleles (common and rare) in 5 of the 12 known populations of *Argania spinosa* by selecting the most diverse and divergent populations. Based on known diversity patterns, the four taxa we studied require a minimum of between 6 populations (43%) of *O. parishii* var. *goodmaniana* and 17 populations (57%) of *A. albens* to capture all alleles and a minimum of between 3 (10%, *A. albens*; 21%, *O. parishii* var. *goodmaniana*) and 9 (29%, *E. parishii*) populations to include all common alleles (Fig. 1). Unfortunately, genetic-

diversity data are lacking for most species and, more important, land-acquisition decisions rarely rely directly on genetic-diversity data. Our results indicate that when conservation decisions are made without such data, larger proportions of populations are needed to ensure representation of alleles than are currently advocated.

Among-population differentiation in a species has obvious implications for the number of populations needed to represent allele diversity. In general, large numbers of populations are required to represent diversity when there is a large proportion of variance among populations. Our results are important in demonstrating, however, that the converse is not true. Although low values of θ_P would lead one to believe that the genetic information in individual populations is highly redundant, even the taxon with the least among-population differentiation (*A. albens*, $\theta_P = 0.01$) (Table 1), required 17 populations to capture all alleles and 16 populations to meet the conservation standard for common alleles. The large number of populations resulted from alleles that occurred only in small subsets of populations, including 14 private alleles (Table 1). Such local diversity is important to capture in conservation efforts and represents variation to which θ_P is not sensitive, that represented by narrowly distributed, low-frequency alleles.

There are a number of possible explanations for the differences between our results and the theoretical expectations of Brown and Briggs (1991). One potential factor is the consequence of agglomerating samples from separate populations. Chakraborty et al. (1988) found that although mean heterozygosity is insensitive to agglomeration, the number of rare alleles (particularly those restricted in distribution to one or few populations) is inflated beyond what one would expect simply as a result of the increased number of individuals sampled. Inflation in rare alleles leads to an increase in the number of populations necessary to capture these alleles. Such effects of agglomeration were likely not incorporated into the allele frequency-distribution models used by Marshall and Brown (1975) and Brown and Briggs (1991). Another explanation for the differences between theoretical predictions and our empirical observations may be that the work of Brown and Briggs (1991) has been both misinterpreted and misapplied. They based their recommendations on the dynamics of a single locus (Marshall & Brown 1975) and relatively few populations. As the number of polymorphic loci and the number of populations increases, the number of populations needed to capture all alleles increases. Yet another important factor is that the four species we studied have large numbers of alleles per locus compared with other plant taxa (Hamrick & Godt 1989; Gitzendanner & Soltis 2000). Certainly, genetic diversity in less diverse taxa would be conserved in fewer populations.

Gene Diversity

In contrast to allelic diversity, average species-level values for H_e were achieved with very few populations. However, the standard for realizing a value of H_e within $\pm 10\%$ of the species-level value was not met by conserving five or fewer populations (Fig. 4). Indeed, to have a 95% probability of being within $\pm 10\%$ of the species value, 20–64% of all populations were required. Our results contradict the expectation that most of the evolutionarily significant genetic variation as measured by H_e is easily represented in a few populations. That expectation is based on the insensitivity of average H_e values to changes in frequencies of uncommon alleles, but it overlooks the large variance in H_e associated with small numbers of populations that our results demonstrate (Fig. 3). Large variances indicate that there is a low probability of maintaining levels of gene diversity when selecting small numbers of populations. Thus, it is misguided to focus simply on average values of H_e as a justification for determining the number of populations necessary to represent this measure of diversity. Not surprisingly, the proportion of populations needed to yield acceptable values of H_e increases with increasing values of θ_P (Fig. 3). The relationship of θ_P and H_e is in concordance with observations by Schoen and Brown (1991), who noted that species with higher θ_P values have more among-population variation in H_e.

General Application

Our results are likely to be applicable to other rare species because the taxa we studied have a range of population differentiation as characterized by θ_P (Table 1), provide broad taxonomic representation (including four genera and three families in distantly related orders [Fabales, Polygonales, and Asterales]), and represent a diversity of life-history characteristics (an annual herb, two short-lived-perennial herbs, and a long-lived-perennial subshrub) (Neel 2000). Data on mating system is available only for *E. ovalifolium* var. *vineum*, and this species is highly outcrossed (Neel et al. 2001). When applying these results to other rare species, it is important to remember that rarity encompasses many different combinations of geographic distribution and population size (Rabinowitz 1981). The taxa we examined are extremely limited in distribution but are locally abundant in appropriate habitat. For rare taxa such as these, there is little immediate risk of extinction as a result of lack of genetic diversity; rather, the focus is on maintaining habitat and protecting populations from threats related to human activities and environmental stochasticity (Soulé & Simberloff 1986). Taxa with these characteristics are precisely those for which saving only a subset of the populations is often necessary and to which these findings are most applicable. Although decisions regarding

conservation of such taxa may not center on genetic concerns, it is still important to maintain existing levels of genetic diversity to prevent future genetic risks (Sherwin & Moritz 2000). Our results provide insight into the number of populations necessary to accomplish this goal.

Generalizations specifically for numbers of alleles should be applied with some caution, because the number of alleles for the four taxa we examined were higher than are typically found in other rare taxa (e.g., Hamrick & Godt 1989; Gitzendanner & Soltis 2000), and fewer populations are required if allele numbers are lower. Although taxa that are less diverse might require fewer populations, it should not be assumed that rare species are genetically depauperate. Narrowly endemic species tend to have less genetic variation than widespread taxa (Hamrick & Godt 1989), but they can be at least as diverse as widespread congenors (Gitzendanner & Soltis 2000). In contrast to number of alleles, our values of H_e were well within range of those for other rare species (e.g., Hamrick & Godt 1989; Gitzendanner & Soltis 2000), so the numbers of populations implied by these values should be widely applicable.

Summary and Conclusions

We demonstrated that in the absence of genetic-diversity data, it is necessary to conserve 53–100% of populations to capture all alleles and to meet the genetic-diversity conservation standard adopted by the Center for Plant Conservation (1991). It is necessary to conserve 20–64% of populations to reliably represent heterozygosity. Thus, currently applied, generalized conservation targets, and especially minimization strategies, are likely to be inadequate to conserve genetic diversity. Conserving the larger numbers of populations necessary to meet existing genetic conservation standards would also provide a more realistic chance of maintaining the existing among-population structure and processes, such as gene flow, that help perpetuate diversity over time. Conserving larger numbers of populations or amounts of habitat also increases the probability of long-term species persistence by reducing stochastic extinction threats and maintaining ecological processes, and thus is in keeping with reserve-design principles based on basic population biology and community ecology (e.g., Simberloff & Abele 1982; Simberloff 1988).

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