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Population genetics of colonizing success of weedy rye in Northern California

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Summary. Genetic parameters of 11 weedy rye populations located in California's northern mountain area and the adjoining Oregon border were compared with those of the putative parents, wild species *Secale montanum* and cultivated rye *S. cereale*. All weedy populations exhibited high levels of genetic variation as determined by isozyme analysis. On average, 44% of the isozyme loci were polymorphic, total genetic diversity was 0.30; and number of alleles per locus was 1.65. High genetic identities, averaging 0.994 ± 0.005 between populations, indicated that little genetic differentiation has occurred among these weedy populations since the initial colonization. Lack of population differentiation could be attributed to a wind-pollinated, self-incompatible breeding system resulting in extensive gene flow among weedy populations, and between weedy populations and local cultivars of rye. Multilocus outcrossing rates of weedy populations ranged from 0.86 to 0.97. The estimated levels of gene flow using the private-alleles method were high among weedy populations, and between cv 'Merced' and weedy populations, with estimated *Nm* values of 14.50 and 8.21, respectively. The colonizing success of weedy rye is discussed and a strategy for its conservation recommended.

Key words: Colonization – Genetic diversity – Outcrossing rate – Gene flow

Introduction

Secale cereale L. and *S. montanum* Guss are self-incompatible species in the genus (*Secale* ($2N = 2X = 14$). Several cytogenetic and evolutionary studies of the genus have indicated that the annual *S. cereale* envolved from the perennial *S. montanum* as a result of progressive cytological and morphological differentiation, and that cultivated rye (*S. cereale* ssp. *cereale*) originated from the weedy races of *S. cereale* through both natural selection and some early artificial selection (Khush and Stebbins 1961; Khush 1962, 1963). Cytologically, *S. cereale* differs from *S. montanum* by two reciprocal translocations involving three pairs of chromosomes (Khush and Stebbins 1961; Stutz 1972). Despite a partial fertility barrier between the two species due to the chromosome rearrangement, spatial contacts between them still result in natural hybridizations, as shown by the occurrence of morphologically intermediate and recombinant individuals in the area of sympatry in central and eastern Turkey (Zohary 1971).

The natural establishment and rapid spread of weedy rye populations in California's northern mountain area was first reported by Suneson et al. (1969). These authors postulated that these populations probably developed from an introduced interspecific hybrid between *S. cereale* and *S. montanum* with recurrent introgression from the many cultivears of rye in the Fall River area. This area seems to be the "center of origin" of weedy rye in California as it shows the highest population densities and genetic diversity. New areas of establishment and abundance, especially along roadsides in the Northern California and adjoining Southern Oregon areas, are prominent today. These weedy populations occur in small or large mixed stands, in mesic or xeric sites, near pastures and fields, in habitats with various degrees of disturbance. In their comparative study of weedy populations, cultivars, and wild perennial *S. montanum*, Suneson et al. (1969) found that the weedy rye was intermediate between the cultivars and *S. montanum* in head-

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Table 1. List of accessions/cultivars of *S. cereale* and *S. montanum* studied

Population designation	Taxon	Inventory/cultivar	Origin	Source[a]
Merced	*S. cereale* spp. *cereale*	Merced	California	Davis, Calif.
Mont. 240285	*S. montanum* ssp. *anatolicum*	PI240285	Turkey	USDA-ARS
Mont. 240286	*S. montanum*	PI240286	Turkey	USDA-ARS
Mont. 272338	*S. montanum*	PI272338	Hungary	USDA-ARS
Mont. 326282	*S. montanum* ssp. *kuprijanovii*	PI326282	Soviet Union	USDA-ARS
Mont. 445973	*S. montanum* ssp. *anatolicum*	PI445973	USA	USDA-ARS

[a] Davis, Calif. – Department of Agronomy and Range Science, University of California, Davis, California USA; USDA/ARS – National Small Grains Collection, USDA-ARS, Small Grains Germplasm Research Facility, P.O. Box 307, Aberdeen, Idaho 83210 USA

ing habit, spike fragility, floret sterility, and morphological characters. There was considerable phenotypic variation for both morphological and quantitative traits within and among weedy rye populations (Jain 1977). These early studies suggested that the causes of interpopulation heterogeneity in these traits could not be explained in terms of selection, migration, or genetic drift.

Weedy rye has proven to be a successful colonizer after only about 50 years (generations) in Northern California. The genetic and ecological features associated with colonizing success in weedy species have been summarized in Baker and Stebbins (1965), Brown and Marshall (1981), Barrett and Richardson (1985), and Barrett and Husband (1990). Studies on the population genetics of colonizing species have shown that (1) colonial populations are depauperate in genetic variation, presumably due to founder effects; (2) predominant self-fertilization or apomixis is common among successful colonizing annuals, which ensures reproduction or progeny fidelity; (3) substantial population differentiation occurs following invasion due to isolation, genetic drift, or natural selection, among other genetic features. However, our population genetic survey of weedy rye populations and their putative parental species showed a unique mode of colonization.

The present study was conducted to: (1) test hypotheses on the hybrid origin of weedy rye and recurrent introgression from cultivars; (2) document allozyme variation within and among the weedy populations and compare the allozyme information with previous morphological measures of variation and extent of population differentiation (Jain 1977); (3) investigate the relative roles of genetic drift, mating system, gene flow, and selection in affecting population genetic attributes of weedy rye in the processes of colonization.

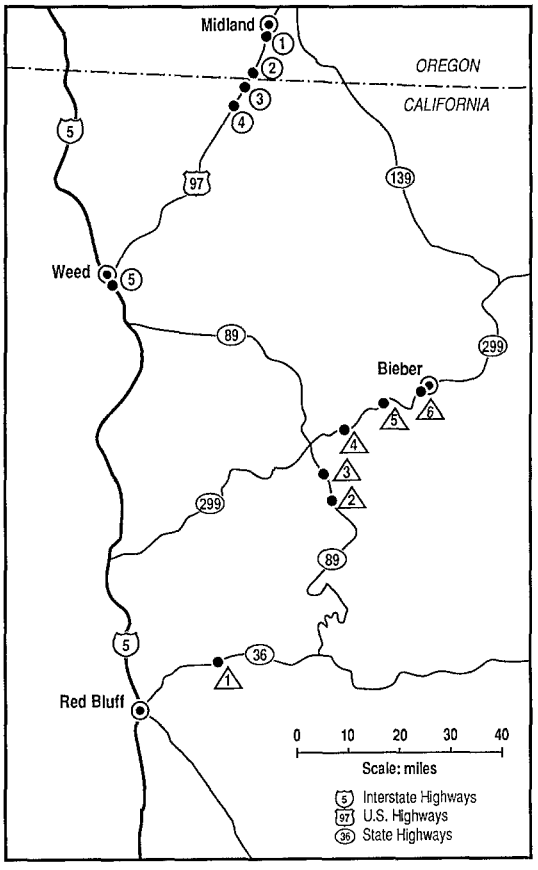

Fig. 1. Map showing location of populations of weedy rye sampled. Populations sampled in 1988 along the transect Highway 97 are labeled 1 to 5 in *circles*; populations sampled in 1989 along the transect Highway 36-89-299 are labeled 1 to 6 in *triangles*

Materials and methods

Seeds were collected from 11 weedy rye populations in California's northern mountain area and the adjoining Oregon border in late summer or early fall of 1988 and 1989. Geographical locations of these populations are shown in Fig. 1, and more detailed information on their locations and habitats are available on request. Seeds collected from different plants were kept separately to provide family progeny arrays for studying breeding system parameters. Seeds were collected as a bulk from only one population (*Rye5* 1988, along the transect Highway 97) because most spikes had shattered at time of collection. The sources of seeds of *S. cereale* cv 'Merced' and of accessions of the wild species *S. montanum* are listed in Table 1. These seeds were obtained as a population bulk in all cases.

One-week-old seedlings were used for electrophoresis. Enzymes were extracted using the buffer described by Sun and

Table 2. Genetic parameters and outcrossing rates in populations of weedy rye along the transect Highway 97

Population	Sample size family/individual	*PLP* (%)	*A*	*H*	h_0	h_e	t_m	t_s	t_f	*F*
Rye1 1988	36/359	40.0	1.53	0.12 (0.05)[a]	0.31 (0.01)	0.29 (0.01)	0.97 (0.03)	0.96 (0.04)	1.07	−0.02
Rye2 1988	37/399	33.3	1.53	0.13 (0.06)	0.33 (0.01)	0.34 (0.01)	0.93 (0.04)	0.91 (0.05)	1.00	0.00
Rye3 1988	36/456	33.3	1.53	0.13 (0.06)	0.31** (0.01)	0.35 (0.01)	0.86* (0.04)	0.78 (0.05)	0.85	0.09**
Rye4 1988	39/537	40.0	1.67	0.13 (0.05)	0.31 (0.01)	0.32 (0.01)	0.92 (0.04)	0.88 (0.04)	0.97	0.02
Rye5 1988	59/59	50.0	1.67	0.16 (0.05)	0.27** (0.02)	0.32 (0.02)	–	–	0.90	0.10**
Mean		39.3	1.59	0.13	0.31	0.32	0.92	0.88	0.96	0.04

* $P<0.05$ between t_m and t_s; ** $P<0.01$ between h_0 and h_e (*t*-test)
PLP, Percentage of loci polymorphic; *A*, mean number of alleles per locus; *H*, mean genetic diversity; h_0, mean observed heterozygosity over polymorphic loci; h_e, mean expected heterozygosity over polymorphic loci; t_m, multilocus outcrossing rate; t_s, mean single-locus outcrossing rate; t_f, mean single-locus outcrossing rate based on gene fixation index; *F*, mean gene fixation index
[a] Numbers in parentheses are standard errors

Ganders (1990). Samples were loaded onto a 12% starch gel, and electrophoresis was conducted in a cold chamber at 4 °C for 5 h. Two buffer systems were used to resolve 13 enzymes. A gel buffer containing 0.005 *M* DL-histidine, 0.0135 *M* TRIS, and 0.004 *M* citric acid (pH 7.0), and an electrode buffer containing 0.135 *M* TRIS and 0.044 *M* citric acid (pH 7.0), were used for aconitase (ACO, E.C.4.2.1.3), aspartate aminotransferase (AAT, E.C.2.6.1.1), glucose-6-phosphate dehydrogenase (G6P, E.C.1.1.1.49), β-glucosidase (GLU, E.C.3.2.1.21), isocitrate dehydrogenase (IDH, E.C.1.1.1.42), phosphoglucomutase (PGM, E.C.2.7.5.1), 6-phosphogluconate dehydrogenase (PGD, E.C.1.1.1.44), and shikimic acid dehydrogenase (SKDH, E.C.1.1.1.25). The buffer system "L" described by Shields et al. (1983) was used for aldolase (ALD, E.C.4.1.2.13), diaphorase (DIA, E.C.1.6.4.3), glutamate dehydrogenase (GDH, E.C.1.4.1.3), leucine-amino peptidase (LAP, E.C.3.4.11.1), and phosphoglucose isomerase (PGI, E.C.5.3.1.9).

Genetic parameters were computed for a total of 18 isozyme loci. Genetic diversity within a population (*H*) was calculated as $H=1-\Sigma p_i^2$, where p_i is the frequency of the *i*th allele at a locus. Mean genetic diversity was obtained by averaging the *H* values over all loci. Distribution of genetic variation among populations was estimated using Nei's genetic diversity statistics (Nei 1973). Total genetic diversity (H_T), mean diversity within populations (H_S) and among populations (D_{ST}), and the coefficient of genetic differentiation among populations (G_{ST}) were computed. The dendrogram of Nei's genetic distance (Nei 1972) was constructed using the unweighted pair group method (UWPGM) of Sneath and Sokal (1973). Wright's fixation index, *F*, (Wright 1969) was estimated for each polymorphic locus as $F=1-(H_O/H_e)$, where H_O is observed heterozygosity and H_e is expected heterozygosity under panmixia at the locus. The estimates of single-locus *F* values were averaged to obtain the mean *F* value for each population. Multilocus outcrossing rates (t_m) and mean single-locus outcrossing rates (t_s) were estimated from progeny arrays using the maximum log-likelihood method of Ritland and Jain (1981) for four populations along the Highway 97 transect. A different method of estimating outcrossing rates is to use Wright's fixation index *F*, with $t_f=(1-F)/(1+F)$. This estimate assumes that the population is at inbreeding equilibrium, whereas the t_m method is independent of this assumption. When comparisons were made between estimates of t_m with those of t_f, the values were found to be similar in all four populations where outcrossing rates could be estimated using both methods. Outcrossing rates (t_f) were computed for all populations obtained as a bulk and for the six populations collected along the Highway 36-89-299 transect.

The level of gene flow, *Nm* (the number of migrants per generation, where *N* is the effective population size and *m* is the rate of migration), among populations was estimated using the private-alleles method (Slatkin 1985; Barton and Slatkin 1986; Slatkin and Barton 1989). The relation used was $\log_{10}[\bar{p}(1)]=a\log_{10}(Nm)+b$, in which $\bar{p}(1)$ is the average frequency of private alleles and $a=-0.612$, $b=-1.21$ for a sample size of 50 per population. Another estimate of *Nm* was obtained using the G_{ST} method (Slatkin and Barton 1989): $Nm=(1-G_{ST})/4G_{ST}$, where G_{ST} is Nei's estimator of Wright's F_{ST} (Wright 1951).

Results

Genetic variation

Eighteen isozyme loci were resolved for 13 enzyme systems. Of the 18 loci, 8 were monomorphic including *Aat2*, *Ald1*, *Gdh1*, *Gdh2*, *G6p1*, *G6p2*, *Lap1*, and *Skdh1*. The 10 polymorphic loci were *Aat1*, *Aco1*, *Aco2*, *Dia1*, *Glu1*, *Idh1*, *Pgd1*, *Pgd2*, *Pgi2*, and *Pgm1*. A total of 39 codominant alleles were observed at these loci. The polymorphic loci have the same common alleles with the exception of the *Dia1* locus. A smaller sample size was available for this locus in most of the populations studied. Allele frequencies for the 11 weedy populations, cv 'Merced', and five accessions of *S. montanum* are available on request.

The level of genetic variation in the weedy populations was high and was comparable to the level displayed

Table 3. Genetic parameters and outcrossing rates in populations of weedy rye along the transect Highway 36-89-299

Population	Sample size family/individual	*PLP* (%)	*A*	*H*	h_0	h_e	t_f	*F*
Rye1 1989	72/72	44.4	1.56	0.15 (0.05)	0.34 (0.02)	0.35 (0.02)	0.93	0.06
Rye2 1989	55/55	44.4	1.56	0.18 (0.05)	0.40 (0.03)	0.40 (0.03)	1.01	0.01
Rye3 1989	40/40	50.0	1.78	0.16 (0.05)	0.28** (0.02)	0.32 (0.02)	0.95	0.10**
Rye4 1989	40/40	50.0	1.72	0.16 (0.05)	0.33 (0.02)	0.33 (0.02)	1.05	−0.01
Rye5 1989	44/44	50.0	1.72	0.17 (0.05)	0.33 (0.02)	0.34 (0.02)	0.98	0.03
Rye6 1989	42/42	50.0	1.83	0.18 (0.06)	0.36 (0.02)	0.36 (0.02)	1.02	0.01
Mean		48.1	1.70	0.17	0.34	0.35	0.99	0.03

** $P<0.01$ between h_0 and h_e (*t*-test)
Abbreviations as in Table 2

Table 4. Genetic parameters and outcrossing rates in cv 'Merced' and accessions of *S. montanum*. Means of all weedy populations from Table 2 and 3 are given below for comparison

Population	Sample size family/individual	*PLP* (%)	*A*	*H*	h_0	h_e	t_f	*F*
Merced	129/129	38.9	1.61	0.15 (0.05)	0.39 (0.02)	0.38 (0.02)	1.16	−0.07
Mont. 240285	17/17	44.4	1.56	0.17 (0.05)	0.40 (0.04)	0.39 (0.04)	1.18	−0.05
Mont. 240286	19/19	44.4	1.56	0.17 (0.05)	0.43 (0.03)	0.41 (0.04)	1.21	−0.05
Mont. 272338	61/61	38.9	1.67	0.14 (0.05)	0.38 (0.02)	0.37 (0.02)	1.05	−0.01
Mont. 326282	42/42	38.9	1.44	0.14 (0.05)	0.30 (0.02)	0.31 (0.02)	0.95	0.04
Mont. 445973	42/42	38.9	1.50	0.14 (0.05)	0.38 (0.03)	0.36 (0.03)	1.18	−0.05
Mean		40.7	1.56	0.15	0.38	0.37	1.12	−0.03
Mean of all weedy populations		44.1	1.65	0.15	0.32	0.34	0.97	0.03

Abbreviations as in Table 2

in cv 'Merced' and in the accessions of *S. montanum* (Tables 2, 3, 4). The mean values of percentage of polymorphic loci (*PLP*) and gene diversity (*H*) in the weedy populations from the transect Highway 97 were significantly lower than those from the transect Highway 36-89-299 (*t*-test, $P<0.05$). These differences might be an artifact caused by missing data at three polymorphic loci in four of the populations along the transect Highway 97. No significant difference was detected between the two transects in the mean number of alleles per locus (*A*), observed heterozygosity (h_O), expected heterozygosity (h_e), outcrossing rate (t_f), and fixation index (*F*). However, the mean values of *PLP* and *A* in the weedy rye populations were significantly higher than in their putative parents (*t*-test, $P<0.05$). In the weedy populations, the observed heterozygosity at polymorphic loci (h_O) averaged 0.32, while the expected value (h_e) was 0.34 (Table 4). The deficiency of heterozygotes resulted in a positive *F* value of 0.03. In contrast, negative *F* values were found in cv 'Merced', and four out of the five

Table 5. Distribution of genetic variation in populations

Populations	N	All loci			Polymorphic loci			G_{ST}
		H_T	H_S	D_{ST}	H_T	H_S	D_{ST}	
Weedy populations	11	0.155	0.150 (0.053)[a]	0.005 (0.005)	0.296	0.287 (0.079)	0.009 (0.008)	0.032
S. montanum	5	0.175	0.154 (0.049)	0.021 (0.011)	0.315	0.277 (0.067)	0.038 (0.017)	0.120
Merced and *S. montanum*	6	0.172	0.153 (0.049)	0.019 (0.009)	0.309	0.275 (0.067)	0.035 (0.014)	0.110
Total	17	0.163	0.151 (0.052)	0.012 (0.008)	0.304	0.283 (0.075)	0.022 (0.013)	0.074

N, Number of populations; H_T, total genetic diversity; H_S, genetic diversity within populations; D_{ST}, genetic diversity among populations; G_{ST}, coefficient of population differentiation

[a] Numbers in parentheses are standard errors

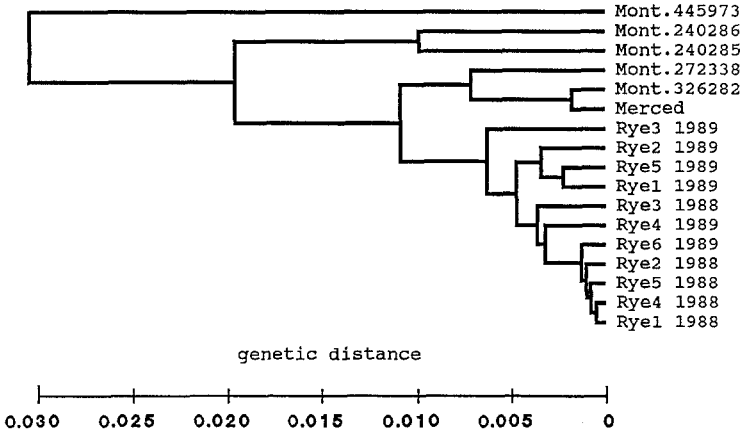

Fig. 2. Dendrogram of Nei's genetic distances between the weedy rye populations, cv 'Merced', and accessions of *S. montanum*, based on gene frequencies at 17 isozyme loci

accessions of *S. montanum*, ranged from −0.01 to −0.07, with an average of −0.03. The negative F values indicated a slight heterozygote excess in comparison with the Hardy-Weinberg expectation. The difference in the mean values of F between the weedy rye and its putative parents was also significant.

Outcrossing rates

The breeding system of rye is predominantly outcrossing (Table 2, 3, 4). The multilocus outcrossing estimates (t_m) ranged from 0.86 to 0.97, averaging 0.92 in the four weedy populations from the transect Highway 97. The mean single-locus outcrossing rates (t_s) for these populations ranged from 0.78 to 0.96, averaging 0.88. The single-locus outcrossing rate was significantly less than the multilocus estimate in only one population, *Rye3* 1988, suggesting the presence of biparental inbreeding (i.e., mating to relatives) in this population. Mean outcrossing rate, t_f, obtained using Wright's fixation index F, was slightly higher than t_m in three of the four populations. The equation $t_f = (1-F)/(1+F)$ assumes that the population is at inbreeding equilibrium. Violation of this assumption may result in a biased outcrossing estimate. The t_f value thus provides only an approximation of t_m, which cannot be computed for populations sampled as a bulk. The average t_f value of 11 weedy populations was 0.97. The t_f values were significantly higher in cv 'Merced' and in the accessions of *S. montanum*, ranging from 0.95 to 1.21, with a mean value of 1.12. An outcrossing estimate (t_f) higher than 1.00 is due to a negative F, and suggests that outcrossing events occur more frequently than expected from random mating, as in the case of self-incompatibility or disassortative mating. Alternatively, if selection favored heterozygotes at the marker loci, the outcrossing rate estimates using F values would be biased upward.

Population genetic differentiation

Distribution of genetic variation among populations is shown in Table 5. Total genetic diversity (H_T) in weedy populations was 0.155 for all loci and 0.296 for polymorphic loci. Slightly higher H_T values were found in the cv

'Merced' and accessions of *S. montanum* i.e. 0.172 for all loci and 0.309 for polymorphic loci. Most of this genetic diversity was distributed within populations, and little was found among populations, as shown by a high H_S value and a low D_{ST} value in each group of populations. The lowest G_{ST} value, 0.032, was found in the 11 weedy populations, indicating that little genetic differentiation has occurred among these populations. A G_{ST} value four-fold higher was found for the *S. montanum* accessions. This is not surprising because these accessions originated from many different countries that are geographically isolated (see Table 1).

High genetic identity values (average 0.994 ± 0.005) between weedy populations further indicated the lack of genetic differentiation. High genetic identity values were also found for other pairwise comparisons. Genetic identity between accessions of *S. montanum* was 0.969 ± 0.022, between cv 'Merced' and *S. montanum* 0.980 ± 0.018, between cv 'Merced' and the weedy populations 0.984 ± 0.008, and between *S. montanum* and the weedy populations 0.981 ± 0.011. A dendrogram of Nei's genetic distances, based upon allele frequencies at 17 loci (*Dia1* excluded due to a smaller sample size), is given in Fig. 2. The weedy populations grouped together, showing a closer genetic relationship to each other than to cv 'Merced' and the accessions of *S. montanum*. Figure 2 also shows that genetic differentiation has not occurred between the two geographical transects within the weedy rye group.

Gene flow among populations

Estimates of gene flow calculated using the average frequency of private alleles, $\bar{p}(1)$, are given in Table 6. By definition, private alleles are those found in only one population or sampling location. Only 2 of the 37 alleles were private alleles in the weedy populations. However, more information on gene flow can be obtained by computing $\bar{p}(1)$ for different subsets of the data (Slatkin 1985). Considering separately the two geographical transects of the weedy populations, 2 alleles were found only in the transect Highway 97 and 6 alleles only in the transect Highway 36-89-299. Using information on the distribution of private alleles in the subsets of samples, we estimated levels of gene flow, *Nm* among all weedy populations, between geographical transects of weedy populations, between cv 'Merced' and weedy populations, and between *S. montanum* and weedy populations. In addition to $\bar{p}(1)$, the spatial distribution of all other alleles can be used to estimate levels of gene flow (Slatkin 1981). The average frequencies of alleles found in i of the d populations sampled, $\bar{p}(i)$, are shown in Fig. 3. The shape of the $\bar{p}(i)$ curve is consistent with those found in other species with high levels of gene flow (see Fig. 3 in Slatkin 1981).

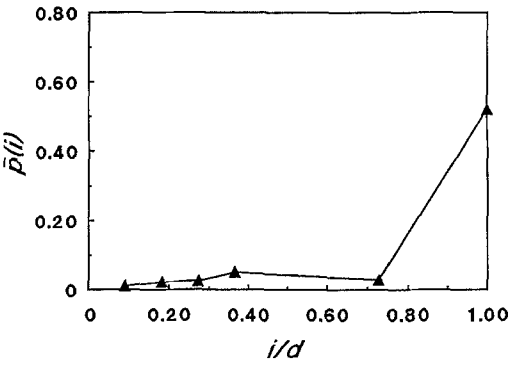

Fig. 3. Conditional average allele frequency $\bar{p}(i)$ plotted against i/d, in weedy populations, where $\bar{p}(i)$ is the average frequency of the alleles present in i populations ($i=1, 2 \ldots, d$), and $d\,(=11)$ is the total number of weedy populations sampled

Table 6. Estimates of gene flow among populations of weedy rye Highway, between geographical transects (populations along Highway 97 versus 36-89-299), between cv 'Merced' and weedy populations, and between accessions of *S. montanum* and weedy populations using Slatkin's private-alleles method

Population	No. of populations	N	No. of private alleles	$\bar{p}(1)$	Nm
All weedy populations	11	50	2	0.012	14.50
Highway 97 versus Highway 36-89-299	5 vs. 6	50	8	0.025	4.37
Merced versus weedy populations	1 vs. 11	50	6	0.017	8.21
S. montanum versus weedy populations	5 vs. 11	50	6	0.042	1.87

$\bar{p}(1)$, mean frequency of private alleles. *Nm*, the average number of migrants exchanged between populations per generation

The estimate of *Nm* based on the G_{ST} value of 0.032 for all the weedy populations was 7.56, which is lower than the private-alleles estimate. The discrepancy between the two estimates could be due to the fact that although in principle G_{ST} and private-alleles methods are expected to give comparable estimates of *Nm* for the same samples, in practice, the G_{ST} method may provide a better estimate because it uses all of the gene-frequency data and, thus, is less sensitive to experimental error. The private-alleles method requires that a reasonable number of private alleles be present and that these alleles be accurately identified. In addition, the private-alleles method assumes that each population sampled is at genetic and demographic equilibrium. The weedy rye may not have reached such an idealized population structure.

Discussion

Genetic variation

In contrast to the low levels of genetic variation commonly found in colonizing populations, all of the weedy rye populations studied contained high levels of genetic variation comparable to or greater than that found in their putative parental species *S. montanum* and *S. cereale* cv 'Merced'. This suggests that founder effects might be insignificant in the weedy colonies. On the other hand, evolution subsequent to an initial population bottleneck might lead to higher levels of genetic diversity by means of hybridization with related species (Brown and Marshall 1981). Nei et al. (1975) showed that the level of genetic variability in colonizing populations depends not only on the number of founders but also on the speed with which large population size is recovered, and that the effect of population bottlenecks on the average number of alleles at each locus is greater than the effect on average heterozygosity. The average number of alleles per locus in the weedy rye populations was significantly higher than in the accessions of *S. montanum* and cv 'Merced', especially in the four populations near the Fall River area (i.e., *Rye3-6* 1989), where initial establishment of the weedy rye population occurred. It can be inferred that the original colonial population may have carried much of the genetic diversity from its source and/or that subsequent introgression with many rye cultivars enriched its allelic composition. The presence of many rye cultivars in the Northern California region provided maximum opportunity for recurrent introgression. Evidence of considerable introgression with local cultivars has been observed in field studies (Suneson et al. 1969). Jain (1977) found comparable high levels of genetic variation for many morphological traits in weedy rye populations. The presence of purple straw and the high frequency of blue aleurone in weedy rye has been attributed to gene flow from cultivars (Suneson et al. 1969; Jain 1977).

The allelic composition of the weedy rye cv 'Merced' and *S. montanum* confirms the hybrid origin of weedy rye. Among the 37 alleles detected in the weedy rye populations, 34 were shared with either, or both, cv 'Merced' and the accessions of *S. montanum*. The three rare alleles not found in the parental samples are likely present in other cultivars or other accessions of *S. montanum*, though it cannot be ruled out that these rare alleles could be the result of mutation since the original hybridization event.

Breeding system and population structure

An outcrossing breeding system has not been encountered frequently in successful annual colonizers, whereas predominant self-fertilization or apomixis is common among them (Stebbins 1957; Allard 1965; Brown and Marshall 1981). The present study indicates that weedy rye is predominantly outcrossed. The estimates of outcrossing rate in the weedy populations ranged from 0.85 to 1.07, with an average of 0.97 (t_f values). However, positive F values, which result from a significant deficiency of heterozygotes relative to that expected from random mating, were detected in a few weedy populations (Tables 2 and 3). Heterozygote deficiency could result from selection against heterozygotes, inbreeding, or a Wahlund effect (the inclusion of two or more genetically distinct units into a single population sample). The inbreeding explanation seems the most convincing in the present situation. Selfing, or biparental inbreeding, could occur in these weedy populations. Jain (1977) reported that about 8% of the spikes of weedy rye set seeds after being bagged. In the population with the lowest estimates of outcrossing rate (*Rye3* 1988), biparental inbreeding, in addition to selfing, might be present, as detected by the significant difference between the values of t_m and t_s (K. Ritland personal communication). Much higher F values were detected at two morphological loci; these ranged from 0.29 to 0.47 and 0.17 to 0.55, respectively, in most of the weedy populations studied by Jain (1977). The discrepancy between the morphological and isozyme measures of fixation index is likely to be because the morphological traits have been subject to stronger selection pressures. The negative F values in cv 'Merced' and the wild species *S. montanum* in the present study may be indicative of the effect of self-incompatibility.

Outcrossing rates in plants are normally under both genetic and environmental regulation. Although rye possesses an effective gametophytic two-locus multiallelic incompatibility system (Lundqvist 1956), the presence of self-compatible alleles, high temperature before and during anthesis, or low population density has been reported to decrease the efficiency of self-incompatibility (Wricke 1979; Schmidt-Stohn et al. 1986; Vaquero et al. 1989). These factors may also account for variation in outcrossing rate both within and among the weedy populations. Vaquero et al. (1989) found a high level of selfing, up to 30%, in a few cultivars of rye. They attributed the selfing to a pseudocompatibility possibly caused by environmental factors. However, we did not detect any significant selfing in the cv 'Merced', nor in the wild species *S. montanum*.

The significant difference in outcrossing rates between the weedy rye and its parental species may suggest a shift in mating system under colonization. The presence of low levels of selfing or biparental inbreeding in some weedy populations could be due to natural selection favoring self-compatibility. One of the advantages of self-compatibility is "reproductive assurance", especially in colonizing species (Baker 1955; Allard 1965). Environmental differences between parental species and colonial populations also may affect the mating system. Some

examples of shifts in mating system under colonization have been reviewed by Brown (1979) and Brown and Marshall (1981).

Population differentiation and gene flow

The total allozyme diversity found in the weedy rye populations was high (averaging 0.296 over polymorphic loci). This is comparable to outcrossing, wind-pollinated plant species, which average 0.293 ± 0.011 over polymorphic loci (Hamrick and Godt 1990). Much of this diversity was distributed within populations, and only about 3% among populations. This pattern of distribution of genetic variation in weedy rye populations suggest that: (1) little loss in the amount of genetic variation occurred in founding new colonies, (2) the fairly recent and rapid spreading of weedy rye has not allowed sufficient time for populations to diverge, and/or (3) high levels of gene flow exist among local populations.

The estimated value of *Nm* is 14.50 (private-alleles method) or 7.56 (G_{ST} method) in the weedy populations. These values are much higher than needed to prevent population differentiation caused by genetic drift or natural selection. In theory, the level of gene flow, $Nm > 1$, is sufficient to prevent population differentiation through the effect of genetic drift alone (Wright 1931; Maruyama 1970, 1972). If $m/s > 1$, gene flow could overcome the effect of natural selection (s) in favor of a locally adaptive allele (Haldane 1930; Nagylaki 1975; Slatkin 1985). A high level of gene flow was found between the cv 'Merced' and weedy rye, with an estimated *Nm* value of 8.21. 'Merced' has been widely used as a representative of cultivated rye in *S. cereale*, and also has been used as the putative parent of weedy rye in the comparative study conducted by Suneson et al. (1969). A close genetic similarity (0.99) has been found between 'Merced' and other cultivars of rye (Perez de la Vega and Allard 1984). Although the exact history of contacts between weedy rye and individual cultivars is not known, the evidence of marker gene introgressions (Suneson et al. 1969) does support the high estimated level of gene flow between them. Gene flow between *S. montanum* and the weedy populations may be purely historical, since *S. montanum* has not been found growing in the wild in California.

On the other hand, the high level of genetic similarity between *S. montanum* and *S. cereale* (Vences et al. 1987a, b; this study), which probably resulted from gene exchanges between the two species in areas of natural contact, may have given rise to the high genetic identities between weedy rye, cultivars, and *S. montanum*.

The present estimates of gene flow among the weedy rye populations could be upwardly biased since these populations may share common ancestry. This bias may, however, be insignificant. Slatkin (1985) pointed out that this situation would give a biased estimate of gene flow only if the time (generations) since colonization were less than the effective population size. Weedy rye has been present in Northern California since the 1930's (Suneson et al. 1969). If the effective population size of the weedy colony is no more than 50, we would not expect to see a high migration pattern in the absence of gene flow simply because of its co-ancestry. Given the characteristics of colonization, it is unlikely that the weedy rye has a large effective population size. The present estimates of *Nm* in the weedy rye are also comparable with the *Nm* values found in other outcrossing, wind-pollinated species (range from 5.3 to 37.8, Hamrick 1987).

Although the levels of allozyme variation within weedy populations are comparable with the morphological measures by Jain (1977), discordant patterns were found between allozyme and morphological differentiation among populations. Frequently, evolution at the morphological and allozyme levels is uncoupled. The significant interpopulation heterogeneity in morphological and quantitative traits in weedy rye reported by Jain (1977) is likely caused by strong local selection pressures on these traits. This notion is supported by this study since there was no evidence of significant genetic drift or differential amounts of introgression from the cultivar.

Colonizing success of weedy rye and its germplasm conservation

The colonizing success of weedy rye in Northern California can probably be attributed to the combination of the following factors. (1) Its hybrid origin and outcrossing breeding system maintained the high levels of genetic diversity present in the parental species. (2) Introgression with rye cultivars in the region may have further enriched its genetic variability. Suneson et al. (1969) postulated introgressions from at least 20 different rye cultivars since 1939. (3) Preservation of certain characters from the wild parent *S. montanum*, such as spike fragility, seed dormancy, and small seed size, may contribute to its ability to disperse and establish in wild habitats under the favorable environmental conditions in the area.

Further spreading of the weedy populations seems to be in progress. The unique genetic properties present in the weedy rye suggest its potential use as a crop germplasm resource. Based on the amount and distribution of genetic diversity in the weedy rye populations, we suggest that the best conservation strategy would be to collect from only two or three populations (e.g., *Rye3*, *Rye4*, and *Rye6* 1989) in the Fall River area, which has the highest allelic diversity. The rare alleles can be retained by increasing the number of individuals sampled within populations. *In situ* conservation can also be easily achieved because of the colonizing ability of weedy rye in this region.

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