magnetic resonance imaging. J. Amer. Soc. Hort. Sci. 118:92-96.
22. Webster, A. D. and M. S. Hollands. 1999. Orchard comparisons of Cox's Orange Pippin grown on selections of the apple rootstock M.9. J. Hort. Sci. and Biotech. 74:513-521.
23. Webster, T. and K. Tobutt. 1994. European and East Malling rootstock research-new and promising apple rootstocks. Compact Fruit Tree 27:17-24.
24. Wertheim, S. J. 1997. Useful differences in growth vigour between subclones of the apple rootstock M.9. Acta Hort. 451:121-128.

Journal American Pomological Society 55(2):100-108 2001

The Utilization of Related Prunus Species for Almond Variety Improvement

T. M. GRADZIEL,[1] P. MARTÍNEZ-GÓMEZ,[2] F DICENTA,[2] AND D. E. KESTER[1]

Abstract

Germplasm from a range of Prunus species in the subgenus Amygdalus, sections Euamygdalus, Spartoides, and Lyciodes has been evaluated for potential value to almond (*P. dulcis*) breeding through an international, U.S.-Spain cooperative project. Species evaluated include *Prunus argentia*, *P. bucharica*, *P. fenzliana*, *P. mira*, *P. persica*, *P. scoparia* and *P. webbii*. The absence of severe crossing barriers in the initial hybridization and in subsequent backcrosses demonstrates a direct accessibility of this rich germplasm to almond breeding. The performance of interspecific hybrids, as well as their subsequent backcrosses to cultivated almond, further demonstrate valuable opportunities for transferring useful traits including self-compatibility, resistance to important pests and diseases, the improvement of seed oil quality, tree growth architecture and bearing habit, and tolerance to aberrant environments. The international collaboration has allowed a more thorough evaluation of related germplasm and avoids quarantine restrictions on the U.S. importation of new Prunus accessions.

Introduction

Almond (*Prunus dulcis* Miller, syn. *P. amygdalus* Batsch, syn. *Amygdalus communis* L.) in the subgenus *Amygdalus*, section Euamygdalus is an important tree crop of worldwide distribution (38). In 1999, California production was estimated at over 800 million pounds from plantings of approximately 500,000 acres. Spain, with the second largest commercial production has plantings of over 1,480,000 acres, but with the total production of less than 200 million pounds due to the widespread practice of low-input, dryland agriculture. The cultivated almond is thought to have originated in the arid mountainous regions of Central Asia (Fig. 1) (19, 31). Several wild species are also found in these mountainous areas, from the Tian Shan mountainous in western China, through Afghanistan, Kurdistan, and Turkestan, and into Iran and Iraq (5, 18, 19, 20, 29). The *Prunus* species *P. fenzliana*, *P. bucharica* and *P. argentia* (of the Section Euamygdalus) from these regions are described as the wild species most closely related to almond (5, 19, 20), and may be the ancestral species of the modern cultivated almond (29). Ladizinsky (31), however, identifies only *P. fenzliana* as the wild ancestor of almond. *P. webbii*, which is thought to have originated on the Balkan peninsula, and *P. persica* and *P. mira* from China are also described as closely related to almond (19, 20, 29). The limited genetic base for cultivated almond has been well documented using molecular markers (3), distribution of self-incompatibility alleles (27) and related genetic studies (32). Environmental limitations, particularly almond's vulnerability to frosts during its early spring flowering, and its susceptibility to foliar diseases under humid production conditions, have limited its cultivation to geographicareas with the characteristic Mediterranean type climate of moderate winter temperatures and low summer rainfall.

While inter-specific gene transfer has

[1]Department of Pomology, University of California, Davis, CA, 95616, USA.
[2]Departamento de Mejora y Patología Vegetal, CEBAS-CSIC, PO Box 4158, Murcia, Spain.

proven a valuable breeding tool for both the agronomic and vegetable crops (2, 22), this approach has not been widely utilized in tree crop improvement (30). In addition, quarantine restrictions to the international exchange of *Prunus* germplasm have been imposed to limit the spread of destructive pests and diseases such as the plum pox potyvirus. The present study is part of a long-term international cooperation between breeding programs in Davis, California and Spain. This program has three primary objectives: a) the preliminary assessment of the potential value of interspecific introgressed germplasm to the improvement of cultivated almond, b) the characterization of available genetic diversity in cultivated almond and related species, and c) the identification of specific germplasm with disease and/or insect resistance, for use in almond breeding. This study presents a preliminary assessment of the opportunities for the introgression of interspecific germplasm to cultivated almond.

Materials and Methods

Barriers to interspecific gene introgression were assessed by examining the level of pollen viability in interspecies hybrids and subsequent backcrosses to almond. Traits evaluated for their value to commercial almond improvement included self-compatibility, seed oil quality, and tree and kernel characteristics.

Pollen viability in interspecies hybrids and backcrosses. The level of pollen viability was evaluated in interspecific hybrids between almond and *P. persica* (32 selections) and their F_2 progeny (83 selections), and interspecific hybrids between almond and *P. webbii* (7 selections) and progeny resulting from their backcross to almond (21 selections). Anthers were harvested at the popcorn stage of bud development. Anther-sacs were collected and dried using standard methods (23, 25). Dehisced pollen was germinated on 0.8 % agar containing 12% sucrose, and the proportion of germinating pollen grains recorded after 8 hours at 25 degrees C. Pollen germination was considered to have occurred when the pollen-tube was at least twice the length of the pollen grain diameter.

Assessment of self-compatibility in backcross progeny from interspecific hybrids. Genotypes tested included backcross progeny from interspecific hybrids between almond and *P. argentia* (1 selections), *P. fenzliana* (2 selections), *P. mira* (1 selections), *P. persica* (4 selections), and *P. webbii* (6 selections). Trees were selected at fourth to eighth leaf for desired tree and nut shape before testing for self-compatibility. Self-compatibility was tested by bagging individual branches with insect-proof, mesh bags. At anthesis, all open flowers on bagged branches were hand pollinated with self-pollen. Viability of self-pollen was verified through in-vitro germination as described above. Each self-pollination was repeated at least twice on alternate days to allow for a more complete pollination of late opening flowers. Fruit set was recorded at least 12 weeks following pollination to allow for late drop of abortive nuts. Self-sets greater than 15% were considered self-compatible based on earlier studies of self-compatible almonds (15, 27, 39). Selfing tests were repeated over 3 years. Fisher's least significant difference (LSD) was calculated using SAS (36) for comparison of genotype effects.

Evaluation of seed oil quality in backcrosses from interspecific hybrids. Total fat content and fatty acid methyl esters were determined for 5 almond selections derived from backcrosses of almond x *P. persica* and *P. mira* hybrids, and 7 cultivars important to California production ('Nonpareil,' 'Mission,' 'Carmel,' 'Ne Plus Ultra,' 'Peerless,' 'Sonora,' and 'Butte'), using a modification of the procedure of Garces and Mancha (13) as reported previously (1). Three separate replicates of each genotype were tested and Fisher's least significant difference (LSD) was used for comparison of genotype effects.

Evaluation of tree and kernel characteristics. Selections resulting from the backcross to almond of interspecific hybrids with *P. argentia* (5 selections), *P. bucharica* (2 selections), *P. fenzliana* (9 selections), *P. mira* (1 selection), *P. persi-*

ca (11 selections), *P. scoparia* (2 selections), and *P. webbii* (14 selections) which exhibited levels of self-compatibility and/or promising agronomic characteristics were propagated on 'Nemaguard' rootstocks, and planted at test orchards at Winters, CA. Data was collected from sixth leaf trees for time of bloom, the percent of total nuts exhibiting a complete shell seal, the crack-out ratio of kernel meat weight to whole nut (kernel meat plus shell) weight, kernel weight, and crop size relative to 'Nonpareil' trees interplanted as standards. Genetic differences between progeny groups were analyzed by ANOVA (36) using data which was similarly collected from the self-incompatible cultivar 'Nonpareil' and the partially self-compatible cultivar 'LeGrand' for comparison.

Results

Pollen fertility in interspecific hybrids and backcrosses. All interspecific hybrids showed moderate to high levels of pollen viability, demonstrating the absence of any significant hybrid sterility for these crosses. The level of pollen viability ranged 22 to 68 percent for hybrids between almond and *P. persica*, and 31 to 62 percent for hybrids between almond and *P. webbii*. For comparison, germination of pollen from the adjacent 'Nonpareil' trees averaged 48 percent with a range from 29-65% using this germination test. Similarly, moderate to high levels of pollen viability were observed in all F_2 and backcross progeny, demonstrating the absence of hybrid breakdown in these crosses. Pollen germination ranged from 33 to 58 percent in F_2 progeny from almond times *P. persica* parents and from 27 to 72 percent in backcross from almond x *P. webbii*.

Assessment of self-compatibility in backcross progeny from interspecific hybridizations. Seed set following controlled self pollinations in backcross (BC) and selfed (F_2) progeny ranged from 3.3 to 43.9 percent for the genotypes tested (Table 1). All selections showed moderate to heavy (up to 54%) sets following open-pollinations. Part of the variation in self-sets observed was due to a generally low set in one of the test years due to poor weather conditions at flowering. The relatively low LSD and standard deviations observed, however, indicate that genetic differences were important determinants of final seed set. While all species tested showed some level of self-compatibility, only *Prunus* mira, *P. persica*, and *P. webbii* had fruit set above 30 percent, (though the latter two species also had greater representation in this evaluation). Breeding populations developed from these parents also segregate for self-compatibility in the expected Mendelian ratios for a single gene (see also 8, 37). The genotypes showing the highest selfing percentages, selection 13,25-75 (a BC_2 progeny from *P. mira*) and selection 13,28-21 (a BC_2 progeny from *P. persica*) also demonstrated a relatively high capacity for autogamy or the ability to autonomously self-pollinate. In one interspecific cross to *P. persica*, cleistogamous autogamy was observed as the dehiscence of the flower anthers prior to flower bud opening (17).

Evaluation of seed oil quality in backcrosses from interspecific hybrids. Total oil content for the genotypes tested ranged from 41.6 to 50.2 percent (Table 2). Be-

Table 1. Average seed set following self-pollinations of breeding selections derived from related *Prunus* species. (Sample standard deviations given in parenthesis).

Source species	Degree Introgression	Selection	Percent Seed Set
P. argentia	BC_1	7920-45	11.5 (1.4)
P. fenzliana	BC_1	7906-13	9.0 (1.3)
P. fenzliana	BC_1	7906-22	9.3 (1.6)
P. mira	BC_2	13,25-75	37.2 (3.5)
P. persica	F_2	10C,20-51	20.8 (1.8)
P. persica	BC_2	13,28-21	43.9 (2.1)
P. persica	BC_2	3,54-39E	36.1 (3.4)
P. persica	BC_1S_1	6,56-89	34.4 (2.4)
P. webbii	BC_1	7927-54	9.6 (0.9)
P. webbii	BC_1	8007-15	3.5 (1.0)
P. webbii	BC_1	8007-24	34.4 (2.6)
P. webbii	BC_1	8011-11	4.4 (1.6)
P. webbii	BC_1	8021-95	3.3 (0.9)
P. webbii	BC_1	8024-12	12.8 (1.1)
LSD			5.3

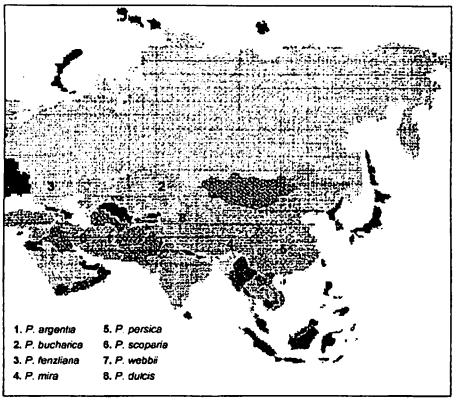

Figure 1. Map of Asia showing origins for *Prunus* species examined in this study. (Cultivated almond *P. dulcis* considered synonymous with wild *P. communis*).

cause of such high levels in almond, oil content and composition are important determinants of almond quality and commercial value. Selections derived from interspecies crosses were intermediate to this range, with the exception of selection 13,25-75 (a BC_2 progeny from *P. mira*) which had the lowest oil content. Selection 13,25-75 also showed the lowest proportions of oleic acid of the genotypes tested. In contrast, the remaining interspecies backcross progeny, which are derived from crosses of almond to *P. persica*, show uniformly high levels of oleic acid, being up to 16 percent higher than commercial almond cultivars. High proportions of oleic acid, a monounsaturated fat, are desirable as it has been associated with reduced susceptibility to kernel rancidity and improved health benefits to the consumer (1). In addition, these higher proportions of oleic acid do not appear associated with any sizable reduction in total oil content.

Evaluation of tree and kernel characteristics in backcrosses from interspecific hybrids. A wide range of tree and nut characteristics were observed among backcross progeny from different interspecies crosses (Table 3). Considerable variability was observed within species groups and significant differences were often observed both within and among species groups. Certain characteristics, however, were retained in many of the progeny from certain species groups. Genotypes derived from *P. argentia* tended to develop into small to medium-sized trees with an early bloom and a hard and well sealed shell. The good shell seal was found to confer resistance to kernel infestations by navel orangeworm (*Amyelois transitella*) and peach twig borer (*Anarsia lineatella*), the two major insect pests of almond in California. The well sealed shell was also associated with the undesirable characteristics of low kernel-to-nut crack-out ratios and smaller kernel sizes. Several progeny, however, possessed a thin, yet highly lignified shell composed primarily of well sealed inner endocarp (Fig. 2) which conferred both insect resistance and high crack-out ratios (up to 73%). Progeny of *P. bucharica* also showed good shell seal with relatively smooth, thin shells and moderate to good kernel crack-outs, a low kernel mass and later blooming but earlier ripening times. *P. bucharica* trees were large to very large. *P. fenzliana* progeny produced generally medium to large, vigorous and upright trees with good cropping potential on both spurs and terminal

Table 2. Almond kernel oil content and proportion of oleic acid for standard almond cultivars and backcrosses from almond x *P. persica*, and almond x *P. mira*. (Sample standard deviations given in parenthesis.)

Genotype	Total oil (% DW)	Oleic acid (% Oil)
13,36-52 (BC_3)	45.0 (2.0)	74.8 (2.4)
LeGrand (BC_3[z])	45.2 (6.2)	73.9 (2.6)
6,56-89 (BC_1S_1)	46.0 (3.3)	73.5 (2.4)
3,54-39E (BC_2)	45.3 (2.1)	73.4 (1.3)
Mission	49.6 (2.8)	71.1 (1.8)
Ne Plus Ultra	47.6 (3.4)	68.1 (2.9)
Nonpareil	43.6 (5.3)	67.4 (2.5)
Sonora	42.2 (2.5)	65.4 (4.4)
Peerless	41.6 (4.3)	65.2 (2.2)
Carmel	44.9 (4.8)	64.5 (2.9)
Butte	50.2 (3.5)	64.3 (1.6)
13,25-75 (BC_2)	43.9 (2.7)	62.9 (1.9)
LSD	2.5	1.9

[z]Lineage estimated from molecular marker data.

shoots, though with a generally poorer shell seal and moderate kernel crack-out ratios and kernel size. *P. mira*, which was represented by only 1 backcross selection, showed a medium-size tree with large crops forming on weepy (peach-like) branches with a late bloom, poor shell seal, and moderate nut size. Progeny from *P. persica*, a close relative to *P. mira*, demonstrated a wide range of variability. As with *P. mira*, and *P. bucharica*, a later blooming period allowed the avoidance of the damaging Spring frosts. Trees tended to be larger but a peachy or willowy growth habit in some genotypes suppressed upward growth and so ultimate cropping potential. Bloom was late and more susceptible to *Monilinia* flower blight (*Monilinia laxa*). Nuts were typically small and flat, often with thick well sealed shells. Resistance to aflatoxin production following *Aspergillus flavus* infection was also observed in these progeny (16). Backcross progeny from *P. scoparia* develop into small to medium sized, upright trees with well sealed though small and thick shelled nuts. Backcross progeny from *P. scoparia* also demonstrated the characteristic *P. scoparia* trait of small linear leaves which tended to abscise early in the growing season resulting in mostly green, willowy, branches where most photosynthesis presumably takes place. This ephemeral leaf trait has been reported to confer a high level of drought resistance (21). Both parent and progeny appear more susceptible to flower blights. The widest range in tree and kernel characteristics was observed in backcross progeny of almond x *P. webbii*, in part due to the larger numbers of selections examined. Considerable variability was observed in bloom period, crop size, shell seal, and shell and kernel size. Low levels of field damage by several important diseases including *Monilinia* flower blight, anthracnose (*Coletotrichum* spp.),

Table 3. Range of tree and nut characteristics of backcross selections resulting from interspecific introgressions. (Self-incompatible 'Nonpareil' and the self-compatible 'LeGrand' provided as references; S-small, M-medium, L-large).

Source	Selections Examined	Tree Size	Crop Size	Bloom Time	Percent Sealed[z]	Kernel-to-Nut Ratio	Kernel size (g)	Promising Resistences[y] and Horticultural Traits
P. argentia	5	S-M	M-L	Early	100	35-73	0.55-0.92	Resist. *Amy, Ana, Alt, Mon,* Self-compat.
P. bucharica	2	L	S-M	Late	100	38-61	0.53-0.73	Late flowering, Early maturity
P. fenzliana	9	M-L	L	Mid	14-100	28-49	0.67-0.94	Drought resist., Late flowering, Resist. *Alt, Ana*
P. mira	1	M	L	Late	8	42	0.86	Self-compat., Oil quality
P. persica	11	S-L	S-L	Late	11-100	31-48	0.47-0.85	Self-compat., Autogamy, Oil quality, Resist. *Asp*
P. scoparia	2	S-M	S-M	Mid	100	23-35	0.53-0.61	Drought resist., Resist. *Amy, Ana, Cap*
P. webbii	14	S-L	S-L	Mid	28-100	37-61	0.51-0.87	Resist. *Amy, Ana, Cap; Alt, Cap;* Self-compat.
P. dulcis	'Nonpareil'	L	L	Mid	8	63	1.19	
	'LeGrand'	L	L	Mid	4	56	1.03	

[z]Percent of nut sample without shell splits exposing the kernel.
[y]Insects: Cap-*Capnodis tenebrionis*, Amy-*Amyelois transitella*, Ana-*Anarsia lineatella*; Fungi: Alt-*Alternaria* spp., Asp-*Aspergillus* spp., Col-*Coletotrichum* spp. and Mon- *Monilinia* spp.

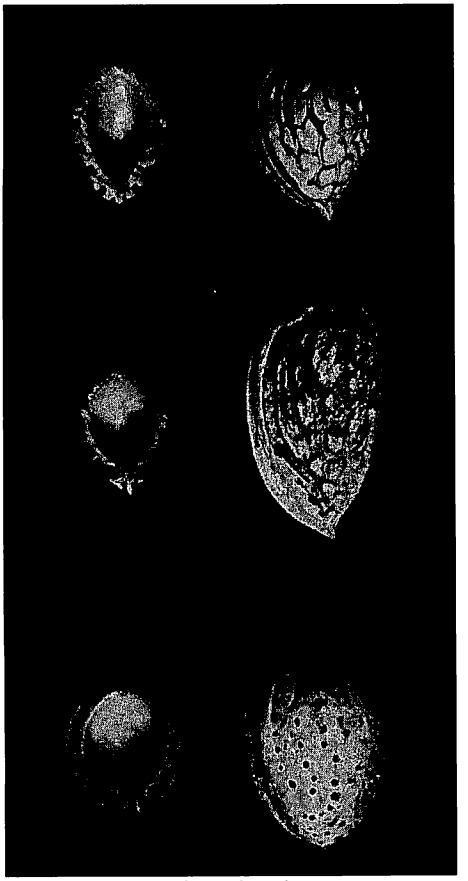

Figure 2. Illustration of endocarp outer surface and cross-section of shells of almond cv. Nonpareil and crosses to *P. argentia* comparing the structurally weaker 3-part (inner, vascular channels, and outer) endocarp of 'Nonpareil' with the more impervious argentia type endocarp where the vascular bundle lies outside of the endocarp. In backcross progeny, the single layer argentia-type endocarp is retained but sometimes as a thin but hard shell conferring both insect resistance and high crack-out ratios.ø

and California (navel orangeworm and peach twig borer) and Spanish insect pests (capnode, *Capnodis tenebrionis*) indicate promising levels of pest resistance in this germplasm. A high proportion of *P. webbii* progeny maintained the characteristic webbii branching habit of numerous current season laterals on new growth (Fig. 3). This ability to rapidly develop extensive fruit bearing wood appears to be a major contributor to the high relative cropping potential of several *P. webbii* selections compared to other selections and the standard almond cultivar 'Nonpareil' (Fig. 4). Extensive development of such current season laterals has also allowed rapid renewal (and so resistance to crop loss) of young bearing wood destroyed by anthracnose (*Colletotricum acutatum*) and Alternaria Leaf Spot (*Alternaria alternata*) in several advanced selections. In some progeny, however, these current season laterals produce only short thorn-liked spurs.

Discussion

A preliminary survey of backcross and F_2 progeny of *Prunus* species closely related to almond has identified a rich diversity in traits with potential value to commercial almond improvement. Relatively high levels of pollen fertility in these progeny support the absence of severe crossing barriers to the interspecific introgression of controlling genes. While molecular studies are now underway examining linkage drag in advanced interspecific breeding lines, a relatively high level of horticultural variability is tolerated in this nut crop. This is because of the large amount of phenotypic variation in tree and nut characteristics present in current varieties, and from the common horticultural practices such as pruning and tree hedging routinely used to modify tree growth and bearing habits in commercial orchards.

Somewhat surprisingly, the most promising sources of new genes may be the more distantly related species of *P. persica*, *P. mira* and *P. webbii*. Both *P. persica* and *P. mira* have been classified by some taxonomists in a separate genus or subgenus. For example Browicz and Zohary (5) have classified *P. persica* and *P. mira* in the genus *Prunus* while almond and the other 'almond-like' species discussed here were placed in the genus 'Amygdalus.' Rehder (33) has placed all of the species examined in this survey in the genus *Prunus*. *Prunus argentia*, *P. dulcis*, *P. bucharica*,

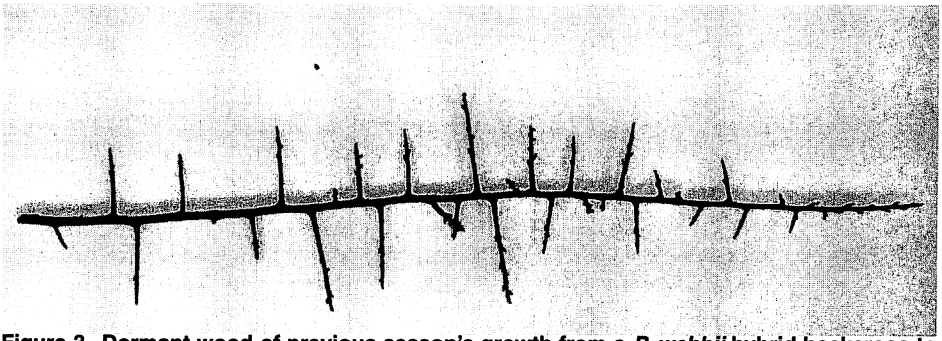

Figure 3. Dormant wood of previous season's growth from a *P. webbii* hybrid backcross to almond showing the retention of the webbii-type bearing habit where low apical dominance results in extensive development of highly productive current season lateral branches.

and *P. fenzliana*, are placed in the subgenus Amygdalus, section Euamygdalus, *P. scoparia* is placed in the section Spartiodes, and *P. webbii* placed in the section Lycioides.

While several earlier reports have discussed recovery of genes for self-compatibility from related species through either natural or controlled crosses (7, 8, 12, 15, 34, 37, 38), only Richter (35), Grasselly (18,20), Denisov (7), and Kester et al. (26) have previously reported on other possible uses of wild species to almond improvement. The historical use of these species and their hybrids as almond rootstocks should facilitate future introgressions. The use of wild species directly as a rootstock for dryland almond has been widely re-

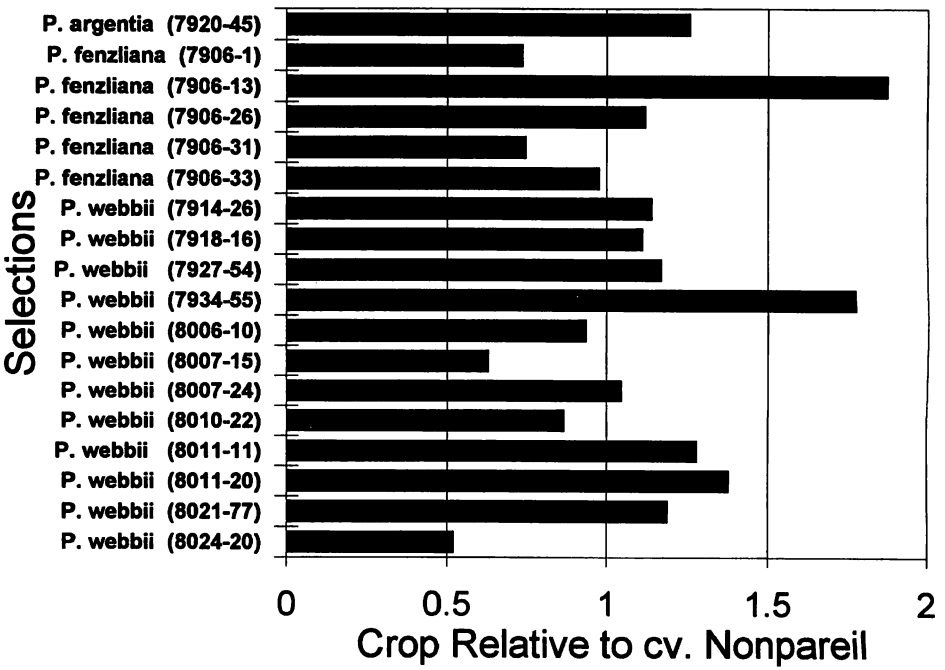

Figure 4. Tree nut production relative to the cv. 'Nonpareil' of selected backcrosses to almond of interspecies hybrids ('Nonpareil' = 1).

ported, including *P. spartioides* in Iran (14), *P. bucharica* (10, 24) and *P. fenzliana* in Russia (6), *P. webbii* in Turkey (9), and *P. fenzliana*, *P. bucharica*, *P. kuramica*, *P. argentea* (18) and *P. dehiscens* and *P. kotschyi* (21) at lower incidence in these and nearby areas. Crosses between almond and related species have been readily achieved under controlled conditions (4, 12, 15, 18, 21, 28, 29). Interspecific crosses between these related species (mainly *P. persica* x almond but also *P. webbii* x almond) have been used for almond rootstock breeding in France (4), USA (28), Spain (11) and Yugoslavia (40). In addition, Browicz and Zohary (5) and Ladizinsky (31) have recently reviewed evidence for a high level of spontaneous interspecific hybridization in the wild between species with overlapping ranges.

The limited germplasm tested, while demonstrating exciting opportunities for almond improvement, probably represent only a small fraction of its full breeding potential. The recent and often dramatic losses of conventional agro-chemical based management options for pest and soil water management have led to an urgent need for new genetic solutions. The exotic origins of this breeding material may offer the needed genetic diversity to solve such problems. This exotic nature, in turn, will require the development of very efficient selection strategies for its timely incorporation into new varieties. Molecular studies of this material are presently underway to better characterize the degree of genetic diversity, as well as develop markers to accelerate promising gene introgression.

Acknowledgment

The authors acknowledge the support of Spanish Ministry of Education and Culture in the stay of Pedro Martínez-Gómez at the Department of Pomology, University of California, Davis, CA.

Literature Cited

1. Abdallah, A., M. H. Ahumada and T. M. Gradziel. 1998. Oil content and fatty acid composition of almond kernels from different genotypes and from different California production regions. J. Amer. Soc. Hort. Sci. 123:1029-1033.
2. Allard, R. W. 1960. Principles of Plant Breeding. Wiley Press, N.Y. Pp. 485.
3. Bartolozzi, F., M. L. Warburton, S. Arulsekar, and T. M. Gradziel. 1998. Genetic characterization and relatedness among California almond cultivars and breeding lines detected by randomly amplified polymorphic DNA (RAPD) analysis. J. Amer. Soc. Hort. Sci. 123(3):381-387.
4. Bernhard, R. 1949. The peach-almond and its utilization. Revue Horticole. 121:97-101.
5. Browicz, K., and D. Zohary. 1996. The genus *Amygdalus* L. (Rosaceae): Species relationships, distribution and evolution under domestication. Genetic Resources and Crop Evolution. 43: 229-247.
6. Denisov, V. P. 1980. Drought resistance of some almond species in Turkmenistan. Trudy Prikladnot Bot., Genet., Selk. 67(1):129-135.
7. Denisov, V. P. 1988. Almond genetic resources in the USSR and their use in production and breeding. Acta Hort. 224:299-306.
8. Dicenta, F. and J. E. Garcia. 1995. Inheritance of self-compatibility in almond. Heredity 70:313-317.
9. Dimitrovski, T. and B. Ristevski. 1973. Ispitivanje podonosti divijeg Badena Amygdalus webii Sprach. Yugosl. Vocarstvo Brez. 23:15-21.
10. Evreinoff, A. 1952. Quelques observations biologiques sur l'amandier. Rev. Int. Bot. App. Agr. Tropical. 32:442-459.
11. Felipe, A. J. 1975. F_1 hybrids of peach and almond trees as a model for both species. Agricultura. 44: 661-663.
12. Felipe, A.J. 1984. Etat de l'arboretum des espèces sauvages d'amandier a Saragose. Options méditerranéennes. 84/I:203-204.
13. Garces, R. and M. Mancha. 1993. One-step lipid extraction and fatty acid methyl esters preparation from fresh plant tissue. Anal. Biochem. 211:139-143.
14. Gentry, H. S. 1956. Almond culture in southern Iran. Almond Facts. 21:6-7.
15. Gradziel and D. E. Kester. 1998. Breeding for self-fertility in California almond cultivars. Acta Horticulturae. 470:109-117.
16. Gradziel, T. M., N. Mahoney & A. Abdallah. 2000. Aflatoxin Production Among Almond Genotypes is Not Related to Either Kernel Oil Composition or *Aspergillus flavus* Growth Rate. HortScience 34:937-939.
17. Gradziel, T. M. and S. A. Weinbaum. 1999. High relative humidity reduces anther dehiscence in apricot, peach and almond. HortScience 34:322-325.
18. Grasselly, C. 1975. L'arboretum amygdalus. 2 Colloque du groupe de recherche et d'étude meditarranéen pour l'amandier. 1-2.

19. Grasselly, C. 1976a. Origine et évolution de l'amandier cultivé. Options méditerranéennes. 32:45-50.
20. Grasselly, C. 1976b. Les espèces sauvages d'amandier. Options méditerranéennes. 32:28-44.
21. Grasselly, C. 1992. Considerazioni sulle caratteistique di alcune specie selvatiche di mandorlo e sulle loro possibilita di utilizzazione in programmi di miglioramento genetico. Progr. de recherche Agrimed. Nimes (France), June, 1990. pp. 135-144.
22. Hadley, M. M. and M. Openshaw. 1980. Interspecific and intergeneric hybridization. In (W. R. Fent and M. M. Hadley, eds.) Hybridization of Crop Plants. Am. Soc. Agron. Pp. 133-160.
23. Hill, S. J., D. W. Stevenson, and B. K. Taylor. 1985. Almond pollination studies: pollen production and viability, flower emergence and cross-compatibility tests. Aust. J. Expt. Agr. 25:697-704.
24. Jadrov, D. A. 1970. Porte-Greffes de l'amandier. Sadovostvo. 11:32.
25. Kester, D. E. and T. M. Gradziel. 1996. Almonds. In: Fruit breeding. J. Janick and J. N. Moore (eds.). New York (USA).
26. Kester, D. E., T. M. Gradziel, and C. Grasselly. 1991. Almonds. In: Genetic resources of temperate fruit and nut crops. J. N. Moore and J. R. Ballington (eds.). Wageningen (Netherlands). pp. 701-758.
27. Kester, D. E., T. M. Gradziel & W. C. Micke, 1994. Identifying pollen incompatibility groups in California almond cultivars. Journal of the American Society for Horticultural Science 119:106-109.
28. Kester, D. E. and C. J. Hansen. 1966. Rootstock potentialities of F_1 hybrids between peach (*Prunus persica* L.) and almond (*Prunus amygdalus* Batsch.). Am. Soc. Hort. Scince. 89:100-109.
29. Kester, D. E., K. Pelletreu,, L. Liu, and R. N. Asay. 1988. Interspecific hybridization for culture improvement in almond. Proc. Int. Symp. of Hort. Germplasm, cultivated and wild. Beijing, China.
30. Layne, R. E. C. 1985. Hybridization. In J. N. Moore and J. Janick (eds.) Methods in Fruit Breeding. Purdue University Press. Pp. 48-65.
31. Ladizinsky, G. 1999. On the origin of almond. Genetic Resources and Crop Evolution. 46:143-147.
32. Lansari, A., D. E. Kester and A. F. Iezzoni. 1994. Inbreeding, coancestry, and founding clones of almonds of California, Mediterranean shores and Russia. J. Am. Soc. Hort. Sci. 119:1279-1285.
33. Rehder, A. 1967. Manual of cultivated trees and shrubs. 2nd. ed. Macmillan, N.Y.
34. Reina, A., V. Giorgio, and A. Godini. 1985. Autres types autocompatibles parmi la population d'amandiers des Pouilles. Options méditerranéennes. 85/I:25-29.
35. Rickter, A. A. 1972. L'amandier. Academie des Sciences Agricoles. Jardin Botanique de Nikits. Yalta.
36. SAS Institute. 1988. SAS/STAT users guide. Release 6.03. SAS Institute, Cary, NC.
37. Socias i Company, R., and A. J. Felipe. 1988. Self-compatibility in almond: Transmission and recent advances. Acta Horticulturae. 224:307-317.
38. Socias i Company, R., and Felipe, A.J. 1992. Almond: A diverse germplasm. HortScience. 27 (7): 718, 863.
39. Socias i Company, R., D. E. Kester and M. E. Bradley. 1976. Effects of temperature and genotype on pollen tube growth in some self-incompatible and self-compatible almond cultivars. J. Am. Soc. Hort. Sci. 101:490-493.
40. Vlasic, A. 1977. L'*amygdalus webbi* Spach ed I suolsi ibridi col pesco com portaninnestro del mandorlo. 3d Colloque GREMPA, Bari, Italy: 80-81.