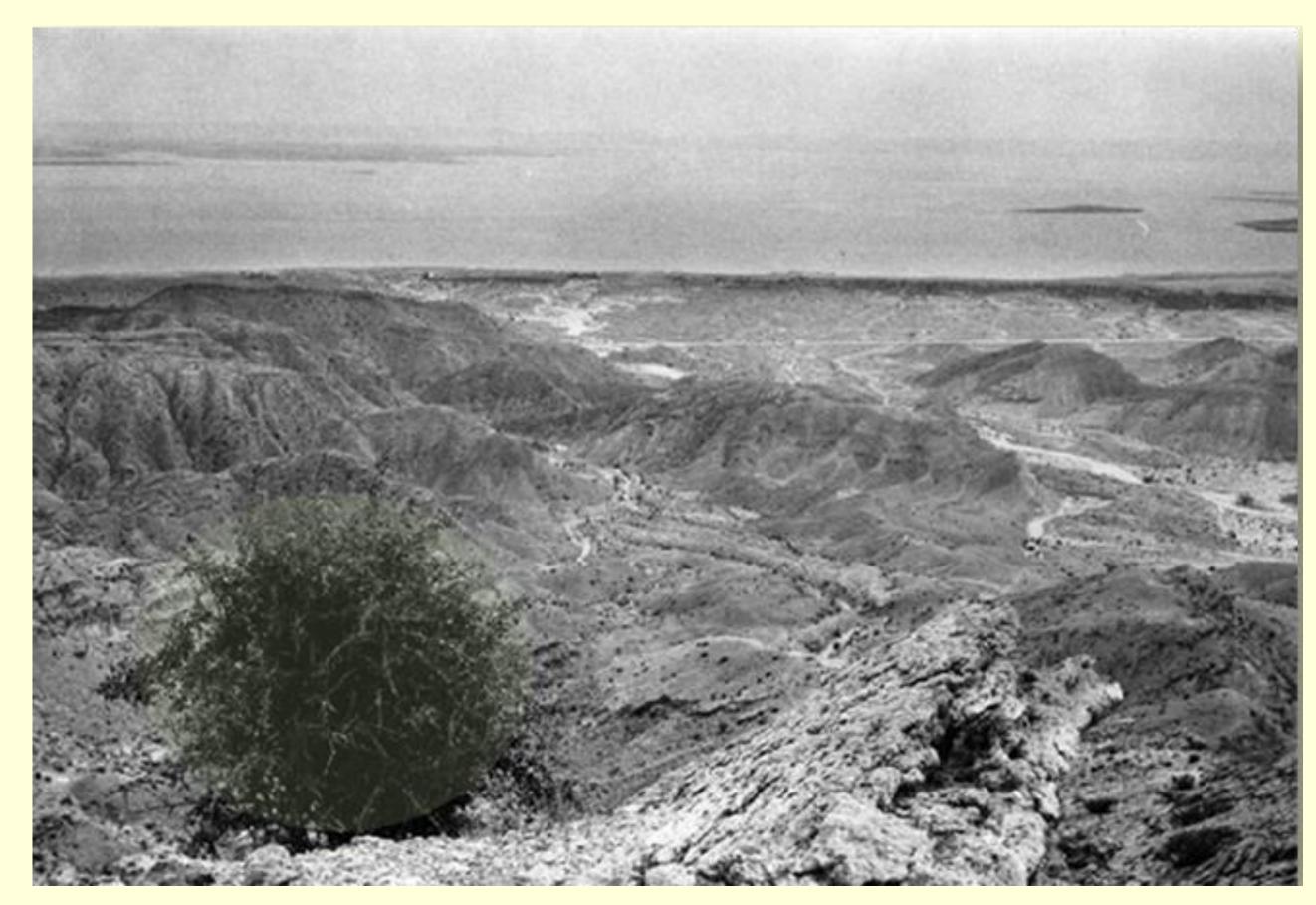
Interspecific Breeding Germplasm for **Rootstock Research and Development** 

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Plant Sciences, University of California/ Davis Location:



Prunus tangutica from Central Asia

## Introduction

Changes in planting practices, including the quantity and quality of land and irrigation water, have led to the need for a new generation of almond rootstocks and a number of public and private efforts have been initiated to develop and test new candidates. Germplasm derived from other species, either directly or through species hybridization, is often pursued to attain

the greatest range of vigor, stress and disease tolerance and desirable horticultural traits. However, the acquisition of such exotic germplasm is often difficult and time-consuming. At UC Davis, a diverse germplasm has already been developed combining almond, peach as well as related species including P. argentea, P. bucharica, P. davidiana, P. fenzliana, P. domestica, P. mira, P. orthosepala, P. scoparia, P. tangutica and P. webbii. In this project, selected germplasm from the UCD almond and peach breeding programs with value for rootstock improvement have been characterized both phenotypically and at the molecular marker level. Detailed pedigree relationships as well as trait expression data have been transferred to the RosBreed web-site (http://www.rosbreed.org/) and published to allow end-user analysis. This germplasm demonstrates extensive diversity both genetically and in the range of traits useful to rootstock improvement programs. Field evaluations have identified unique and potentially useful characteristics in parents and progeny, including modification of tree architecture from *P. scoparia* hybrid rootstocks, drought tolerance in a peach by *P. argentea* hybrid, and a pronounced invigoration of scion growth in advanced interspecies lines.

			(%)		Kernel	Nut	Kernel	Nut	Kemel	Nut	Kemel	Soluble	R-		
No.	Genotype	Origin	Almond genome	Length (mm)	Length (mm)	Width (mm)	Width (mm)	Thicknes s (mm)	Thicknes s (mm)	Mass (g)		protein (g/100g)	ELISA	R-WB	R-DB
	A7-23	P. argentea (bitter seed)	0	19.03	-	15.25				1.47	0.37	17.28	0.61	0.64	0.61
	A7-25	P. webbii (bitter seed)	0	28.97	the subscription of the local division of th		11.75				0.82	19.09			
	F10D,2-14	P. fenzliana (F2)	0	30.55	-						1.03	19.21			-
	Andross	Peach (P. persica) (bitter seed)	0	35.31							0.36				
	A10-4	P. bucharica (bitter seed)	0	Contraction of the			_	-	4.66	-	0.21	20.94	and the second division of the second divisio	-	
87	A7-28	P. webbii (bitter seed)	0	25.72		14.13		and the second se	6.3		0.49	21.04		1.11	
101	F10D,2-12	P. fenzliana (F2)	0	26.49	20.61	16.11	10.78	11.51	7.04	1.41	0.77	21.38	1.53	1.06	1.44
71	P11-58	P. mira (bitter seed)	0	26.55	14.48				4.33	2.48	0.29	23.39	0.53	0.79	0.83
76	A13-1	P. persica $\times$ P. davidiana (bitter seed)	0	21.47	13.77	20.69	11.41	17.83	6.1	3.83	0.46	23.41	0.45	0.5	0.9
51	40A-17	Peach (P. persica) (bitter seed)	0	24.25	13.35	16.83	7.18	12.45	3.35	1.81	0.11	23.74	0.51	0.52	1.02
106	A2-11	P. tangutica (bitter seed)	0	16.54	13.36			12.4	8.28	1.34	0.49	25.44	0.7	0.94	0.87
88	F5,4-42	P. webbii (F2)	0	26.82	18.54	14.98	9.46	10.83	6.7	1.96	0.55	25.8	0.64	1.1	1.06
5	F5,4-10	P. webbii $\times$ (Nonpareil $\times$ P. persica ) BC1	38	27.5	19.69	18.32	11.94	12.78	7.22	2.69	0.78	22.12	0.53	1.02	0.96
77	Hansen2168	Almond $\times P$ . persica	50	44.06	27.95	28.46	15.71	18.29	7.34	9.07	1.44	12.35	1.57	0.81	1.31
97	F10D,3-24	P. webbii (BC1)	50	25.71	19.33	19.52	13.23	13.29	6.13	2.66	0.71	13.39	1.27	0.95	1.16
79	Nickels	Almond $\times P$ . persica	50	36.88	23.87	28.7	16.37	20.85	8.75	9.18	1.53	13.79	0.75		
104	F10D,3-50	P. fenzliana (BC1)	50	36.2	27.32	19.3	13.93	13.31	8.75	2.37	1.59	15.37	2.18	0.73	0.91
96	F10D,3-13	P. webbii (BC1)	50	25.39	19.4	19.08	12.02	13.66		1.85	0.83	17.07	0.47	0.57	0.87
100	F10D,3-3	P. argentea (BC1)	50	29.57	23.42					1.88	0.96	17.47			
93	F10D,3-2	P. webbii (BC1)	50	30.57	19.71	17.83	11.09	13.64	6.99	1.53	0.77	17.84	0.66	0.51	1.09
	F10D,2-5	P. webbii (BC1)	50						8.07	1.23	0.76	17.99		0.75	-
		P. webbii (F2BC1)	50		and the owner where the party of the local division of the local d	The subscription of the local division of th		Name and Address of the Owner water or other	The second se		0.96	18.58	_	-	
		(Nonpareil × P. persica ) F2	50	a second second second							1.08	19.32			
		P. webbii (BC1)	50						7.15	1.59	0.84	20.4			
		P. webbii (BC1)	50						7.57	1.94	0.95	20.5			
		P. webbii (F2BC1)	50							2.45	0.97	21.05			-
		Almond $\times P$ . persica	50								1.12	21.06			0.77
		P. webbii (BC1)	50						7.45		0.93	21.17			
		(Nonpareil × P. persica) F2 (bitter seed)	50							2.43	1.1	23.87			
	F5,16-60	(Mission × P. argentea ) F2	50					11.9		1.56	0.87	24.08			
		Padre × F5,4-4	69	27.45		-			and the second division of the second divisio	2.32	0.84	14.48		-	_
	F5,20-42	Padre × F5,4-10	69			17.85					1	16.72			
		Nonpareil × F5,4-4	69			24.82					1.11				
	F8N,7-4	F5,4-10 × Sonora				16.12						19.52			
	8010-22	Nonpareil × F5,4-10				19.31				-		21.06			
		Nonpareil × F5,4-10		NA		NA	12.54			NA	1.17				
		Nonpareil × F5,4-4 (see No. 4)				17.53									
	F8N,6-68	F5,4-10 × Solano				19.93						23.47		-	
	F10D,3-7	Almond $\times P$ . webbii $\times P$ . persica (BC2)				16.6									-
	97,1-232	SB13,25-75 × Winters (see No. 55)		NA		NA	13.42		or the owner of the owner of the owner of the owner.	NA	1.29	the second s			
	F5,13-54	(Mission × P. fenzliana) BC1 × Sonora				19.52						16.28			
	F5,10-9	(Mission × P. fenzliana) BC1 × Sonora		A Real Property lies and the second se	the second se	18.82	the second se	and the second data was not second as a second s			the second se	18.11		the second se	
	F10D,2-3	(Mission × P. fenzliana) BC1 × Sonora				20.09									-
	F5,6-13	(Mission × P. fenzliana) BC1 × Sonora				17.25									
	F5,6-1	(Mission × P. fenzliana) BC1 × Sonora				23.68				5.08		25.88			-
	97,3-40	Almond × P. webbii × P. persica (BC3)		NA			15.14			NA 2 15		25.31			
	2004,9-1	Nonpareil × 97,1-232				23.78						14.54			
		Nonpareil × 97,1-232				21.45						15.81			
		Nonpareil × 97,1-232 (see No. 72)				22.52			the subscription of the local division of th	2.96		19.84	the second se	1.15	
		(Nonpareil × P. persica) BC3				15.83									
	97,2-240	P. webbii (BC4)		NA 27.00		NA 26.46	12.61		9.45			22.22		0.61	
		(P. persica) BC4								7.31		23.91			-
	2000,2-3	Almond × P. webbii × P. persica (BC4)		NA		NA	11.62			NA		19.89			
	2000,8-27	Almond × P. webbii × P. persica (BC4)		NA 38.43		NA 27.67	12.13			NA 5.45		23.92			
	Tuono	Almond variety		38.43 NA		27.67 NA	13.12			5.45 NA		17.14			
	2004,18-20 Mission	Almond variety				NA 19.79				NA 2.55		18.72			
		Almond variety				23.06				-		19.17			-
	Ferragnes 95.1-26	Almond variety Almond variety		30.35 NA		23.00 NA	14.18		9.47		_	19.37 20.94			
	Sonora	Almond variety Almond variety				NA 18.89						22.07			
	Marcona	Almond variety	and the second se	and the second se		25.83	and the local division in which the local division in which the local division is not the local division in which the	and the second division of the second divisio	The subscription of the local division of th	5.55	successive statements where the second statement is not second as a second statement of the second sta	22.22			
	Winters	Almond variety				19.25								-	-
		Almond variety Almond variety		30.41 NAc	20.33		13.49			NA NA	1.31	and the second se			1.5
	Sweetheart	Almond variety Almond variety				18.98			-			25.52			1.16
	Kahl	Almond variety Almond variety				17.03						25.52			
						17.03	and the second data in the secon	Statement of the local division in the local		2.02				and the second se	The subscription of the local division of th
02	Chips	Almond variety	100	28.00	21.51	19.45	12.08	14./1	8.18	2.02	1.00	20.40	1.08	0.91	1.1/

 
 Table 1. UCD breeding germplasm
parents and introgression lines analyzed for seed characteristics. (A common introgression line would involve the transfer of the desired trait such as self-fruitfulness or nematode resistance from the donor species to the cultivated crop through a series of recurrent backcrosses to the target species). Color shading allows visualization of the relative differences within each trait (column) with red being low to green being high. A large range in phenotypic expression for each trait is available in parent species with much of this diversity successfully transferred through multiple backcrosses to recipient breeding line supporting good heritabilities and so opportunities for future breeding progress.

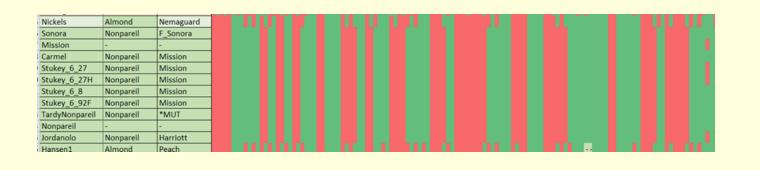
Traits	Cooperator	Germplasm evaluated	<b>Species</b> P.argentea, P. bucharica, P.davidiana, P.mira, P. persica, Plum spp., P. scoparia, P. tangutica, P. webbii.					
Compatibility. Nematode (ring and root knot) resistance	Burchell Nursery	50 clones						
Disease & Nematode resistance, Replant	Sierra Gold Nursery	20 clones & ~1000 seed	P.argentea, P. davidiana, P.mira, P. persica, P. scoparia, P. tangutica, P. webbii.					
Productivity, Compatibility, Architecture	Fowler Nursery/Paramount	5 clones	P.argentea, P. davidiana, P.mira, P. persica, P. scoparia					
Disease/pest resistance, hybrid vigor	UCD scion breeding	50 clones & ~4000 seedlings	P.argentea, P. bucharica, P.davidiana, P.mira, P. persica, Plum spp., P. scoparia, P. tangutica, P. webbii.					
Productivity	Mathew Gilbert	15 clones	P.argentea, P. fenzliana, P.mira, P. persica, P. webbii.					
Botryophaeria resistance	Jose Chaparro (UF)	40 clones	P.argentea, P. bucharica, P.fenzliana, P.mira, P. pedunculata, P. persica, Plum spp., P. tangutica, P. triloba, P. webbii.					
Root lesion and root-knot nematode resistance	Andreas Westphal	7 clones	P.argentea, P. davidiana, P.mira, P. persica, P. tangutica, P. webbii.					
Phytophthora resistance	Greg Browne	3 clones & ~200 seedlings	Plum interspecies, Almond spp. Seedlings & seed					
Crown gall resistance	Dan Kluepfel	200 seedlings & 400 seed	P. persica x P. tangutica (F <sub>2</sub> , F <sub>3</sub> )					



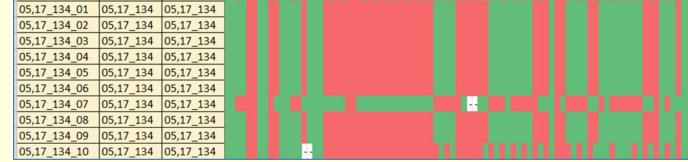
**Table 2.** List of current germplasm and cooperators pursuing: a) large-scale field evaluations and trait validations (blue) and b) gene discovery including mechanisms of gene action (gold).

## Approach

The genetic characterization of parent species, species-hybrids and subsequent breeding progeny has been completed using over 500 molecular markers showing a fairly uniform distribution over all eight of the Prunus chromosomes. While rootstock improvement has benefited from the increased understanding of both the extensive genetic as well as phenotypic (disease/stress resistance, etc.) variability available in almond and its related species, traditionally breeding remains very tedious and time-consuming. The use of molecular markers is increasingly being used to improve breeding efficiency by allowing geneticist to identify genotypes and so traits of interest at much earlier stages in the breeding program. These molecular markers have frequently been shown to be species-specific, which confounds their utility when wide species crosses or divergent species introgression is attempted. To evaluate molecular marker cross-species efficacy, genetically diverse Rootstock Breeding lines were evaluated using available RosBreed markers to determine whether markers were able to detect genetic differences in this highly variable germplasm.



**Figure 1**. Representative molecular marker plots for different almond cultivars and genotypes as compared to the peach by almond hybrids Nickels and Hansen showing failure of the markers to discriminate among the known heterozygotes in almond. (Note that some markers on the right side of the plot were able to be discriminated, but these were rare).



_104	05,11_	134															
134	05,17_1	134															
134	05,17_1	134															
134	05,17_1	134															
134	05,17_1	134															
134	05,17_1	134			• •												
Par	rent2	ST ST ST S	si si <mark>si</mark>	SI SI SI	sr SI sr S	ST ST ST S	SI SI SI SI	SI SI sr SI	ST ST ST SI	SISIS	I SI SI SI	ST ST ST ST	SI SI SI SI S	I SI SI S	I SI SI	SI SI S	I SI
F_1	18_8_11																
P_1	Tang																

Figure 3. Molecular marker inheritance pattern of peach (top row), a peach by *Prunus* tangutica interspecies hybrid (2nd row) with lower rows showing patterns for 6 F2 progeny (selfs of the species hybrid) showing that distortion from expected inheritance patterns differs with different species introgression lines.

Figure 2. Results from a almond by peach hybrid introgressed to peach, where parents

are highly heterozygous but subsequent introgression progeny show a very distorted

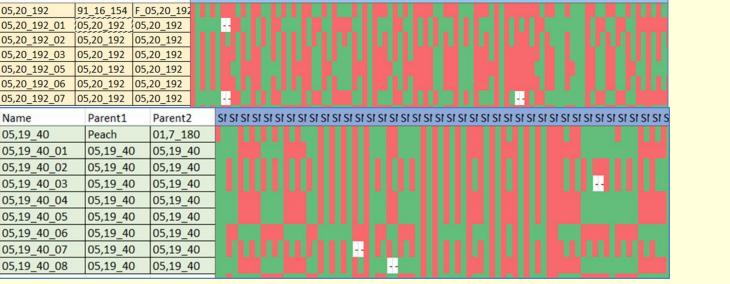
Figure 4. Molecular marker inheritance patterns of primal genotypes derived from

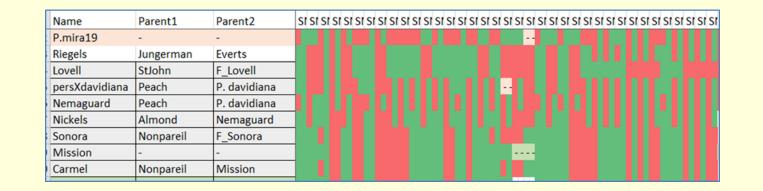
progeny, respectively, showing largely normal inheritance patterns.

advanced almond to peach introgression lines (05, 20-192 at top) in *Prunus argentea* to

peach introgression lines (05, 19-40 at bottom) as well as patterns for 7 and 8 selfed

inheritance often depending upon individual genotype.





**Figure 5**. Representative molecular marker patterns from chromosome 1 for different species and species hybrids, showing the value of general trends in identifying possible parentage. The pattern for Nemaguard shows a greater similarity between an established (peach x P davidiana) interspecies hybrid than either almond or Prunus mira, supporting the general perception that *Prunus davidiana* was a parent of Nemaguard and the probable donor of Rootknot nematode resistance.

## Summary

In much of this diverse germplasm, molecular markers developed for peach showed varying levels of efficacy when applied to species hybrids and their introgression lines. Despite a frequent failure of molecular markers developed in one species to discriminate the genetic alleles of different species, the use of marker-assisted-breeding proved valuable for understanding general inheritance trends as well as identifying possible barriers as well as potential novel opportunities for exotic gene transfer for rootstock improvement.