Interspecific Breeding Germplasm for Rootstock Research and Development

Project No.: **14-HORT10-Gradziel**

Project Leader: Tom Gradziel
Department of Plant Sciences
UC Davis
One Shields Ave.
Davis, CA 95616
530.752.1575
TMGRADZIEL@UCDAVIS.EDU

Project Cooperators and Personnel:
B. Lampinen, S. Marchand, J. Fresnedo, S. Overstreet, C. Crisosto, S. Metcalf, M. Gilbert, UC Davis
J. Adaskaveg, Plant Pathology, UC Riverside
G. Browne, Plant Pathology, UC Davis
T. Michailides, KAC, Parlier
C. Ledbetter, USDA/ARS, Parlier
J. Connell (Emeritus), UCCE – Butte County
J. Preece, M. Aradhya, USDA Clonal Germplasm Repos.
S. Sathe, Food Science, University of Florida
C. Peace, RosBreed Project, WSU
J. Slaughter, R. Robinson, CA nurseries

Objectives:

A. Compile information from UCD interspecific breeding populations for dissemination to interested public and private researchers.
B. Propagate the most promising selections for transfer to the USDA Germplasm Repository for public access.
C. Determine the value of marker assisted selection (MAS) in characterizing and prioritizing populations of differing interspecific backgrounds.

Interpretive Summary:

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 (UCD), 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 identified, catalogued and propagated. Detailed pedigree relationships as well as trait expression data have been transferred to the RosBreed website (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 several advanced interspecies introgression lines.

Disease and nematode evaluation plots are concurrently being developed in cooperation with UCD, USDA, nursery and grower cooperators. 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. Initial inheritance studies indicate that some markers may be unreliable as indicators for specific trait selection, presumably a unique consequence genetic masking resulting from their inter-species origin.

Field crosses have generated over 5000 seed. Approximately 25% of this seed has been germinated and undergoing testing. Some material is being saved as seed for later germination and testing including seedling of almond by peach (F1, F2,), almond by Nemaguard, almond by 40A17 (nematode resistant source), peach by *Prunus mira*, (F1, BC1), peach by *P. tangutica* (F1, F2), peach by *P. davidiana* (F1, F2), peach by Nemaguard, peach by *P. argentea* (F2, BC1), peach by almond *by P. scoparia* (F2, BC1). Progeny population sizes range from only a few to several hundred individuals depending upon the difficulty of the cross. Recent field crosses have generated an additional 3000 seed and/or seedling of almond by peach (F2,), Nemaguard by CP45 (almond parent of Nickels rootstock), almond by 40A17 (nematode resistant source), peach by Prunus mira, (BC1), peach by *P. tangutica* (F2).

Over 20 clonal genotypes as well as over 2000 seed from interspecies introgression lines have so far been distributed to cooperating researchers, including nurseries and growers for rootstock trait evaluations. Traits currently being evaluated for include drought and salt tolerance, verticillium resistance, crown gall resistance, phytophthora's/waterlogging resistance, nematode resistance, scion architecture modification, compatibility with Nonpareil, and capacity for rootstocks to develop more compact scion tree size. Test sites are located in Butte, Colusa, Yolo, Stanislaus, Fresno, and Kern counties.

Materials and Methods:

A. Compile information from UCD interspecific breeding populations for dissemination to interested public and private researchers. Breeding population information is being disseminated through these ABC annual reports (available at Almonds.com/ ResearchDatabase), as well as the variety development annual report as well as through the RosBreed website (www.rosbreed.org) in publications #1, 5, 6, 8, 9, 12, 13, 14.

B. Propagate the most promising selections for transfer to the USDA Germplasm Repository for public access. Propagations were done by softwood cuttings or T-bud-propagations for distribution to interested researchers. We also used bulk and self-pollinations to generate seed populations of 100-500 individuals for germination/greenhouse planting or seed storage for later germination assays. Crossing targets included *P. persica, P. mira, P. webbii I P. tangutica, P. scoparia, P. davidiana* and *P. dulcis* interspecies F2's and backcrosses.

C. Use RosBreed developed molecular markers to test whether molecular markers assort normally from parent to progeny in interspecies crosses or whether aberrant segregation patterns (due to chromosomal rearrangements such as translocations/inversions and associated gene methylation) are evident. Distortions from expected patterns are identified using standard genomic analysis software such as Pedimap and also to visual examination of the data. Where discrepancies are identified, inheritance patterns of individual formative markers have been characterized using standard qualitative methods.

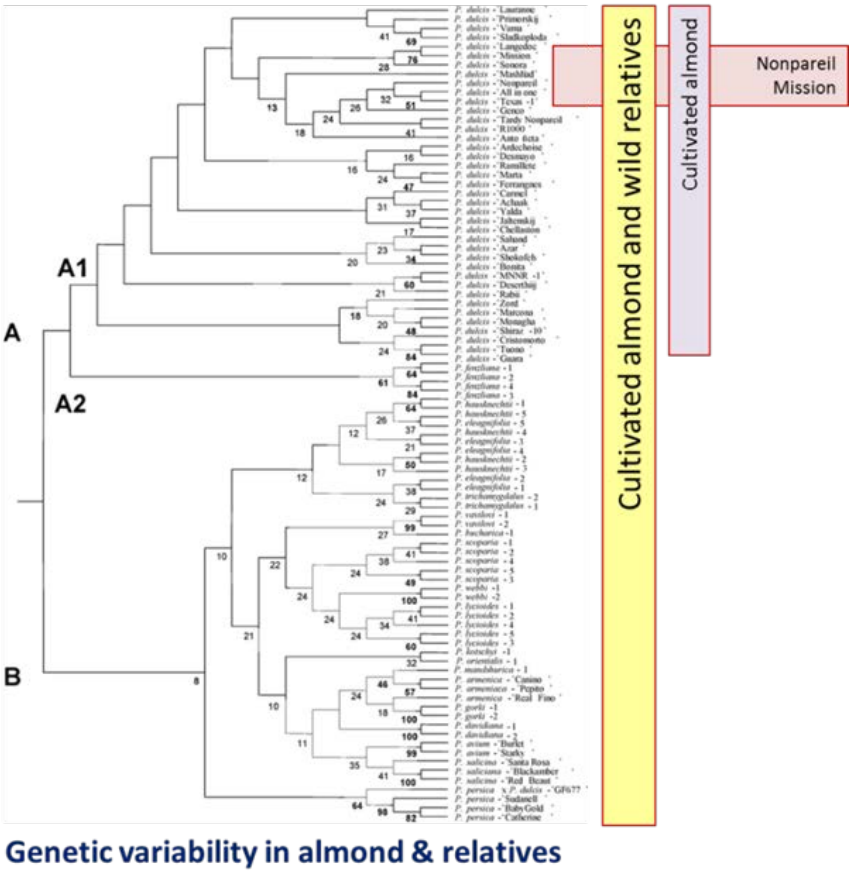

Figure 1. Revised genomic relationships among species evaluated (Ref. #5 & 6) demonstrating the large germplasm diversity exploitable for rootstock breeding.

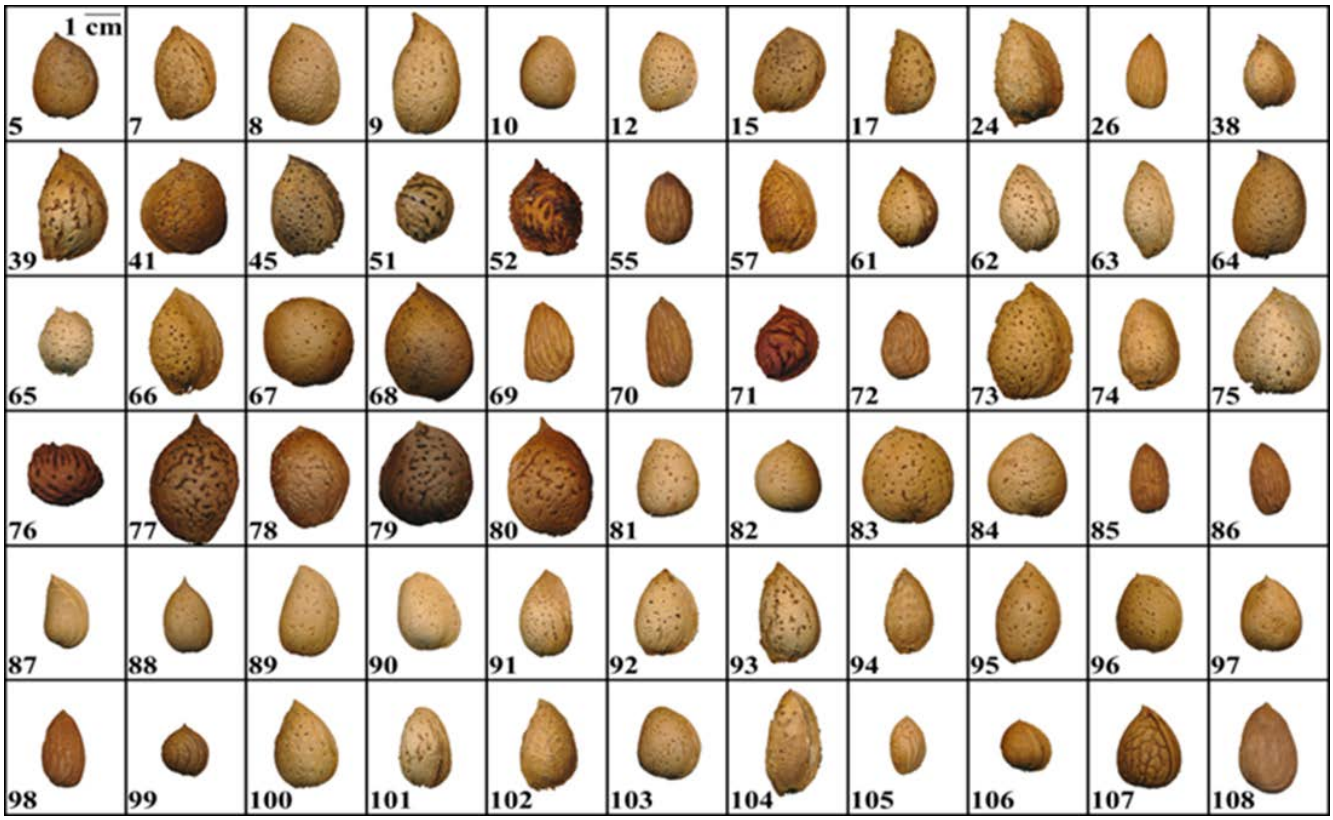

Figure 2. Representative sample showing the diversity of almond types present in this germplasm. The numbering of the seeds is the same as in **Table 1**. All seeds are in-shells except # 26 (Nonpareil), 55, 69, 70, 72, 85, 86, 98, and 108. [Results are from a published (ref. #14) collaborative study with Dr. Sathe's group at the University of Florida].

Results and Discussion:

Compile information from UCD interspecific breeding populations for dissemination to interested public and private researchers.

Species represented within the UCD almond germplasm *include Prunus tangutica, P persica, P davidiana, P mira, the argentea, P scoparia, P dulcis,* and *P webbii* which taken together represent a much broader range of genetic breeding options then present in the traditional almond breeding germplasm and in particular the California germplasm which is largely derived from 2 cultivars: Nonpareil and Mission (**Figure 1**). An extensive diversity is also captured for each species within introgression lineages (see annual Variety Development Reports). Considerable variability has been captured and demonstrated for both fruit type (**Figure 2** and **Table 1**) and tree types, including germplasm previously shown to have disease resistance value when utilized as a rootstock (**Figure 3**). Genotypes range from interspecies hybrids with a well-established rootstock value owing to their typical high vigor and so related disease/stress resistance (**Figure 4**), to genotypes demonstrating the ability to modify scion architecture and physiology (**Figure 5**). The species donor in **Figure 5** is *Prunus scoparia*, which also possesses tremendous adaptability to arid environments. (The inset shows native *P. scoparia* growing at a 3000m mountain pass in the Zagros Mountains of Iran were conditions are so extreme that no other plants survive).

Table 1. UCD breeding germplasm analyzed for seed characteristics in a collaborative study with University of Florida. The numbering of the individual genotypes is the same as in **Figures 7** and **8**.

No.	Genotype	Origin	(%) Almond genome	Nut Length (mm)	Kernel Length (mm)	Nut Width (mm)	Kernel Width (mm)	Nut Thickness (mm)	Kernel Thickness (mm)	Nut Mass (g)	Kernel Mass (g)	Soluble protein (g/100 g)	R-ELISA	R-WB	R-DB
99	A7-23	*P. argentea* (bitter seed)	0	19.03	13.38	15.25	9.73	12.05	6.04	1.47	0.37	17.28	0.61	0.64	0.61
107	A7-25	*P. webbii* (bitter seed)	0	28.97	20.39	18.28	11.75	13.66	7.26	2.93	0.82	19.09	0.51	0.75	0.57
102	F10D,2-14	*P. fenzliana* (F2)	0	30.55	22.3	16.48	11.38	11.33	8.43	4.54	1.03	19.21	1.66	0.78	1.1
52	Andross	Peach (*P. persica*) (bitter seed)	0	35.31	17.77	26.12	11.43	19.5	3.89	6.21	0.36	20.65	0.39	0.53	1.01
105	A10-4	*P. bucharica* (bitter seed)	0	19.14	14.33	10.33	6.62	7.35	4.66	0.58	0.21	20.94	0.59	1.01	0.82
87	A7-28	*P. webbii* (bitter seed)	0	25.72	18.43	14.13	9.1	10.2	6.3	1.39	0.49	21.04	0.88	1.11	1.03
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	17.75	9.86	12.82	4.33	2.48	0.29	23.39	0.53	0.79	0.83
76	A13-1	*P. persica* × *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	15.23	10.28	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* × (Nonpareil × *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 × *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 × *P. persica*	50	36.88	23.87	28.7	16.37	20.85	8.75	9.18	1.53	13.79	0.75	0.09	0.85
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	8.03	1.85	0.83	17.07	0.47	0.57	0.87
100	F10D,3-3	*P. argentea* (BC1)	50	29.57	23.42	18.62	12.41	13.8	7.01	1.88	0.96	17.47	0.26	0.4	0.65
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
94	F10D,2-5	*P. webbii* (BC1)	50	28.66	20.83	14.57	9.81	11.31	8.07	1.23	0.76	17.99	0.47	0.75	0.86
89	F10D,3-15	*P. webbii* (F2BC1)	50	33.32	24.03	20.99	12.86	14.64	7.18	4.1	0.96	18.58	0.33	0.91	0.82
41	F10C,12-28	(Nonpareil × *P. persica*) F2	50	35.09	20.24	23.93	13.04	17.99	9.02	4.96	1.08	19.32	1.76	0.71	1.19
92	F10D,1-2	*P. webbii* (BC1)	50	29.97	20.76	19.8	12.21	14.21	7.15	1.59	0.84	20.4	0.68	1.11	1.14
91	F10D,1-4	*P. webbii* (BC1)	50	30.79	23.09	18.07	11.93	13.31	7.57	1.94	0.95	20.5	1.32	0.62	1.1
90	F10D,1-22	*P. webbii* (F2BC1)	50	28.91	21.59	21.35	12.65	15.24	7.72	2.45	0.97	21.05	1.78	1.01	1.11
78	Hansen536	Almond × *P. persica*	50	34.51	23.82	24.61	13.93	18.9	7.46	7.44	1.12	21.06	0.66	1	0.77
95	F10D,3-26	*P. webbii* (BC1)	50	33.55	24.05	20.27	11.4	14.4	7.45	3.23	0.93	21.17	1.06	1.02	0.89
45	F10C,20-51	(Nonpareil × *P. persica*) F2 (bitter seed)	50	35.12	25.14	21.25	12.63	14.98	7.31	2.43	1.1	23.87	0.56	0.7	0.69
57	F5,16-60	(Mission × *P. argentea*) F2	50	32.85	23.77	17.06	11.1	11.9	7.34	1.56	0.87	24.08	0.44	0.57	0.99
84	F10D,3-23	Padre × F5,4-4	69	27.45	20.37	19.82	11.85	13.38	7.71	2.32	0.84	14.48	1.49	0.76	1.11
12	F5,20-42	Padre × F5,4-10	69	26.76	21.42	17.85	12.07	14.03	8.18	1.87	1	16.72	0.65	0.99	0.66
83	F10D,1-26	Nonpareil × F5,4-4	69	30.84	23.05	24.82	14.16	15.84	6.87	3.88	1.11	17.64	1.61	1.27	0.99
17	F8N,7-4	F5,4-10 × Sonora	69	31.96	22.74	16.12	10.66	10.66	6.21	1.17	0.76	19.52	0.65	1.14	0.81
39	8010-22	Nonpareil × F5,4-10	69	37.57	24.6	19.31	12.5	14.1	7.07	1.9	1.05	21.06	2.09	1.43	0.89
55	SB13,25-75	Nonpareil × F5,4-10	69	NA	23.08	NA	12.54	NA	7.76	NA	1.17	22.18	1.78	0.94	1.49
82	F10D,2-18	Nonpareil × F5,4-4 (see No. 4)	69	24.86	19.04	17.53	10.8	13.13	8.47	1.95	0.8	22.4	0.76	0.65	1.2
15	F8N,6-68	F5,4-10 × Solano	69	30.72	21.57	19.93	12.47	14.38	7.19	1.89	0.96	23.47	0.88	1.02	0.91
81	F10D,3-7	Almond × *P. webbii* × *P. persica* (BC2)	84	26.25	20.45	16.6	10.62	12.64	6.74	1.41	0.69	15.35	0.42	0.55	0.77
72	97,1-232	SB13,25-75 × Winters (see No. 55)	85	NA	23.62	NA	13.42	NA	8.16	NA	1.29	20.61	2.06	0.99	0.9
9	F5,13-54	(Mission × *P. fenzliana*) BC1 × Sonora	88	37.19	23.69	19.52	11.92	16.68	8.31	2.94	1.05	16.28	0.7	0.54	0.68
10	F5,10-9	(Mission × *P. fenzliana*) BC1 × Sonora	88	27.28	21.12	18.82	12.24	14.15	7.04	3.08	0.82	18.11	0.61	1.08	0.99
103	F10D,2-3	(Mission × *P. fenzliana*) BC1 × Sonora	88	27.63	21.82	20.09	13.17	16.3	8.92	3.24	1.13	20.71	1.56	0.89	1.15
7	F5,6-13	(Mission × *P. fenzliana*) BC1 × Sonora	88	32.03	22.06	17.25	10.83	10.51	6.71	1.66	0.84	25.6	0.95	0.83	0.84
8	F5,6-1	(Mission × *P. fenzliana*) BC1 × Sonora	88	33.78	23.01	23.68	14.64	16.75	7.38	5.08	1.33	25.88	0.92	1.13	0.94
108	97,3-40	Almond × *P. webbii* × *P. persica* (BC3)	92	NA	33.26	NA	15.14	NA	8.7	NA	2.08	25.31	0.9	0.9	0.87
75	2004,9-1	Nonpareil × 97,1-232	93	34.28	24.96	23.78	13.46	18.07	7.54	3.15	1.24	14.54	1.89	1.1	0.86
74	2004,8-201	Nonpareil × 97,1-232	93	32.14	24.05	21.45	12.97	13.97	8.13	2.06	1.26	15.81	1.67	1.09	1.22
73	2004,8-160	Nonpareil × 97,1-232 (see No. 72)	93	38.45	28.58	22.52	14.18	15.41	8.64	2.96	1.77	19.84	2	1.15	0.85
38	SB13,54-39E	(Nonpareil × *P. persica*) BC3	94	26.19	16.93	15.83	10.23	12.33	8.18	1.05	0.7	21.51	1.96	0.72	1.16
98	97,2-240	*P. webbii* (BC4)	94	NA	23.76	NA	12.61	NA	9.45	NA	1.28	22.22	0.4	0.61	1
80	2005,20-192	(*P. persica*) BC4	94	37.09	20.55	26.46	14.58	19.27	7.37	7.31	0.99	23.91	0.63	0.88	1
85	2000,2-3	Almond × *P. webbii* × *P. persica* (BC4)	96	NA	23.91	NA	11.62	NA	8.97	NA	1.17	19.89	1.93	0.96	0.75
86	2000,8-27	Almond × *P. webbii* × *P. persica* (BC4)	96	NA	24.26	NA	12.13	NA	8.62	NA	1.2	23.92	0.55	0.87	0.9
68	Tuono	Almond variety	100	38.43	26.35	27.67	16.27	18.3	8.22	5.45	1.58	17.14	0.32	0.96	0.8
69	2004,18-20	Almond variety	100	NA	26.34	NA	13.12	NA	8.7	NA	1.54	18.72	0.68	0.85	0.99
61	Mission	Almond variety	100	27.89	20.76	19.79	12.36	15.77	8.87	2.55	1.04	19.17	0.46	0.84	0.93
64	Ferragnes	Almond variety	100	36.35	26.8	23.06	14.18	17.04	8.29	4.09	1.48	19.37	1.56	1.02	1.21
70	95,1-26	Almond variety	100	NA	29.66	NA	14.25	NA	9.47	NA	1.98	20.94	1.1	0.66	0.74
24	Sonora	Almond variety	100	37.02	27.7	18.89	13.08	12.69	7.8	2.25	1.52	22.07	0.74	1.08	1.13
67	Marcona	Almond variety	100	29.38	21.96	25.83	17.26	19.62	8.75	5.55	1.55	22.22	0.88	1.02	1
66	**Winters**	Almond variety	100	36.41	26.33	19.25	11.87	14.08	8.13	2.09	1.21	22.37	1.05	0.73	1.3
26	**Nonpareil**	Almond variety	100	NAc	24.74	NA	13.49	NA	7.86	NA	1.31	23.07	1.02	1	1
65	Sweetheart	Almond variety	100	22.47	19.1	18.98	12.51	14.33	8.84	1.54	0.98	25.52	1.73	1.12	1.16
63	Kahl	Almond variety	100	34.27	25.95	17.03	12.11	14.99	8.04	2.2	1.2	26.29	1.22	1.12	0.9
62	Chips	Almond variety	100	28.66	21.51	19.45	12.68	14.71	8.18	2.02	1.06	26.46	1.68	0.91	1.17

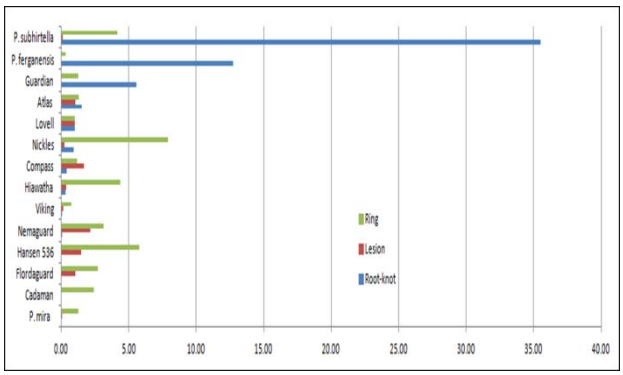

Figure 3. Differences in interspecies germplasm resistance to various nematodes as documented by previous studies by Bliss et al. at UCD.

Figure 4. Increased tree size and vigor typical of an interspecies hybrid (left) versus standard peach seedling (right).

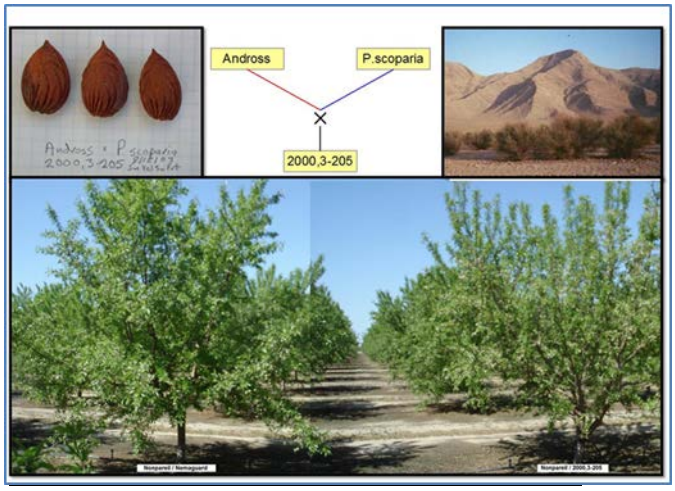

Figure 6. Seedling at right is an advanced backcross to peach of the *P scoparia* interspecies hybrid shown in **Figure 6** demonstrating heritability of some aspects of drought tolerance from the wild species.

A major challenge to rootstock breeding is the maskings affect that interspecies hybrid vigor has on any specific disease resistance genes within the germplasm. This is because the rootstock may be essentially able to outgrow the disease even though it is susceptible. Since the individual can be clonally propagated, this genotype can still be used directly as a rootstock, but often multiple improved traits are desired in the next generation of California rootstocks (nematode resistance, soilborne disease resistance, graft compatibility, scion architecture, etc.) which would require additional breeding to recombine all desired traits into one genotype. Because hybrid vigor is lost in subsequent self's in crosses, any species-hybrid-vigor advantage would be lost in rootstock breeding progeny which would confound both selection progress as well as the use of molecular markers to identify traits of interest. The only way to accurately determine the breeding value of such material would be to make and analyze the appropriate crosses. For example, **Figure 6** shows an advanced backcross to peach of the *P scoparia* interspecies hybrid shown in **Figure 5** which was left to grow under dryland conditions after its 3rd season of irrigated growth (and prior to removal). Tolerances to non-irrigated conditions are evident in the continued tree growth and development as well as by the setting and cropping of a heavy fruit set. Consequently rootstock breeding is typically pursued by 2 separate strategies: 1) generate interspecies rootstocks which combine desired traits (such as nematode resistance and graft compatibility) with vigorous root and tree growth derived from the nature of the interspecies hybrid, and 2) recombine multiple desired traits from

multiple parents using traditional crossing and backcrossing methods. The development of interspecies hybrids can be rapidly obtained and resultant vigor captured through clonal propagation, though the number of desirable traits combined in a single rootstock is limited. Traditional breeding allows the recombination of the greater number of traits but involves a considerable amount of time for rootstock development. While molecular markers would facilitate traditional breeding, the multigenerational breeding pedigrees required to validate these markers would need to be developed and so require multiple years to develop and characterize. An example of such wild species trait introgression's demonstrated in **Figure 7**. In this example interspecies hybrid was backcross to California adapted peach for a total of 4 consecutive generations while at the same time selecting for mildew resistance and good commercial peach quality. Several such introgressions have been included in the UCD germplasm providing both immediate opportunities for new trait introduction as well as required populations for developing useful molecular markers.

In our breeding program we have also identified a rare but intriguing introgression-type which combines the vigor and growth habit typically found in interspecies hybrids with greater genetic uniformity (i.e. greater heritability in progeny) which we have characterized as a primal-type since the tree characteristics almost always shows the more primal species phenotypes.

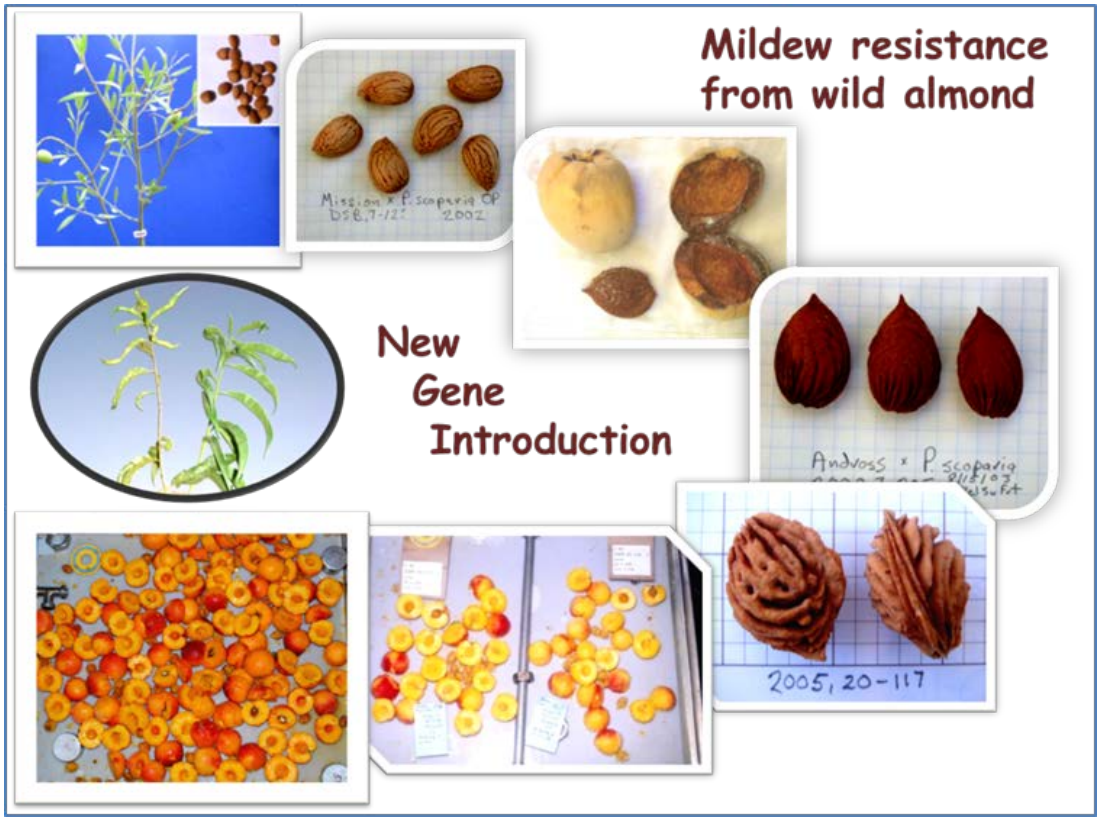

Figure 7. Introgression of mildew resistance from wild *P scoparia* to cultivated peach. Images denote different parents and the multi-generation backcross to peach.

Often found in advanced introgression lines (**Figure 8**) at very low frequencies (1/500-1/1000), these rare individuals appear to be reversions to a more primal, undomesticated phenotype which almost always displays the almond-like leaf, tree and fruit traits characteristic of wild Prunus (**Figure 9**). This reversion-type also typically shows augmentation of tree vigor and tree architecture commonly associated with interspecies hybrids (**Figure 8**). Although rare, the size and genetic diversity of UCD breeding program has allowed the collection of over 30 primal-type genotypes, most often in peach-almond and in peach-mira introgression lines though less frequently in crosses involving other species. In some advanced introgression material, primals have been selected which display a high quality, sweet kernel in an otherwise hybrid vigor background and so may have unique opportunities for commercial almond production under high disease/environmental stress conditions (**Figure 9**). Because of their rarity, there is virtually no information concerning such genetic throwbacks in the literature. Based on the UCD experience, such interspecies derived germplasm may be relatively common in domesticated almond (see references 5 & 6) and some of the domesticated plums. Peach-almond reversion lines are particularly useful because they are easy to identify given the distinctive tree/leaf/fruit characteristics, and because that specific species hybrid group has proven particularly valuable for rootstock development. Over 10 of these unique genotypes are currently being propagated for the rootstock trials with cooperating nurseries and growers. Almond by peach interspecies hybrid breeding lines represent only a small proportion of the potential variability in tree growth and performance available with hybridization to other species. Examples of the range of tree architectures and growth habits are shown in **Figure 10** while the range in performance (potential drought tolerance as indicated by the critical leaf temperature) of advanced breeding lines is shown in **Figure 11**.

Figure 8. The appearance of a primal -type in an almond times *P mira* backcross introgression breeding line. While rare, primal types are readily identified by an increase in figure and associated disease resistance as well as leap and fruit traits more characteristic of the wild type.

Figure 9. Characteristic leaf and fruit traits typical for the primal-group of interspecies introgression genotypes. This tree was derived from an initial almond by peach hybrid and has medium-large sweet kernels.

Figure 10. Range in tree growth habits of selected UCD interspecies breeding lines which are among the over 200 individuals which have been transferred to USDA/ARS germplasm repository for public access by rootstock researchers and breeders.

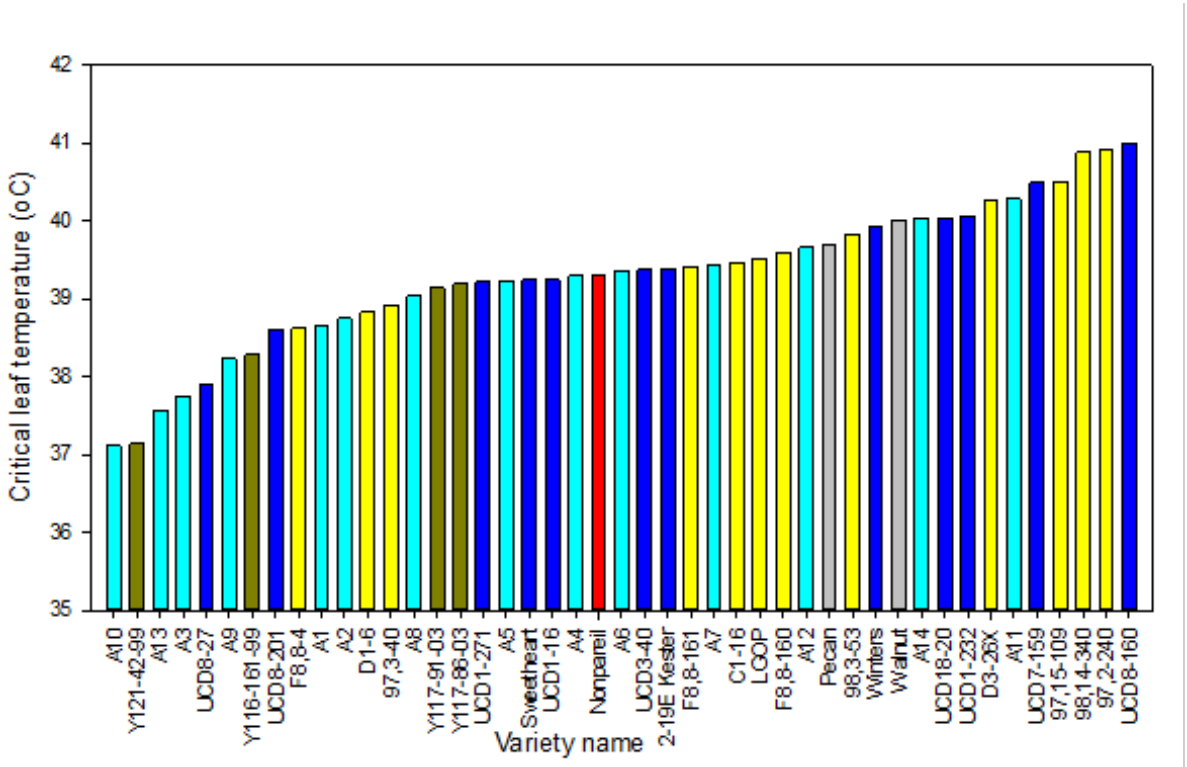

Figure 11. Range in potential drought tolerance as indicated by the critical leaf temperature for UCD and USDA/ARS almond and interspecies germplasm planted at the Chico and Arbuckle RVT and analyzed in 2014 by Matthew Gilbert, UCD.

Propagate the most promising selections for transfer to the USDA Germplasm Repository for public access.

Key selections, representing diverse species lineages, have been propagated for distribution to interested researchers. Using both selfed pollinations as well as controlled hybridizations we have generated over 6000 new seed/seedling genotypes in to 2015 for use in trait/molecular marker segregation projects. Germplasm now available in our core collection as clonal material and/or seed/seedling populations is shown in **Tables 2** and **3**. Advanced seedling progeny generations of this material as well as novel germplasm, including primals, currently in the hybridization program is summarized in **Table 4**. Over 20 clonal genotypes as well as over 2000 seed from interspecies introgression lines have so far been distributed to cooperating researchers, including nurseries and growers for rootstock trait evaluations. Traits currently being evaluated for include drought and salt tolerance, verticillium resistance, crown gall resistance, phytophthora's/waterlogging resistance, nematode resistance, scion architecture modification, compatibility with Nonpareil, and capacity for rootstocks to develop more compact scion tree size. Test sites are located in Butte, Colusa, Yolo, Stanislaus, Fresno, and Kern counties. In addition, approximately 200 individuals of a F2 population of a *P. tangutica* by peach interspecies hybrid which has shown promise for crown gall resistance are being planted to test for consistency and heritability of the trait, and opportunities for developing molecular markers to the controlling gene(s).

Table 2. Species hybrids and introgression lines presently available in our core collection. Items which have been vegetatively propagated in 2013 and 2014/15 are highlighted in yellow while lineages maintained through seed propagation followed by field planting in to be highlighted in blue.

Species	F1	F2	BC1	Other
Peach x P. argentea	2	130	85	150
Peach x P. scoparia	1	10	410	200
Peach x P. dulcis	90	420	80	800
Peach x P. mira	3	150	200	600
Peach x P. davidiana	1	90	40	80
Peach x P. tangutica	12	450		
Peach x plum	2			
Almond x P.persica	240	800	5	1400

Table 3. UCD peach and almond species germplasm selections for which detailed molecular and phenotype information is now available through the RosBreed website (http://www.rosbreed.org/). Items which have been vegetatively propagated in 2013/14are highlighted in yellow while lineages maintained through seed propagation followed by field planting in 2013 to15 are highlighted in blue.

Selection	Parent1	Parent2	Source	Number of Selections, F1 or F2s
2008_3_196	Loadel	Yumyeong	*P.persica*	22
2005_17_1	Loadel	Vilmos	*P. dulcis*	42
2005_17_255	Carson	persXdavidiana	*P.davidiana*	23
2005_17_155	Loadel	persXdavidiana	*P.davidiana*	1
2006_1_107	18_8_11	P_Tang_Mix	*P.tangutica*	1
2003_1_329	DrDavis	P.mira19	*P. dulcis*	1
P.mira#19	P.mira	P.mira	*P.mira*	1
2001_7_180	Andross	P.argentea	*P.argentea*	1
2000_2_8	Loadel	P.argentea	*P.argentea*	2
2005_20_192	91_16_154	Ogawa	*P. dulcis*	1
Hansen1	Almondseed2	Nemaguard	*P.davidiana*	1
Nickels	CP_5_33	Nemaguard	*P. dulcis*	1
2000_3_205	Andross	MissionxScoparia	*P.scoparia*	1
Carmel	Nonpareil	Mission	*P. dulcis*	1
Jordanolo	Nonpareil	Harriott	*P. dulcis*	1
Panamint	BabcockxBoston	GoldminexRioOsoGem	*P.persica*	1
2000_2_16	Loadel	F8_5_166	*P. dulcis*	1
2005_22_204	91_17_195	F8_5_159	*P. dulcis*	2
98_2_132	Pallas	F8_1_96	*P. dulcis*	1
2005_16_172	OHenry	F8_1_42	*P. dulcis*	19
99_16_131	F8_1_121	F8_1_121	*P. dulcis*	1
2005_17_5	Loadel	F10C_12_28	*P. dulcis*	1
Woltemade	Kakamas	F_Wolvamade	*P.persica*	1
Stukey_6_9BF	Nonpareil	F_Stukey69BF	*P. dulcis*	1
Stukey_6_8	Nonpareil	F_Stukey68	*P. dulcis*	1
Stukey_6_27H	Nonpareil	F_Stukey627H	*P. dulcis*	1
Stukey_6_27	Nonpareil	F_Stukey627	*P. dulcis*	1
St_John	ChineseCling	F_StJohn	*P. dulcis*	1
Sonora	Nonpareil	F_Sonora	*P. dulcis*	1
F5C_6_9BF	Nonpareil	F_F5C_6_9BF	*P. dulcis*	1
F5C_6_8	Nonpareil	F_F5C_6_8	*P. dulcis*	1
F10C_20_51	F8_76_45	F_F10C_20_51	*P. dulcis*	1
F10C_12_28	F8_72_33	F_F10C_12_28	*P. dulcis*	1
2007_12_209	2000_16_133		*P. dulcis*	1

Table 3 (continued). UCD peach and almond species germplasm selections for which detailed molecular and phenotype information is available through the RosBreed website (http://www.rosbreed.org/). Items which have been vegetatively propagated in 2013/14are highlighted in yellow while lineages maintained through seed propagation followed by field planting in 2013 to15 are highlighted in blue.

Name-TMG	Parent1	Parent2	Source	Number of Selections, F1 or F2s
2005_20_11	Carson	-	P.persica	1
2005_18_151	2001_7_180	-	P.argentea	1
2005_17_208	Carson	-	P. dulcis	1
2005_17_148	Loadel	-	P. dulcis	1
2008_58_18	2000_8_150	DrDavis	P.persica	1
54P455	GoldenGlory	Bonanza	P.persica	1
98_9_7	93_3_159	Bolinha	Bolinha	1
F8_7_179	Nonpareil	A80_10_22	P. dulcis	1
2009_19_18	Andross	96_9_292	P.persica	3
2005_29_95	92_14_73	92_14_73	P.persica	1
2001_18_215	91_18_6	91_18_6	P.persica	1
Ogawa	90_10_91	90_10_91	P. dulcis	6
F8_1_42	90_1_4	90_1_4	P. dulcis	1
2008_13_194	Loadel	2003_1_329	P.mira	11
2005_19_40	19_2_72	2001_7_180	P.scoparia	1
2005_18_244	Rizzi	2001_7_180	P.argentia	20
2009-28-152	18_8_11	2001_18_215	P.persica	1
2007_10_244	2000_8_150	2000_8_150	P. dulcis	3
2005_19_139	19_2_72	2000_3_205	P.scoparia	1
2005_20_117	Carson	2000_3_205	P.argentia	7
2005_18_191	2000_2_8	2000_2_8	P.argentia	3
2007_12_234	2000_16_133	2000_16_133	P. dulcis	35
2007_11_249	2000_15_122	2000_15_122	P. dulcis	1
Vilmos	F10C_12_28	*VP	P. dulcis	1
Mission_BF	Mission	*MUT	P. dulcis	1
NonpareilBF	Nonpareil	*MUT	P. dulcis	1
TardyNonpareil	Nonpareil	*MUT	P. dulcis	1
40A_17	-	-	P.persica	1
Hansen536	-	-	P. dulcis	1
Mission	-	-	P. dulcis	1
Nemaguard	-	-	P. dulcis	1
Nonpareil	-	-	P. dulcis	1
persicaXdavidiana	-	-	P.davidiana	1
Winters	-	-	P. dulcis	1
Yumyeong	-	-	P.persica	1

Table 4. Advanced introgression populations developed from core germplasm.

Primals	Seed	Pollen
2005,20-192	91,16,154	Ogawa
2007,12-209	2000,16-133	Self
2009,17-302	Andross	96,9-292
2009,17-325	Andross	96, 9-292
2009,18-151	Andross	96,9-292
2009,19-331	Andross	96,9-292
2009,24-337	DRDAVIS	96,9-292
2009,25-36	DRDAVIS	96,9-292
2009,26-185	DrDAVIS	96,9-292
2009,26-205	DrDAVIS	96,9-292
2009,26-266	Dr.DAVIS	96,9-292
2008,61-38	91,17-262	E22-59
2009,19-247	Andross	96, 9-292
2009,27-175	Dr. DAVIS	96, 9-292
2009,18-87	Andross	96,9-292
2008,25-101	Andross	2000_8_157
2008,25-113	Andross	2000_8_157
2008,53-47	Riegels	00,12-110
2008,58-18	91,17-195	00,8-150
2009,17-340	Andross	96,9-292
2009,19-252	Andross	96, 9-292
2009,19-32	Andross	96, 9-292
2009,19-85	Andross	96, 9-292
2009,20-89	KLAMT	96, 9-292
2009,21-1	ROSS	98, 4-177 BRR
2009,21-168	ROSS	98, 4-177 BRR
2009,23-109	RIZZI	E 22-59
2009,23-28	RIZZI	2001_18_215
2009,28-152	18_8_11	2001_18_215
2009,29-107	Lt. ROSS	96, 9-229 BRR
2009,29-15	Everts	2001_18_215
2009,33-175	Halford	2001_18_215
Introgression lineages		
Almond x P. mira (BC1)		
Almond x P. argentea (BC1)		
Almond x P. fenzliana (BC1)		
Almond x P. mira (BC3)		
Almond x P. davidiana (BC3)		
Almond x P. argentea (BC3)		
Almond x P. mira (BC2)		
Almond x P. webbii (BC1)		
Almond x P. webbii (BC3)		
Almond x P. webbii (BC4)		
Almond x P. webbii (F2)		
P. orthosepala		
Almond x P.persica (BC3)		
Almond x P.persica (BC4)		
Almond x P. bucharica		
Almond x P. webbii x P.persica		

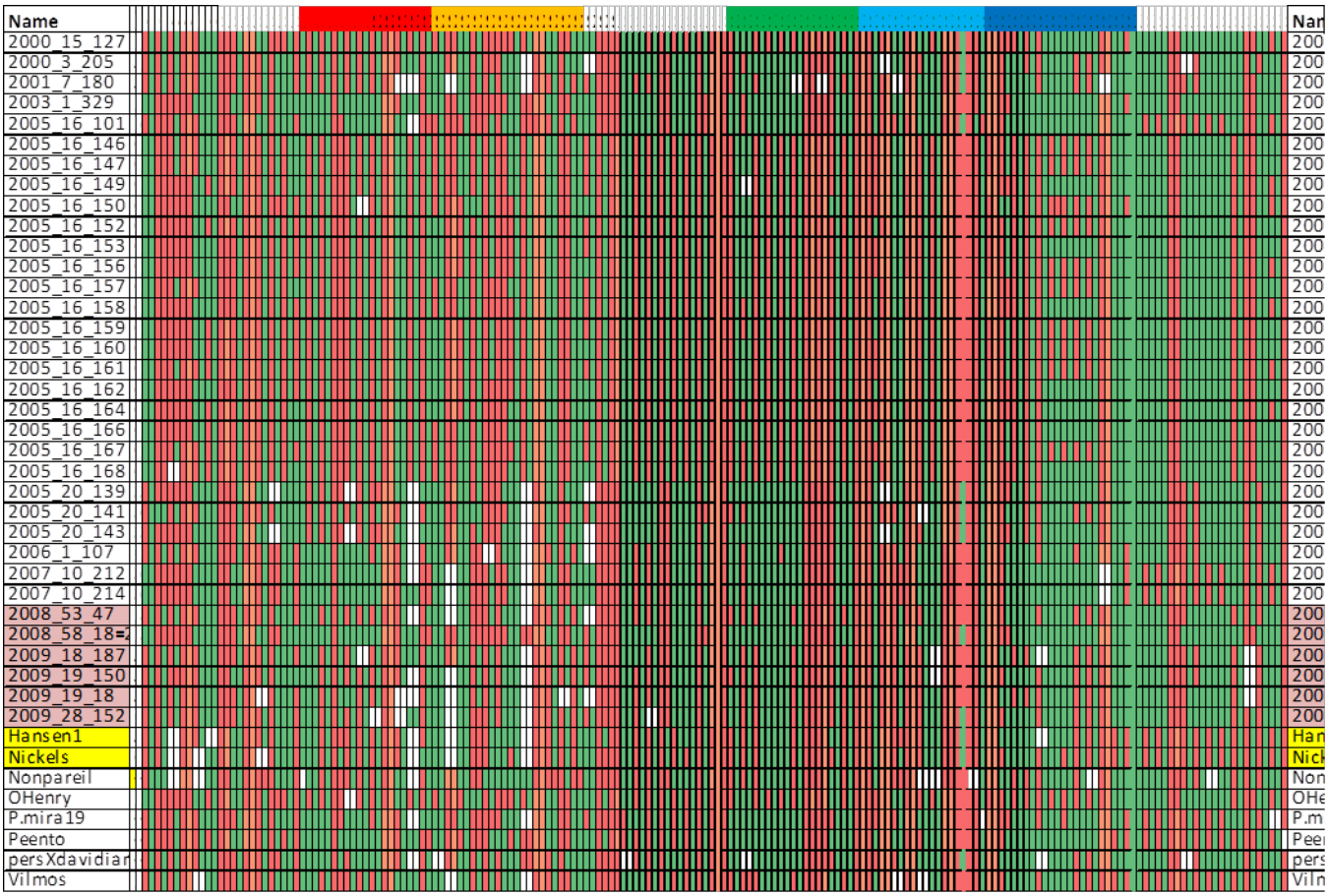

Figure 12. Representative RosBreed marker patterns for each of the eight Prunus chromosomes analyzed (colored bars in the top row). [While only about 30 markers are shown for each chromosome, they are representative of the over 500 markers analyzed.]

Determine the value of marker assisted selection (MAS) in characterizing and prioritizing populations of differing interspecific backgrounds.

The now completed analysis of RosBreed marker data documents an extensive genetic diversity and variability within the UCD breeding program. As detailed in the 2012/13 and 2013/14 Almond Breeding and Interspecific Breeding Germplasm reports, this extensive diversity also led to a large error rate and identifying specific alleles and their associations with specific traits. This is primarily because the markers utilized were initially developed from the peach genome and while the relative position would be expected to be similar (though several exceptions in the form of translocations and inversions are presented in the 2012/13 and 2013/14 reports), the specific allelic signature was often not fully recognized and so accurately recorded for many interspecific relatives and even their advanced introgression lines (see the Almond Variety Development reports). Consequently, the value of individual markers for MAS needs to be determined on a lineage by individual marker basis. However, the overall patterning of marker assortment appears useful for distinguishing between different interspecies lineages and, in particular, characterizing primal's. **Figure 12** shows the typical patterning for an advanced almond-peach introgression line (2005-16-133) as compared to some of the early primal's included in the RosBreed analysis (red

highlight), the almond-peach interspecific hybrids *Hansen* and *Nickels* (yellow highlight) in the various species parents (at base). Frequently, the markers identified in primal's are not in agreement with those of the parents, possibly due to the previously discussed translation errors common when interspecies are analyzed. While, in the interspecies hybrids *Hansen* and *Nickels,* the expected heterozygosity (because one set of alleles is inherited from almond and the other from peach) is observed at most sites, a much more pronounced genetic variability is observed within the primals. Even in introgression lineage 2005-16-133 between parents as diverse as peach and almond, the identity by descent (common parentage) results in a generally consistent patterning over all 8 chromosomes with the expected relatively low number of discrete changes in individuals. A highly variable patterning is observed in the primal's (which also argues against an accidental cross-pollination of the peach parent by outside almond pollen-which would have been simplest explanation). Each primal analyzed, including those with a common parentage, shows a distinctive patterning arguing against a crossing error, even with the high level of translation errors expected in this material.

In 2013/14 and again in 2014/15, additional molecular data was generated through a 9K SNP mini-array based on RosBreed markers but developed with a private service provider (Ilumina). A large number of additional primal's were included in this analysis. The Ilumina array was originally pursued to see if this platform could successfully translate the trait predictions developed from the larger RosBreed data from the much more limited array results. As with the earlier RosBreed analysis, simple translation from marker presence to trait presence is not possible and it still requires molecular and breeding expertise to identify the most promising markers, interpret results, and troubleshoot system glitches. Nonetheless, large haplotypic diversity in UCD peach breeding germplasm is documented. In general, this DNA test was simply predictive (i.e. it was easy to interpret results) for approximately 60% of germplasm but require a more careful analysis for the remainder. In particular, a large number of trait prediction failures were common in the primal material (as in **Figure 12**). For example, markers scored with two heterozygous groups (e.g. "AC (2)") are always for interspecies introgression populations, since they detect the presence of a third allele coming from non-peach species but cannot uniquely identify that allele.

The 9K SNP mini-array patterns for the over 1000 markers evaluated was highly variable for the interspecies introgression lines as well as the primals, similar to that previously seen with the RosBreed markers. However, because more primal genotypes were analyzed, preliminary analysis has identified intriguing trends for a small proportion of the markers, 27 of which are summarized in earlier reports. These markers clearly distinguish primals from the general peach population as well as the peach almond hybrids lineages [AxP] and almond. We are currently examining the possible significance of these marker consistencies (for example, are they from the same segments of the chromosome or involved in similar developmental processes, etc.). In addition we now have F2 populations of several primal genotypes which have become available for genetic and horticultural (disease screening, etc.) evaluation in spring, 2015 and are currently under evaluation.

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