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ALMOND BOARD OF CALIFORNIA ANNUAL REPORT

Project No. 88-M1 Almond Variety Improvement

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<u>Objectives</u>: The long range goals of this research program are to develop pollinizers for current varieties, particularly Nonpareil, and develop replacement varieties for Nonpareil (and other market types) that are self fertile with a range of bloom time and maturity.

<u>Current objectives</u>: (1) Complete the analysis and summarization of the data and information obtained in the described material to date. (2) Confirm self-fertility characteristics and clarify the relationship between selfcompatibility and self-pollination. (3) Make additional crosses that will provide future generations of seedling material that will incorporate low BF-potential, self-fruitfulness, good yield potential, and desirable nut and tree characteristics.

INTERPRETIVE SUMMARY

Information from this project was formerly included either in the Bud-failure or the Variety Evaluation project. This year, this new project was initiated to concentrate information on the specific efforts of genetic improvement in almond cultivars. Emphasis is placed on establishing the genetic basis for pollination control (emphasis toward self-fertility), yield components, and other characteristics essential to almond variety improvement. Genetic improvement also requires an understanding of the genetic basis for BF and how to control it.

During the past few years, the basic genetic germplasm in the almond collection at UCD has been inventoried and evaluated, that which is unneeded eliminated and material useful for maintaining a genetic germplasm base for almond eliminated. This effort has been largely completed but data analysis is still in progress.

Considerable emphasis is being placed on understanding the genetic basis for the unique reproductive system of the almond, including the inheritance and nature of the incompatibility alleles, as we have reported in the Almond Variety Evaluation Project. An anomaly has arisen in the identification of the hypothesized 4th incompatibility from Nonpareil-Mission progeny. Some possible progress was made during the past spring in identifying some seedlings with this group in a population of Nonpareil x NePlus Ultra. This will be the basis of tests in 1989.

Another major effort has been to identify sources of self-fertility among almond germplasm not only to detect self-compatibility (the ability of the pollen tube to grow down and fertilize the egg apparatus) but also self-pollination (the ability of the flower to pollinate itself without the aid of insect vectors). The methods to measure these parameters have been found to be more complex than one might expect due to seasonal effects and to the need to separate between low set due to lack of fertilization and that due to dropping of nuts. Differences among individual plants are not always clear-cut due apparently to quantitative factors affecting set, to the interactions of environment and cultural conditions and to physiological parameters (e.g., heavy crop load, which may influence the tree during the subsequent year. Insufficient light penetration and excess tree competition results in heavy nut drop after initial set and reduces yield.

Nevertheless it has been possible to identify a number of germplasm sources that not only have the capacity for self-compatibility but also the capacity for self-pollination. These include (a) one imported variety 'Trusito' and individual seedlings from an open pollinated population of 'Tuono', an Italian variety showing self-fertility, (b) various hybrids of almond with peach (<u>Prunus persica</u>) and <u>Prunus mira</u> hybrids, both F2 and almond backcrosses and (c) hybrids of almond with other almond species, particularly <u>Prunus webbii</u>. These groups include individual plants of various sizes and growth habit types. Selections have been made of this material for variety potential but have just now been planted into new test blocks.

Considerable effort has been made in the last few years to establish the genetic basis for yield potential in almond and the means for its early screening (see 1987 Variety Evaluation report). This report includes further analysis of these concepts carried out in 1988 with representative selections.

Finally, future almond improvement programs must take into account the possibility for high BF potential among individual selections. Although an exact test to measure BF-potential objectively is still lacking, there is much information now about its inheritance and progress has been made in identifying germplasm breeding lines with relatively low BF-potential although not necessarily BF-free.

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PART I. Analysis and summarization of orchard data on seedlings and selections associated with this project.

Breeding and variety collections at UCD and WEO orchards are of two types: One is the collection of old varieties, species, hybrids, advanced selections and miscellaneous germplasms. The second includes various seedling populations that were established more or less annually until 1983.

An inventory was made of the first category in 1986. Plants on the list were evaluated and items placed into the following groups (a) old California varieties, (b) imported varieties and selections, (c) recent California varieties of chance seedling origin (d), advanced selections or germplasm, (e) species and species hybrids, and (d) mutations and clonal variants.

A program followed in 1987 to repropagate and relocate desirable items. The basic material has now been placed into the following collections:

- 1. National Clonal Germplasm Repository (NCGP), located at Wolfskill Experimental Orchard, Winters. This collection will include the basic species and species hybrid germplasm which will be made available for public use and will be gradually expanded from other collections and almond growing areas around the world.
- Department of Pomology Variety collection. At present this collection mostly includes old varieties of the collection of historical interest. This material will be indexed for viruses in 1989. The collection will be supplemented with other varieties of particular interest in California.
- 3. Department of Pomology Breeding collection. This material includes germplasm of specific interest in future breeding programs. Much of this material has arisen from earlier breeding or has been collected for that use. This collection includes several subgroups of particular interest in the future of this project.
 - a. Self-fertile selections of varied origins.
 - b. Species and interspecies hybrids of almond with other Prunus including peach, P. mira, P. webbii, P. fenzliana, P. argentea, etc.
 - c. Advanced selections representing specific breeding lines of potential use as parents. Includes lines of high producing, late blooming selections from Tardy Nonpareil.
- 4. Selection Blocks for evaluating potentially commercial varieties. This collection now includes two kinds of entries:

- a. Older selections which are carryovers from earlier breeding but which are being reevaluated in the light of current variety needs of the industry.
- b. New selections that have been made (1987) from the various seedling populations where studies have recently been completed.

Most seedling populations from recent breeding studies have been removed as studies are complete. At present two blocks, both planted in 1983, remain but should be removed within a year as studies reach completion. These include populations of peach-almond origin being studied for segregation of BF factors, self-fertility and tree and nut types.

Collections listed in No. 1, 2, 3 (part) and 4 were established in winter 1987-88. Some individuals in group 4 remain in place from older plantings. Most of the blocks, however, were planted in 1987-89 and several years will be required to bring them into bearing. This material provides opportunity to evaluate them for early yield potential and tree size utilizing some of the concepts described in recent reports for yield evaluation and growth habit types.

PART II.

IDENTIFICATION AND CHARACTERIZATION OF SELF-FERTILITY AND SELF-INCOMPATIBILITY ALLELES

The 1987 annual (Variety Evaluation) report reported a survey of self-fertile germplasm. Several questions arose during these studies concerning methods to evaluate self-fertility. One concerned the significance of an early count (1 month after bloom) vs. a late or final count (3 months after bloom) for evaluating self-fertility. Kester found evidence that heavy drop after the initial count was largely due to shading and other physiological factors and concluded the early count was a better indicator of self-fertility as long as sufficient time elapsed to allow the non-fertilized nuts to drop. He also concluded that hand self-pollination bagged tests should be matched with a hand cross-pollination test for comparison.

Identification and inheritance of different cross-incompatibility alleles has also been a major part of the variety evaluation project. One objective has been to identify the four hypothesized incompatibility groups among varieties believed to be 'Nonpareil' x ' Mission' hybrids. A second objective has been to identify varieties that show unilateral incompatibility with Jeffries. The interrelationships of these two problems was described in the 1987 report.

<u>Procedures</u>: Pollination procedures have been described in previous reports. All pollinations were carried out under bags with nonemasculated flowers. Special effort was made to evaluate the two questions posed in the previous discussion.

<u>Results:</u>

1. Inheritance of pollen incompatibility alleles (also see Variety evaluation project).

Genetic incompatibility theory indicates that in seedling populations of Nonpareil x NePlus Ultra, 1/2 of the progeny should have the same incompatibility alleles as the parental Ne Plus Ultra (also of Merced One half should be a new group corresponding to the hypothesized group). 4th group. A population of Nonp x NPU seedlings was prepared several years ago and pollen from ten individual seedlings of this population was tested against four varieties (Merced, Grace, Monterey and Valenta). In 1988 we obtained some nut set (up to 10%) from all combinations of this group, including the unpollinated checks). However, there was a separation between high (40 to 50 per cent) setting and low (10- 20 per cent) setting combina-Consequently it appeared that a possible segregation among the two tions. types occurred such that specific seedlings may belong to the new group. Tests are planned to confirm this in 1989.

2. 'Jeffries'

Selfing of 'Jeffries' was retested to establish a possible basis of self-fertility. Bagging resulted in 0 to 1 % set but comparative fruit sets

of 3 open pollinated limbs were 33% to 41%. The per cent set counts of the different limbs on April 3 and June 1 were identical.

- Test crosses for self-fertile almond selections.
 - a. Individuals in populations of almond x almond species hybrid seedlings were tested for self-fertility in 1987 by bagging and counting nuts (see 1987 report). A number of the selections were retested in 1988 (Table 1).

The seedling selections tested in 1988 in Table 1 are ranked in order of initial per cent set. The per cent set ranged from a high of 56 to a low of 24 which a range consistent with the per sets of previous experiments. The late set count was consistently less that the early count which in these trees was due to severe rodent infestation as well as some management problems. All early counts were based on emerging and enlarging pistils. In the five selections tested by cross-pollination all but one (which was higher) had a comparable set to open pollination. Four of the eight showed approximately the same set with self as cross under bags (one was lower) but essentially all showed less set under the bag without hand pollination.

Of the group the following appear to be self-fertile 18-82, 19-49, 20-52, and 19-13. Not self-fertile = 16-1, 16-2, 18-64, 20-42.

4. Pollination studies with F2 almond x peach seedlings

Plants of this group involved both normal sized (Dw/-) and genetically dwarfed (dw/dw) seedling plants described in some detail in previous reports. Pollination characteristics, yield potentiality and tree size has been involved. One aspect involved the effect of high foliage density and poor light penetration which apparently strongly inhibited full yield expression in the genetically dwarfed trees.

A. <u>Dwarfed plants</u>. In spring 1988 a number of the dwarfed plants were pruned to open them to the light. Different methods of pollination were also tested. Results are presented in Table 2.

The results were somewhat inconsistent but tended to confirm the previous hypotheses. Large differences occurred between the early and late drop on all of the unpruned trees. However, each of these selections showed comparable results among the different pollination tests. This relationship indicated self-compatibility as well as a high degree of self-pollination. However, full expression of pollination capacity and yield expression requires that some adjustment of the canopy by pruning is necessary.

Two of the four pruned trees showed full expression of fertility between the two counting periods. One selection was fully self-fertile and the other showed a lower level of self-fertility and self-pollination. It is not possible to separate between the two possibilities because hand pollination and self pollination was not done. Selection 30-42 showed poor set overall, a fact apparently related to general poor health and disease conditions of the tree. Similarly Selection 25-43 showed relatively poor set and a heavy drop. It appears to be self-fertile but has other problems.

- B. <u>Non dwarfed trees</u>. Self-pollination stu0dies were also carried out (Weinbaum, Muroaka) on the normal sized hybrid trees of the same F2 peach-almond population. These trees have been previously tested for self-fertility. These trees have now grown to considerable size and are crowded in the close planted rows with considerable competition now occurring. This effect on reduced yield capacity was shown in 1987. Results are shown in Table 3.
- 5. Self-fertility in 'Tuono' seedlings.

A number of open pollinated seed from self-pollinated Tuono were obtained from Italy several years ago. These were planted and a population of seedling grown. Bagging tests were made of these materials in 1988 (Table 3.). Out of the 15 seedlings tested, 7 (47%) showed some degree of self-fertility. These seedlings will now be evaluated for nut and tree characteristics and relocated into germplasm blocks.

PART III. EVALUATION FOR YIELD POTENTIAL

The 1987 report included discussion and data on methods of evaluating yield potential in relation to tree size. Additional observations have been made in relation to growth habit traits. Some of these tests were repeated in 1988 with individual selections and data is shown in Table 4.

This group includes a range of genetically dwarfed seedlings of peach and almond, some of which are described under the pollination test discussion. These populations originated by making F1 crosses between a genetically dwarfed peach 54P455 and various almonds including 'Nonpareil' (Wells source), 'Nonpareil' (BF source), 'Padre', 'Sel. 2-2', 'Sel. 6A-11'and several others which did not contribute to these populations. The seedlings of this group were obtained by collecting the seeds from the F1 plants and growing a second generation which segregated approximately 3/4 normal sized (Dw/DW and Dw/dw) and 1/4 dwarfed (dw/dw).

Trees (canopy diameter) were measure and the average areas were calculated by the standard formula. Tree canopy areas ranged from as small as 4 square feet to as high as 57. A count of the total nut number obtained in July (after the final drop) also showed a very wide range. Crop density (numbers of nuts/square foot canopy) was then calculated. Three of trees shown yielded 55 - 59 nuts per square foot which considered to be nearly full expression of yield, the remainder ranging down to about 15. The data parallel those obtained in 1987. Growth habit types variants are indicated by the percentages of the flower buds found on long shoots (10 inches or more), medium (5 - 10 inches) and short (less that 5 inches) spurs. The relative amount of spurs to shoots is indicative of the relative closeness to a peach or to an almond growth habit.

The identification of those showing self-fertility is shown. None of these seedlings have nut characteristics resembling almond. These dwarfed materials have a direct counterpart in the non-dwarfed seedlings described in Table 3 which are also being evaluated for their potential as sources of self-fertility.

PART IV. EVALUATION FOR BUD-FAILURE POTENTIAL

The original purpose for growing the F2 seedling populations (almond varieties x 54P544 dw/dw peach) was to study the segregation of the BF phenotype in this generation. This project was described in the 1985 annual report and alluded to at other times. Inheritance of additional aspects, i.e., growth habit trait (almond vs. peach) analysis, self-fertility, yield potential, etc.) has made this study one of considerable significance. We have now been collecting data on the segregation of BF and RB (roughbark) factors for 6 years. Previously we have found that the F1 hybrids of various almonds and 40A-17 peach (an early blooming, low chilling) peach segregated severe RB and BF individual seedlings at an early age. Table 5 is a recapitulation of accumulated data of the percentages of affected offspring that have been found over the years. The relative numbers of seedlings and the severity of the BF,RB expression is apparently a reflection of the relative BF-potential of the parent although this relationship is not always direct. Nevertheless note that some individual almond varieties did not produce bud-failure offspring, including Sel 2-2, Padre, This was the basis for using some of this Mission, Price and Butte. material as parents in the F2 study.

Note that the F1 hybrids of 54P544 also did not produce the expected RB,BF offspring, a fact that has been correlated to the high chilling, later blooming background present in this particular peach parent. Consequently it was particularly of interest to determine if any segregation of the RB, BF character appeared in the F2 population. The data shows that segregation of this character does indeed occur in this second generation from progeny of all of the grandparental almond varieties, providing strong evidence for the genetic basis of this phenomenon.

However it has been found that the relative numbers of the BF. RB characters is widely different among the separate almond varieties used in Nonpareil and Nonpareil Bf produced by far the highest the crosses. percentage of BF. RB individuals, a fact that is consistent with expecta-This also occurred with Titan, a selection which produces large tions. numbers of RB, BF individuals in F1 populations. Very low amounts of RB,BF characters were produced by Sel. 2-2, Sel. 6A-11 and Padre. This is interpreted to mean that all of these varieties (as believed with all almond varieties) carry the genetic factor for bud-failure but it occurs at a very low level of potentiality.

GENERAL DISCUSSION

This project was separated from the Almond Variety Evaluation and the Bud-failure projects to concentrate on specific aspects of almond variety improvement. During the past 15 years it has been felt more important to address fundamental questions of almond productivity and genetics then to expend a large effort on variety improvement. Thus a major emphasis has been placed on understanding the genetic basis for and control of specific disorders that have a genetic basis, including noninfectious bud-failure and nonproductive genetic disorders. Also major effort was placed on evaluating the current genetic materials available either for commercial use and/or

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for genetic improvement. Consequently much effort has gone into the RVT plots, the germplasm base accumulating at UCD and the potential for specific kinds of exotic almond species in crosses to almond. These studies has included investigating the basis for early and long term selection for yield potential, the nature of the pollination system (including self-incompatibility, self-compatibility and self-pollination). The genetic basis and methods of evaluating many of the nut and tree characteristics for orchard and market use has been developed. Similarly a great deal of data on bloom and harvest maturity times has been accumulated to guide future improvement This and recent annual reports have presented some of this programs. background and considerable time is yet needed to summarize and put into published form all of the accumulated information.

Nevertheless, it is felt that sufficient background has been achieved so that greater effort can be placed on a specific program of new crosses that will lead to the attainment of specific goals of improved almond cultivars. Looking into the future, it has been concluded that the greatest production problems and limits to constant, sustainable and efficient production is to eliminate the need for multiple variety orchards that require maximum cross pollination by bees. Considerable progress has been made in recent years by European plant breeders to select self-fertile varieties. We have made considerable progress in recent years in both understanding the genetic and horticultural basis for self-fertility and in identifying sources of almond germplasm with both self-fertility and self-pollination potential.

Crosses to incorporate self-fertile germplasm will be made in spring of 1989 with additional crosses continuing in future years.

Table 1.	Test cros	sses of a	almond se	elections	for	self f	ertility	character	cis-
	tics.	Parentage	s invol	ve almond	х	almond	species	hybrids	(P.
	webbii).	Each va	lue is t	he average	of	two bra	anch repl	ications	•

Selection		· • • • • • • • • • • • • • • • • • • •				
	725	Open	cross*	selfed*	unpoll.	
	(ι	inbagged)	(bagged)	(bagged)	(bagged)	Problems
		(%)	(%)	(%)	(%)	
1. 6-1	a)	56	-	-	5.5	
	b)	12.5	-	-	2.5	rodents
2. 6-2	a)	53	72.5	12.5	25	
	b)	12.5	43	3	10.5	rodents
3. 19-13	a)	50	33.5	42	21.5	
	b)	29	21.5	14	5	rodents
4. 19-49	a)	39	45	36	26	
	b)	28	33	26	17	nat.drop
5. 18-64	a)	39	-	-	2.5	
	Ъ)	24	-	-	0	nat. drop
6. 20-42	a)	37	-	-	1.5	
	b)	43	-	-	1	
7. 18-82	a)	35	15.5	22.5	15.5	
po nov 2007682891 14 N	b)	29	9.5	15.5	12.5	rodents
8. 20-52	a)	24	34	34	56	
	b)	0	0	0	0	rodents

a) counts made March 25, all pistils growing out of calyx

b) counts made June 2, trees ravaged by rodents

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* hand pollinated; cross pollen was mixture of NePlus Ultra and Nonpareil

Sdlg. Sel.	date poll.	date counted	Open (unbagged)	Cross* (bagged)	Self* (bagged)	Self (bagged)	Ave all
			(\$)	(%)	(%)	(%)	(%)
			I. Not pr	runed			
24-17	Feb 17	a	74	74	58	74	70
		b	12	6	6	13	9
25-25	Feb 25	a	23	4	21	22	16
		Ъ	13	2	18	12	10
29-26	Feb 15	а	56	-	-	61	58
		a	14	-	-	16	15
25-16	Feb 19	а	45	_	-	61	53
		Ъ	9	-	-	22	15
			II. Pru	ned			
25-2	Feb 19	а	47	-	-	10	29
		Ъ	41	-	-	10	26
25-10	Feb 19	а	39	31	37	31	38
		b	33	26	33	29	30
30-42	Feb 19	а	6	6	4	6	5
		b	5	6	3	6	5
25-43	Feb 19	а	16	-	-	18	17
		b	6	-	-	6	6

Table 2. Per cent nut counts, following initial and final set as determined by self, cross and open pollination. All F2 dw/dw peach-almond genotypes. All values were the average of two branch tests.

a = counts made March 24; b= counts made June 2.

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* hand pollinated. Cross pollination was by a mixture of NePlus Ultra and Nonpareil

Table 3. Effect of different types of pollination on different selections of a population of normal sized F2 peach almond hybrids. 1988. These selections are the normal sized (Dw/-) counterparts of progeny described in Table 2. All data in per cent and the average of two branch replications.

Sdlg sel.	Open (unbagged)	Self-hand (bagged)	Self (bagged)	Self-hand/ Open	Self, Open
561.	(%)	(8)	(%)	(%)	(%)
1. Sel. 2-10	a) 86	92.5	82	93	95
	ъ) 20	19	9	95	45
2. Sel. 8-46	a) 77	35	50	45	65
	b) 62	21	44	34	71
3. Sel. 8-24	a) 67	65	67	97	100
	b) 9	7	5.5	78	61
4. Sel. 11.9	a) 58	52	70	79	106
	b) 24	3.5	1.5	15	6
5. Sel. 9-22	a) 61	88	86	144	141
	b) 30	51	65	170	217
6. Sel. 9-12	a) 57	56	61	98	107
	b) 25	18.5	24	74	96
7. Sel. 12-44	a) 44	18	15.5	45	39
	b) 26	10.5	8	40	31
8. Sel. 8-31	a) 37.5	20	21	55	56
	b) 14	3	10	21	71
9. Sel. 7-31	a) 23	27	30	117	130
	b) 14.5	10.5	6.5	72	45
10.Sel. 8-28	a) 21	11	14.5	52	69
	b) 13	6	4	46	31

a) first count

b) second count

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Seedling No.	under		Open Poll.	Bloom days after Feb 1.	
	(1)	(2)			
	(\$)	(5)	(\$)	(no.)	
1	58	9	40	20	
4	18	28	43	26	
5	17	28	-	26	
6	0	4	0	20	
9	39	34	15	20	
10	0	0	20	26	
11	23	0	20	30	
L6	0	0	33	26	
22	0	3	-	34	
25	0	0	70	20	
28	3	0	20	32	
29	0	-	-	30	
34	12	18	18	20	
37	18	8	0	34	
5	0	-	-	20	

Table 4. Per cent set of nuts produced under a mesh bag in different seedlings of a Tuono OP seedling population. Percent for two replicated branches is shown.

Apparently self fertile individuals in population: 7/15 = 47%

Selec- tion	Grand- parent(1)	Canopy area(2)	No. of total	nuts No./ft	Train- ing(3)	Repr. system(4)		GHT(5) M	S
	· ·····	Sq.ft.	No.	No.			8	€	Å
28-50	BF Nonp	56.7	901	15.9	P	?			
30-42	Sel 2-2	56.7	860	15.2		?	10		90
23-25	Sel 6A-11	56.7	1210	21.3					
25-2	Nonp.	40.9	2846	69.5	P	SF	38	7	55
25-16	Nonp.	38.5	1540	40.0		SF			
25-27	Nonp.	38.5	576	22.5			23	24	53
30-51	Sel.2-2	38.5	678	17.6	P	SF	20	44	36
25-10	Nonp.	38.5	2660	69	P	SF	0	10	90
32-22	BF Nonp	33.2	2284	68.8					
25-43	Nonp	33.2	750	22.5	P		7	18	75
24-17	6A-11	12.5	404	25.2		SF			
25-51	Nonp	9.6	527	54.9					
26-35	Nonp	3.9	77	19.7					
25-25	Nonp	-	-	-		SF			
29-26	Sel. 2-2	-	-	-		SF			
25-43	Nonp	-	-	-		SF			

Table 5. Characteristics of potentially useful dw/dw genotypes. All of the original almond parental types were crossed with Peach Sel. 54P455.

(1). Original almond variety crossed with 54P455 peach

(2). area of circle based on diameter of tree canopy

(3). Pruned = P

(4). SF= self-fertile

(5). Percent of buds produced on long (L), Medium (M) or Short (S) shoots.