Project Number: 75-J

Title: Tree Research: Synthetic growth and maturity regulators.

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I. Objectives and Goals: The aim is two-fold: one is to treat almond with synthetic and natural occurring growth substances to test their effectiveness as looseners and also in advancing nut maturity so that a more efficient harvest and complete recovery of the crop might be made; secondly, to examine the physical and chemical nature of the nut to assess the basis of resistance and attractiveness of the maturing nut to the Navel Orangeworm.

II. Abstract. Chemical treatments. Four synthetic growth retardants were applied to Nonpareil and Mission trees: Alar (Uniroyal), Bay-Hol 1302 (Chemagro), Ro 7-6145 (Hoffman-LaRoche) and Release 5507 (Abbotts Laboratories). Alar was most effective in advancing maturity but a reduction in kernel size was observed which was proportional to the earliness of the application. The other chemicals advanced harvest about 3 days and the kernel size was unaffected. While combination of Alar with Ethephon (Ethrel) was very effective in advancing maturity, the latter often caused gumming. In relation to the ripening studies, radioactive sucrose was applied to a leaf near a bearing spur to ascertain how long into the season the nuts accumulated food from the leaves. Chemical composition of nuts: Cross sections of nuts from several varieties were examined for the deposition of lignin, the compound which makes wood hard. Peerless shell accumulated lignin early and in large quantities which may explain why this variety is most resistance to attack by NOW. Extracts of maturing nuts were made to test whether they would serve as attractants for female NOW moths when placed in bait traps. Preliminary trials were conducted by Dr. Rice using these extractives.

III. and IV. Experimental procedures and requests:

A. Synthetic growth regulators: Alar was applied at 500 and 1000 ppm to Nonpareil trees at the Adrian Orchards, San Joaquin County. Ten trees each were sprayed on the month noted below and the degree of dehiscence rated at weekly intervals as the nuts approached maturity. The findings are noted below:

Date of Spraying	Conc.	Mean dehiscence	Days advancement of maturity	Kernel wt.
May	1000 ppm	6.99 (8/19)	12	1.39 g
June	500 ppm	5.93	3	1.46
June	1000 ppm	5.93	3	1.46
July	1000 ppm	5.90	0	1.55
Control	0	5.33		1.53

Ro 7-6145 at 3000 ppm, Bay Hol 1302 at 1000 ppm and Release 5507 at 100 ppm were applied to Nonpareil and the latter two to Mission trees on campus on August 11. The degree of dehiscence on Nonpareils were rated on August 14, 21, and 28 while the Missions were rated on August 28, September 4, 11, and 28. No difference in kernel among the treatments were noted as compared to control trees. The histograms illustrating the range of dehiscence within a 100-nut sample for Nonpareils treated with Ro 7-6145 as compared to the controls on nearby trees. The finding for the other compounds and Mission were similar and therefore not included here. The advancement in maturity was estimated to be about 3 days over the control.

<u>B. Histochemical analyses</u>: Beginning on May 14 when the shells began to harden, samples of nuts from several varieties were collected at 5-day intervals and placed in preservatives. Comparable nuts were sectioned and immersed in a solution of phloroglucinal:2N hydrochloric acid to bring out the locations of lignin deposits. The sections were then photographed. This technique revealed that lignin which makes the shell hard was accumulated in the Peerless variety in larger quantity than in other varieties. The seal in this variety is probably superior to the others, making Peerless less susceptible to NOW damage. <u>C. Chemical attractants in maturing nuts</u>. Nonpareil nuts were harvested and divided into maturity Class I, II, and III. Nuts within each maturity class was then sub-divided into two samples, were paritioned into various fractions by their solubilities in organic solvents and then finally distilled as shown below:

I. Water extracts: 200 g of hull and shell blended with 300 ml water.

solid residue (discarded)-- Aqueous phase partitioned with pet. ether. Petroleum ether fraction (1) -- Aqueous phase (fats, lipids and pigments) partition with ethyl acetate. Ethyl acetate fraction (2) -Aqueous phase (growth regulators & phenols) add equal amount of methanol and distill under vacuum. Methanolic distillate (3) -Final aqueous phase (4) (trapped in cold finger trap containing dry ice and anti-freeze)

II. Methanol extracts: 200 g of hull and shell blended with 300 ml methanol. Filtered Solid residue (1) ______ Filtrate Methanolic distillate (2) ______ Aqueous residue (3) (trapped in cold finger trap)

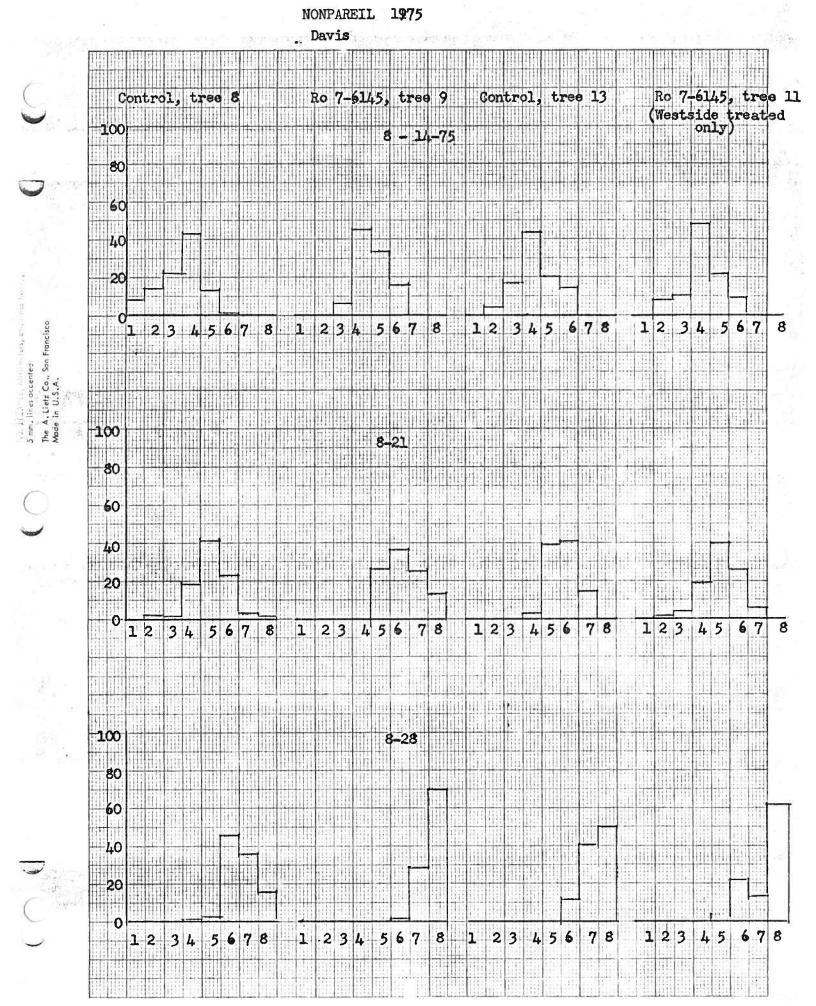
These fractions were then stored frozen or in the solvent until used as constituents in NOW bait traps. Along with this study, crude honey and culture medium used for rearing NOW larva were distilled as above in an effort to isolate chemical attractants to the female moth. Dr. Richard Rice will be doing the field testing.

<u>D. Sucrose transport from leaves to fruits</u>. To ascertain whether nuts approaching harvest maturity will accumulate photosynthates, bearing branches were severed and brought into the laboratory and placed in vases. Radioactive sucrose was then applied to leaves on bearing spurs. After 48 hr, the treated leaves and the different parts of the branches were separated and oven dried. The treated leaves were rinsed in water to determine how much of the radioactivity was left on the surface. The dried samples were pulverized and a portion of the sample combusted to ${}^{14}\text{CO}_2$. The ${}^{14}\text{CO}_2$ was trapped and counted. Six limbs bearing nuts of different maturity were examined in this manner. The distribution of radioactivity is shown on the following page.

Cpm/g of dry tissue

		Limb	number			
Fractions:	I	II	III	IV	v	VI
Rinse (x1000) (surface residue)	378	731	260	46	128	49
Treated Leaves (x1000)	1,121	505	76	182	386	313
Adjacent nuts: pericarp kernel	5,426 4,389	887 1,803	637	7,205	559	11,593
Distal nuts: pericarp kernel		4,617 2,069	-	-	Ξ	-
Untreated leaves on treated spurs	54,548	1,972	8,110	2,415	2,707	5,050
Untreated leaves on distal spurs	452	506	-	-	-	-
Stem & spur next to treated leaves	42,659	5,769	1,251	652	690	789
Stem & spur on distal parts	2,408	740				
Water in vase	57	69	173	89		90

<u>Discussion</u>: While the search for chemical growth regulators to loosen nuts and advance their maturity is necessary, the ultimate answer will lie in developing or acquiring disease and insect resistant almond varieties which will naturally ripen early and abscise freely from the tree. This project has been broadened in scope to seek physical and chemical bases which will assist future breeders identify genetic markers to achieve the industry's goal. To this end, the results obtained this year, and priorly, look promising, in that 1) advancement of maturity by use of select chemicals has been demonstrated, 2) certain shell pesticides will move with photosynthates from leaves to the maturing nuts. Plans are to continue the cooperative work with Dr. Richard Rice in seeking components of maturing nuts which may attract the female NOW moths and with Mr. John Lauck of Chevron Chemicals to study the mode(s) by which insecticides are translocated in almond trees.



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