Epidemiology & Management of Almond Brown Rot, Jacket Rot, Shot Hole, Rust, and Hull Rot in California

Project No.: 14-PATH4-Adaskaveg			
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Objectives:

- I. **Disease management strategies:** Evaluate new fungicides and develop efficacy data based on spectrum of activity, systemic action, and persistence.
 - a. Continue evaluations of new products against brown rot, jacket rot, and shot hole, in experimental orchards at UC Davis and UC Kearney Agricultural Center (KAC) (as well as rust if the disease occurs during the season).
 - Evaluate new fungicides, fungicide pre-mixtures, and adjuvants: fluxapyroxad; penthiopyrad (Fontelis); metconazole – Quash; polyoxin-D - Ph-D or Oso; dodine -Syllit, Luna Experience; Luna Sensation; Quadris Top; Quilt Xcel; Inspire Super; Merivon; as well as biologicals (Serenade Optimum, Botector, Actinovate).
 - Evaluate persistence and post-infection activity of selected fungicides in laboratory studies for management of brown rot.
 - b. For hull rot management, evaluate application timing and new fungicides.
 - * Qol fungicide class, DMI fungicide class, SDHI fungicide class, polyoxin-D, and experimental fungicides, as well as selected pre-mixtures as above.
 - * Alkaline treatments by themselves or in combination with fungicides (pH effects in the shoot tissues will be also evaluated).
- II. **Develop baseline sensitivities** of fungal pathogen populations against new fungicides and determine potential shifts in fungicide sensitivity.
 - a. Characterize baseline sensitivities of *Monilinia* species against SDHIs (penthiopyrad, fluxapyroxad, fluopyram).
 - b. Determine fungicide sensitivities in locations where disease was not satisfactorily managed after fungicide treatments.
- III. Evaluate the new almond variety orchard at UC Davis under simulated rainfall for susceptibility against brown rot blossom blight, shot hole, and other diseases that develop naturally in the springtime.

In this annual report, we can only include 2015 data for the springtime diseases brown rot, shot hole, and gray mold. Trials on summer diseases including hull rot are ongoing, and thus, 2014 data are presented for these objectives.

Interpretive Summary:

In 2014-15, we again evaluated new treatments against major foliar and fruit diseases of almond in California in field and laboratory studies. New fungicides evaluated belong to new (EXP-1) and existing classes (e.g., DMIs - FRAC 3, SDHIs - FRAC 7, anilinopyrimidines -FRAC 9, Qols - FRAC 11, polyoxins - FRAC 19, phosphonates - FRAC 33, isophthalonitriles - FRAC M5, and guanidines - U12). Additionally, several pre-mixtures and rotation programs as well as a natural product were evaluated. FRAC 7 fungicides (boscalid, isofetamid, fluopyram, fluxapyroxad, penthiopyrad – all except penthiopyrad marketed in pre-mixtures) are assigned to three sub-groups that differ in their target binding site. This sometimes results in no cross-resistance among sub-groups; however, cross resistance is still possible because the target site (i.e., succinate dehydrogenase) is the same. With awareness and fungicide stewardship, the arsenal of available fungicide treatments will help prevent the selection and build-up of resistant pathogen populations when applied in rotation or mixture programs. The use of pre-mixtures is a resistance management strategy, and additionally, the spectrum of activity is generally expanded so that several diseases can be targeted by a single treatment. In our research, highly effective single-fungicides and pre-mixtures were identified for the management of brown rot blossom blight, gray mold, and shot hole.

Brown rot incidence in the spring of 2015 was much higher than in 2014, but shot hole only occurred at very low incidence at our trial sites. For brown rot management, most of the singleand pre-mixture-fungicides and rotation programs provided excellent disease control. Studies on the management of hull rot were all conducted in orchards with mainly Rhizopus stolonifer as the pathogen. Over the years, this fungus has been the primary pathogen; the occurrence of Monilinia fructicola has been much more variable. Possibly, microclimatic conditions (temperature) could be responsible for this (e.g., R. stolonifer is more heat-tolerant). We are currently still recommending a two-spray program for hull rot management, with one pre-hull split treatment in early/mid-June (mainly targeting the Monilinia pathogen) and another one at early hull split (mainly targeting the Rhizopus pathogen). In our field studies in 2014, we confirmed previous results, that hull rot caused by *R. stolonifer* can be managed with a single application during early hull split. Our timing studies indicated that earlier applications are also beneficial. If Monilinia species are the main pathogens, however, we previously determined that applications should be done earlier (e.g., early to mid-June). The most effective treatments for hull rot management include FRAC Groups 3+7, 3+9, 7+11, 3+11, and 3+19. In trials where alkalizing treatments were applied to possibly neutralize fumaric acid that is produced by R. stolonifer, lime or sodium bicarbonate significantly reduced the disease compared to the control, but there was no additive effect when applied together with a fungicide. For the most effective integrated management of hull rot, fungicides should be used together with proper water management (i.e., deficit irrigation) and nitrogen fertilization. Lastly, for evaluation of natural host resistance to diseases, our new almond variety block at UC Davis will be available in the coming years.

Materials and Methods:

Fungicide evaluations for management of brown rot, gray mold, and shot hole in experimental orchards – 2015 Research. Field trials were conducted at UC Davis on cvs. Drake, Butte, and Sonora, and at the Kearney Agricultural Research and Extension (KAC) Center on cvs. Wood Colony and Sonora. Treatments were done as single-fungicide, premixture, or rotation programs. Four applications were done at UC Davis from pink bud to petal fall. Trees were inoculated with *M. laxa* after the full bloom application. A single application was done at KAC. This was followed by two 6- to 8-h simulated rain applications, and trees were inoculated with *M. laxa* between these two irrigations. For brown rot evaluation, the number of strikes per tree was counted for each of four or six single-tree replications. All fungicides were also evaluated in laboratory studies on detached blossoms that were inoculated with conidia of *M. laxa* (20,000 conidia/ml) either 20 h before or after treatment. The incidence of stamen infections was then determined after 5 days of incubation at 20C. For efficacy against gray mold, field-treated flower petals were collected after the full bloom spray and incubated on moist vermiculite for 5-6 days at 20C for the development of natural incidence of the disease.

Fungicide evaluations for management of hull rot - 2014 Research. Field trials were conducted in cv. Nonpareil orchards in Colusa, San Joaquin, and Fresno Co. where hull rot was mostly caused by *R. stolonifer*. Treatments to evaluate fungicide efficacy were done as single treatments (at early suture split) or in a two-spray program (early June and early suture split mid-July). In timing studies in Colusa and San Joaquin Co., one to four treatments were applied between June 10 and July 23. Hull rot was evaluated by counting the number of shoot infections in a 30sec evaluation per tree.

Develop baseline sensitivities of fungal pathogens from hull rot-infected fruit against different fungicide classes – 2013-15 Research. Additional isolates of *R. stolonifer* were obtained from almond fruit with hull rot in the summer of 2014. In vitro fungicide sensitivities were determined for difenoconazole, metconazole, fluopyram, fluxapyroxad, trifloxystrobin, and pyraclostrobin using the spiral gradient dilution method as described previously (Forster et al., Phytopathology 94:163-170).

Planting of a new variety orchard. A new variety block was established at UC Davis in the spring of 2014 for evaluation of disease susceptibility of new varieties of almond in the coming years. Twenty-eight varieties on two rootstocks were planted in a randomized block design.

Statistical analysis of data. All data were analyzed using analysis of variance and least significant difference (LSD) mean separation procedures (P > 0.05) of SAS version 9.4.

Results and Discussion:

Brown Rot blossom blight and Gray Mold management – 2015 Research. Although the spring of 2015 was generally very dry, brown rot incidence in the UC Davis field trial on the highly susceptible cv. Drake was very high (average of 148 brown rot strikes per tree). This

was probably due to the inoculation of trees that was followed by some minor rainfall. Disease on cvs. Butte and Sonora, however, was very low.

All treatments significantly reduced the incidence of disease (**Table 1**). Kenja (isofetamid) by itself was among the least effective treatments, but when mixed with the experimental IB18220, it resulted in the lowest disease levels in this trial. Numerically the second most effective program was a rotation of Vangard, Quadris Top, Bravo, and Inspire EC. In the trial at KAC where trees were also inoculated, but in addition received two overhead irrigations for 6 to 8 h after the single application, all treatments were also effective in reducing the incidence of brown rot (**Table 2**). Treatments performed statistically similar on cv. Sonora, but on cv. Wood Colony, Luna Sensation was statistically more effective as some other treatments.

				2/10	2/16	3/5	3/19	Brown rot**		Gray mold***			
								Strikes/					
	Program	Treatment*	Rate/A	PB	FB	PF	PF	tree	LSD^	Inc. (%)	LSD	Severity	LSD
1		Control						148.0	а	76.2	а	2.5	а
2	Single	EXP-1	4 fl oz	@	@	@	0	29.2	bcd	25.3	cdef	0.7	efg
3		Fontelis	20 oz	@	@	@	@	39.0	bc	48.5	b	1.2	cd
4		Kenja	13.7 fl oz	@	@	@	@	42.4	b	37.1	bc	0.9	de
5	Mixtures	Kenja + IB18220	10.3 + 6.9 fl oz	@	@	@	@	4.4	е	13.7	ef	0.3	h
6		Kenja + IB18121	8.6 + 12.9 fl oz	@	@	@	@	26.0	bcde	18.3	def	0.4	fgh
7	Pre-	Luna Experience + NIS	6 fl oz	@	@	@	@	10.2	de	20.3	def	0.3	gh
8	mixtures	Luna Sensation + NIS	5 fl oz	@	@	@	@	17.4	cde	37.3	bc	0.6	efg
9		EXP-2	7 fl oz	@	@	@	@	28.2	bcde	13.1	f	0.4	fgh
10		EXP-3	7 fl oz	@	@	@	@	19.0	bcde	25.7	cdef	0.7	efgh
11		EXP-3	8.5 fl oz	@	@	@	@	20.0	bcde	27.2	cd	0.7	efgh
12		Merivon	5.5 fl oz	@	@	@	@	28.2	bcde	49.7	b	1.4	С
13	Rotations	Syllit	1.5 lb		@	@	@	19.0	bcde	25.5	cde	0.4	fgh
		Tebuconazole	4 fl oz	@	@	@	@						
14		Syllit	2 lb		@	@	@	23.0	bcde	43.7	b	0.8	ef
		Tebucon	4 oz	@	@	@	@						
15		Indar 2F + surf	6 + 16 fl oz	@	@			20.4	bcde	78.0	а	1.9	b
		Dithane + surf	144 + 16 fl oz			@	@						
16		Vangard	5 oz	@				7.0	de	80.9	а	1.5	С
		Quadris Top + DyneAmic	14 fl oz + 16 fl oz		@								
		Bravo	64 fl oz			@							
		Inspire EC	7 fl oz				@						

Table 1. Efficacy of fungicide programs for management of brown rot and gray mold of cv. DrakeAlmonds at UC Davis 2015.

Treatments were applied using an air-blast sprayer at a rate of 100 gal/A on 2-10 (20% bloom), 2-16 (full bloom), 3-5 (1wk after PF), and 3-19-15 (3-wk after PF).

** For brown rot evaluation, the number of brown rot strikes per tree was counted for each of six single-tree replications.
 *** Gray mold was evaluated on flower petals that were collected after the full bloom spray and incubated on moist vermiculite in the laboratory. Incidence of gray mold was based on ca. 50 petals for each treatment replication. Severity was evaluated using a rating scale: 0=0, 1=<25%, 2=26-50%, 3= 51-75%, 4=76-100% petal areas diseased.

Values followed by the same letter are not significantly different based on an analysis of variance and LSD mean separation (P > 0.05).

Most of the field treatments were also evaluated in laboratory pre- and post-infection studies on detached cv. Carmel and Wood Colony blossoms. All seventeen fungicide treatments performed extremely well and reduced the incidence of brown rot from >90% in the control to \leq 4%. The biological treatments Botector and Serenade Opti also performed well, as in 2014, and showed <32% incidence, but the natural product Fracture was less effective.

The efficacy of field fungicide treatments against gray mold was evaluated in a blossom petal assay on cv. Drake (**Table 1**). The lowest levels of disease occurred using the new EXP-1, EXP-2, and EXP-3, as well as mixtures of Kenja with IB18220 or IB18121, Luna Experience, and a Syllit-Tebucon mixture.

Shot hole management – 2015 Research. The incidence of shot hole was very low in the untreated control in 2015 on cv. Drake, Sonora, and Butte at UC Davis and no efficacy data could be obtained.

				Average number of brown rot strikes			
				Sonora		Wood	Colony
No.	Program	Treatment	Rate/A	No.	LSD^	No.	LSD
1		Control		61.4	а	12	а
2	Single	EXP-1	4 fl oz	12.8	b	2.6	bcd
3		EXP-1	5 fl oz	18.6	b	4.6	bc
4		Kenja	13.7 fl oz	Not d	one	3.8	bcd
5		Enable 2F + BreakThru	6 + 16 fl oz	24	b	Not o	done
6		Rhyme	7 fl oz	14.4	b	Not o	done
7		Rhyme + BreakThru	7 + 8 fl oz	13.2	b	Not done	
8		Quash	3.5 oz	15.4 b		Not done	
9	Mixtures	Kenja + IB18220	10.3 + 6.9 fl oz	Not done		1.2	cd
10		Kenja + IB18121	8.6 + 12.9 fl oz	Not done		6	b
11		Kenja + IB18121	10.3 + 16.5 fl oz	Not done		2.6	bcd
12		Syllit + Elite	24 + 4 oz	20	b	3	bcd
13	Pre-	EXP-2	7 fl oz	11.4	b	2.6	bcd
14	mixtures	EXP-3	7 fl oz	13.8	b	2.2	cd
15		EXP-3	8.5 fl oz	17.4	b	1.6	cd
16		Merivon	5.5 fl oz	13.8	b	2.4	bcd
17		Luna Experience + BreakThru	6 + 8 fl oz	14.6	b	3.8	bcd
18		Luna Sensation + BreakThru	5 + 8 fl oz	17.8	b	0.6	d
19		Viathon	2 pt	17.6	b	4.6	bc
20		Viathon	3 pt	12.2	b	3.6	bcd

Table 2. Efficacy of fungicides for management of brown rot of cvs. Sonora and Wood Colony almonds at KAC, Fresno Co. - 2015.

Treatments were applied using an air-blast sprayer at a rate of 100 gal/A at 40% bloom to cv. Sonora (2-15-15) and to cv. Wood Colony (2-19-15) and 6-8 h of overhead sprinkler irrigation was applied to the orchard one and two days after application. Natural rain fall occurred on 2-22, 2-23, and 2-28-15. Trees were inoculated with *Monilinia laxa* (1 gal/tree of 1000 spores/ml) on 2-18-15 (cv. Sonora) or 2-24-15 (cv. Wood Colony). This was followed by 6-h overhead irrigations each after one and two days.

** For brown rot evaluation on 4-20-15, the number of brown rot strikes per tree was counted for 3 min for each of five single-tree replications.

 Values followed by the same letter are not significantly different based on an analysis of variance and LSD mean separation (P > 0.05). Our field studies indicate that numerous highly effective treatments are available for the management of brown rot blossom blight and these performed well even under highly conducive conditions (simulated rainfall) and high disease pressure. Among new compounds, EXP-1, EXP-2, and EXP-3 (FRAC groups not disclosed) are promising treatments with high efficacy against brown rot and gray mold (EXP-1 was very effective against shot hole in 2014), similar to the registered pre-mixtures in FRAC Groups 7/11, 3/11, and 3/7. Our data also indicate that some biologicals can have a role in the management of springtime diseases.

Hull Rot management – 2014 Research. The incidence of hull rot was very high again at some orchard sites. Incubation of diseased fruit indicated that *Rhizopus stolonifer* was the main pathogen at our trial locations. Over the years, this fungus has been the primary pathogen; the occurrence of *Monilinia fructicola* has been much more variable. Possibly, microclimatic conditions (temperature) could be responsible for this (e.g., *R. stolonifer* is more heat-tolerant).

In comparisons of different treatments using the recommended two-spray program (one application in early June mainly for managing *Monilinia* species) and one at early hull split (for managing *R. stolonifer*), all fungicides were statistically similar effective in reducing hull rot strikes, but Luna Sensation numerically had the lowest values (**Table 3**). Long infection periods and the problem of obtaining residues into newly splitting hulls result that the disease is difficult to reduce to very low levels.

			Application		Hull ro	ot strikes
No.	Treatment*	Rate/A	6/5/14	7/10/14	No.	LSD^
1	Control				67.2	а
2	Quash + S2200	3.36 oz + 3.36 oz	@	@	32.3	b
3	Luna Experience	8 fl oz	@	@	26.3	b
4	Ph-D + Tebucon 45 + NuFilm P	6.2 oz+ 8 oz+ 8 fl oz	@	@	25.8	b
5	Inspire Super + DyneAmic	20 fl oz + 16 fl oz	@	@	25.8	b
6	Quadris Top + DyneAmic	14 fl oz + 16 fl oz	@	@	23.5	b
7	Ph-D + Quash + NuFilm P	6.2 oz+ 3 oz + 8 fl oz	@	@	21.8	b
8	Luna Sensation	5 fl oz	@	@	19.8	b

Table 3. Efficacy of fungicide treatments for management of hull rot of cv. Nonpareil almond caused mainly by *Rhizopus stolonifer* - San Joaquin Co. - 2014.

Treatments were applied using an air-blast sprayer at a rate of 100 gal/A. Early hull split was at the 7-10-14 application.
 Evaluations for disease were done on 8-6-14 and the number of hull rot twig strikes was counted for 30 sec for each of the 4 single-tree replications.

 Values followed by the same letter are not significantly different based on an analysis of variance and least significant difference (LSD) mean separation (P > 0.05) procedures.

In other trials alkalizing treatments were applied to possibly neutralize fumaric acid that is produced by *R. stolonifer*. Fumaric acid is considered to be responsible for the dieback symptoms on almond spurs with hull rot-affected fruit. In several trials, lime or sodium

bicarbonate (baking soda) significantly reduced the disease compared to the control (**Table 4**). There was, however, no additive effect when these treatments were applied together with a fungicide. This could be due to pH inactivation of the fungicide. These results, however, are still of value because these alkalizing treatments could possibly be used in organic almond production.

Timing studies in trials with mostly Rhizopus hull rot gave no clear indication as to the optimum fungicide application time over the 6-wk period when fungicides were applied. Pre-hull split treatments (6/10) with Ph-D/Quash were statistically similar effective as early (7/9) or mid 7/22) hull split treatments (**Table 5**).

			Applic.	Hull rot s	trikes
No.	Treatment*	Rate/A	7/9/14	No.	LSD^
1	Control			63.1	а
2	Lime	130 oz	@	37.3	b
3	Quadris Top + DyneAmic	14 fl oz + 16 fl oz	@	35.5	b
4	Baking soda + Ph-D + Quash + NuFilm P	130+ 6.2+3 oz+8 fl oz	@	31.3	b
5	Luna Sensation	5 fl oz	@	29.8	b
6	Baking Soda	130 oz	@	29.3	b
7	Baking soda + Quadris Top + DyneAmic	130 oz+14+16 fl oz	@	28.5	b
8	Baking soda + Luna Sensation	130 oz + 5 fl oz	@	28.3	b

Table 4. Efficacy of fungicide treatments for management of hull rot of cv. Nonpareil almond caused mainly by *Rhizopus stolonifer* - Colusa Co. – 2014.

* Treatments were applied on 7-9-14 (pre-hull split) using an air-blast sprayer at a rate of 100 gal/A.

** Evaluations for disease were done on 8-20-14 and the number of hull rot twig strikes was counted for 30 sec for each of the 4 single-tree replications.

 Values followed by the same letter are not significantly different based on an analysis of variance and least significant difference (LSD) mean separation (P > 0.05) procedures.

			Applications		Hull rot strikes		
No.	Treatment*	Rate/A	6/10	7/9	7/22	No.	LSD^
1	Control					61.9	а
2	Ph-D + Quash + NuF-P	6.2 + 3 oz + 8 fl oz	@			36.3	b
3	Ph-D + Quash + NuF-P	6.2 + 3 oz + 8 fl oz		@		36.3	b
4	Ph-D + Quash + NuF-P	6.2 + 3 oz + 8 fl oz			@	32.5	b
5	Ph-D + Quash + NuF-P	6.2 + 3 oz + 8 fl oz	@	@	@	28.0	b

Table 5. Efficacies of selected timings of Ph-D/Quash mixtures for management of hull rot of cv. Nonpareil almond caused mostly by *Rhizopus stolonifer* - Colusa Co. – 2014.

* Treatments were applied using an air-blast sprayer at a rate of 100 gal/A.

** Evaluations for disease were done on 8-20-14 and the number of hull rot twig strikes was counted for 30 sec for each of the 4 single-tree replications.

 Values followed by the same letter are not significantly different based on an analysis of variance and least significant difference (LSD) mean separation (P > 0.05) procedures. The three-spray program reduced the disease to numerically the lowest values. Therefore, early timings are also beneficial for Rhizopus hull rot. *Rhizopus* spp. depend on injuries and senescent host tissue to invade the host and are unlikely to infect the non-split almond fruit at these earlier timings. The beneficial action of fungicides therefore may be in reducing fungal populations in the orchard. We previously demonstrated that if *Monilinia* species are the main pathogens, applications should be before hull split (e.g., early to mid-June) because these fungi can invade the intact, immature hull and do not require injuries. Therefore at this time, we still recommend a two-spray program to most effectively manage hull rot. This is also because the pathogen population causing hull rot is generally not known for a particular orchard site and both pathogens are usually present at varying frequencies among locations and years. The most effective treatments for hull rot management include FRAC Groups 3+7, 3+9, 7+11, 3+11, and 3+19. For the best integrated management of hull rot, fungicides should be used together with proper water management (i.e., deficit irrigation) and nitrogen fertilization.

		EC50 (mg/L)				
Fungicide	FRAC	Range	Average			
Difenoconazole	3	0.04 - 0.30	0.102			
Metconozole	3	0.04 - 0.15	0.082			
Fluopyram	7	0.12 - 0.64	0.292			
Fluxapyroxad	7	0.20 - 1.90	0.850			
Trifloxystrobin	11	0.001 - 0.004	0.002			
Pyraclostrobin	11	0.001 - 0.005	0.002			

Table 6. In vitro sensitivities of R. stolonifer against fungicides belonging to three FRAC groups

 EC_{50} values were determined using the spiral gradient dilution method.

Determine baseline sensitivities of fungal pathogen populations against new fungicides and potential shifts in fungicide sensitivity - 2013/2015 Research. Baseline sensitivity studies were continued for the hull rot pathogens for representative fungicides in FRAC groups 3, 7, and 11. In 2014, 32 additional isolates of *R. stolonifer* were obtained. Sensitivity ranges and average values for a total of 36 isolates are shown in **Table 6**. With very low EC₅₀ values, the FRAC 11 QoI compounds were more active against this pathogen than FRAC 3 or 7 compounds. Sensitivities for each fungicide were within a ten-fold range, and thus, all isolates are considered sensitive.

Planting of a new variety orchard in spring of 2014. A new variety block was established at UC Davis for evaluation of disease susceptibility of new varieties of almond. Twenty-eight varieties (many numbered accessions) on two rootstocks (Krymsk 86 and Nemaguard) were planted in a randomized block design. Standard cultivars such as Nonpareil, Winters, and Wood Colony were included. The orchard will not receive foliar fungicides and will be evaluated for foliar diseases from spring to fall in the each of coming years (e.g., first leaf, second leaf, etc.).