

STRATEGIES FOR APHID MANAGEMENT IN ALFALFA

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INTRODUCTION

Alfalfa is an important commodity for dairy and livestock enterprises and is the first in terms of acreage planted in Arizona and second to almond in California. It dominates the cropping systems in the Western US given the importance of the dairy industry and other livestock enterprises.

Alfalfa is considered the insectary of the west; providing refuge to wildlife and a variety of beneficial insects, improves soil characteristics, contributes to atmospheric nitrogen fixation through rhizobacteria, traps sediments and takes up nitrate pollutants, mitigates water and air pollution, and provides appealing and pleasing open landscape. Alfalfa also plays an important role in insecticide resistance management by acting as a refuge for susceptible genotypes; e.g., the sweetpotato whitefly *Bemisia tabaci* Gennadius. Beneficial insects move from alfalfa fields into other crops, where they play crucial roles in pollination and biological control. Broad-spectrum insecticides use in alfalfa affect beneficial insect populations and causes potential long-term effects across the entire landscape.

A complex of four alfalfa aphids in the region, pea aphid (PA) *Acyrtosiphon pisum* (Harris), the blue alfalfa aphid (BAA) *Acyrtosiphon kondoi* L., the spotted alfalfa aphid (SAA), *Therioaphis maculata* (Buckton), and the cowpea aphid, (CPA), *Aphis craccivora* C.L. Koch. The first two aphids are the most common and dangerous in the southwest desert of the U.S. The Aphid populations are highest in the winter and spring months when the first cuttings of the year occurred. These first cuttings are in high demand for the dairy industry.

The spotted alfalfa aphid was the first and most devastating alfalfa aphid outbreak on record in 1954. This SAA outbreak destroyed millions of acres of alfalfa in the U.S. Several insecticide-resistant SAA biotypes were identified (Stern and Reynolds, 1958; Nielson and Don 1974). Consequently, resistant alfalfa variety line was produced in respond of the SAA outbreak (Nielson et al. 1970)

Cowpea aphid outbreak in the late-1980's in the Imperial Valley (Natwick 1991) causing serious injury to alfalfa and moved northward and eastward (Natwick 1999a, 1999b, 1999c; Summers 2000a, 2000b) attacking alfalfa in most alfalfa producing regions across the U.S. (Berberet et al. 2009). A CPA-resistant alfalfa varieties were released in 2014.

By 1975, reports showed declining in the efficacy of chemical controls of pea aphid in alfalfa fields in the region (Sharma et al. 1976). That followed by the establish of blue alfalfa aphid in

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the desert production area of alfalfa in the late 80's. The PA and BAA, are similar in size and color and often occur in mixed populations so they are easily misidentified.

The BAA spread quickly across the U.S., particularly in warmer regions (Nielson and Kodet 1975; Nielson et al. 1976a) and it was first easily controlled by different groups of insecticides (Sharma et al. 1975). However, BAA re-infestation was extremely quick and difficult to control. BAA-resistant alfalfa varieties such as CUF 101 were developed (Nielson et al. 1976b) and economic threshold levels were also reported (Sharma and Stern 1980).

In 2013, BAA re-emerged as the most serious alfalfa aphid in the low elevation desert of California and Arizona, and the problem quickly spread northward (Barlow and Natwick 2013, Ramirez, unpublished, Mostafa, unpublished). Although alfalfa pest management practices have worked, at different degrees, for aphid management over the last 40 years, new challenges have arisen, mainly the lack of “softer” chemistry options, mono cropping and the absence of major IPM tools like practical sampling method, well-studied economic threshold, timing of insecticide applications, proper understanding of the roles and conservation of natural enemies. Broad-spectrum insecticides such as organophosphates and pyrethroids are frequently used in alfalfa production to control aphids. If not used correctly, these insecticides are destructive to natural enemy populations, pose risks to applicator health, and have many environmental concerns such as contamination of waterways, and non-target effect. Lower risk selective insecticides are available for sucking insects such as aphids, however they are slow in getting registered for alfalfa, more expensive and less likely to be used as first choice.

Most alfalfa fields are treated with insecticides at least once annually for aphid control, but since the spring of 2013, growers in CA and AZ have been using chemicals multiple times during the season to control BAA. In the past 5–10 years, insecticide applications per season use have increased due in part to higher hay prices and quality demands from the dairy industry. Insecticide cost for alfalfa ranges from \$12–\$40 per acre, which represents about 2-5% of production costs. Some Pest Control Advisors (PCAs) and growers have reported lower insecticide efficacy compared to previous years. In 2011, a need assessment survey among growers and agricultural professionals in AZ indicated that different pyrethroids, as well as chlorpyrifos and malathion, all broad-spectrum insecticides, account for 100% of the insecticides applied to control alfalfa weevil and aphids, the major and most destructive winter/spring alfalfa pests in the region. Dairy and livestock industries are continuously demanding a higher quality product, thus, to meet this goal, many growers have increased their chemical control approach to decrease pest damage. In low desert alfalfa production areas, aphid populations are highest in the winter and spring months when the highly demanded, premium quality first cuttings of the year occurred.

Starting 2015 we observed infections of alfalfa aphids in central and western Arizona with entomopathogenic fungi. These infections spread to California and then northward. The entomopathogenic fungal infection persisted and increased through 2016-2017 season; giving an indication of long term presence of this entomopathogenic fungus in the region.

The work proposed in this project is part of a larger ongoing research and extension IPM program to develop improved IPM practices for alfalfa weevil and alfalfa aphid complex, key alfalfa pests, develop their economic thresholds, verify their use in commercial fields, study

efficacies of available insecticides, promote and evaluate adoption and impact of new practices through a coordinated outreach program.

INSECTICIDE EFFICACY TRIALS FOR BAA AND PA

Several insecticide efficacy trial for BAA and PA have been conducted by Ayman Mostafa at the University of Arizona Maricopa Agricultural Center from 2014-2017. Each treatment replicated four times in a Randomized Complete Plot Design (RCPD). Each plot measured 20 x 20 ft with 5 ft alley between plots. Insecticide treatments were applied using a modified John Deere Hi-Cycle with a 6-row boom and a compressed air system. All sprays were delivered with a tractor-mounted, ground-rig at 20 GPA (ca. 30 psi, TeeJet TwinJet 8003 nozzles, 24 per plot, broadcast over the top). Eight, seven, eleven, eight and eleven treatments were tested in 2014 trial I, 2014 trial II, 2015, 2016 and 2017, respectively.

Evaluations of treatment effects were made by estimations of number of aphids per 5 stems from the radial of each plot. Samples were processed by using paint brush and beating on white sheet. Aphids were sorted to species by microscope. A mechanical yield of 75 sq ft was harvested from the middle of each plot at the time of regular cut using Carter® harvester.

Statistical analyses of ANOVA of treatment means were conducted using SYSTAT™ 13 software. Comparisons of means were tested using LSD contrasts.

RESULTS AND DISCUSSIONS

2014 – Efficacy Trial I

Results of the two aphid species are summarized in Figs. 1, 2 & 3. In general, all treatments were “effective” in that there were fewer aphids following the spray compared to the UTC, with few exceptions. BAA was the major concern of growers and PCAs during that time. For BAA and total aphids, statistical analyses showed that the two rates of Transform, Beleaf, Dimethoate, Warrior II and the tank mix of Warrior II and Malathion 8 have no significant differences in term of their effects. Lorsban and Malathion 8 has no significant differences compared to the UTC. Similar trend has been detected for the effects of treatments on hay yields ($P = 0.005$) (Fig. 4).

2014 – Efficacy Trial II

Results of the two aphid species are summarized in Figs. 5, 6 & 7. Similar to Efficacy trial I, all treatments were “effective” in that there were fewer aphids following the spray compared to the UTC, with few exceptions. Statistical analyses showed no significant differences among treatments in term of their effects on BAA and PA populations. However, all treatments produced significantly higher hay yield compare to UTC ($P = 0.005$) (Fig. 8).

2015 Efficacy Trial

Statistical analyses for insecticides treatment against BAA) and PA at different sampling days after treatment (DAT), in summary, showed all insecticide treatments had fewer and for some significantly fewer Aphid complexes than the untreated plots during the whole experimental period (Table 1).

Statistical analyses showed that all insecticides treated plots had significantly ($P < 0.05$) fewer BAA than the untreated plots, 5 DAT (Table 1). In the 11 DAT it was only Transform at a rate of 1 oz./A that had significantly fewer BAA when compared to untreated check. However, 18 DAT Transform, Pyriproxyfen and Cobalt advanced had significant effect against BAA as compared to untreated plots. At 25 DAT plots treated with Sivanto at both rates (7.0 and 10.5 oz./A), Pyriproxyfen at a rate of 3.2 oz./A, Beleaf 50 at a rate of 2.85 oz./A, Besiege at a rate of 9 oz./A, and Dimethoate 4E at a rate of 1 pt./A had significantly ($P < 0.01$) fewer BAA than the untreated plots. However, there were no significant differences among the insecticides treated plots.

Sivanto at both rate (7 oz./A & 10.5 oz./A), Cobalt advanced at a rate of 24 oz./A, Warrior II at a rate of 1.92 oz./A and Beleaf50 at a rate of 2.85 oz./A insecticide treatments had significantly fewer PA than the untreated check for the 5 DAT. Each insecticide treatment had significantly fewer PA than the untreated check at the 11 DAT. Beleaf 50, Besiege and Pyriproxyfen treatments had significantly fewer PA than the untreated check for the 18 DAT samplings. Post sampling at 25 DAT indicated that plots treated both rates of Sivanto and Pyriproxyfen, Dimethoate, Beleaf 50 and Besiege had significantly fewer PA than the check.

2016 Efficacy Trial

The infestations of alfalfa aphids both at experimental sites and commercial fields were lower in 2016 than in recent years. At the experimental site, there was significantly lower population of BAA (only 2 % BAA) than Pea aphids (87 % PA). Fungal infection was observed in many fields including the experimental sites where up to 34% of BAA infected (Table 2). This naturally occurring entomopathogenic fungus, apparently decreased aphid population. BAA was the major concern of growers and PCAs during the last few years. In 2016, the BAA population reached its lowest level in this area since we start monitoring aphids in alfalfa in 2011.

Initial count (02/10/2016) was made a day before chemical spray. Aphid population was initially 87% pea aphid, 10% cowpea aphid, and 2% BAA and only 1% SAA on stem basis. However, the total populations of the four species were as low as two aphids per stem.

The 2016 result showed slight shift in population dynamics of aphids' population. BAA remarkably infected by fungus compared to other species of aphids (Table 2). Unlike the past three to four years' data, population of Cowpea aphid was slightly higher than that of BAA population. However, populations of BAA, CPA and SAA remained lower throughout the sampling period both in treated and untreated plots, only one to two aphids per stem.

Significant differences in cumulative pea aphid population and over all aphids' population resulted from the insecticidal treatments (Table 3). Plots treated with Dimethoate, Endigo, Beleaf50, Sivanto and Transform insecticides had significantly fewer pea aphid-days per stem than did the untreated control. The rest of treatment had fewer but not significantly different from untreated check. Generally, all insecticide treated plots resulted in higher yields as compared to the untreated check (Table 3).

2017 Efficacy Trial

Eleven insecticides were tested for their efficacies against aphids in alfalfa, including were two microbial insecticides (PFR-97 & BotaniGard).

For the second consecutive year there was significantly lower population of BAA than PA. The fungal infection was repeatedly seen in many fields including the experimental sites.

Following the insecticide treatments applied on January 26, 2017, both PA and BAA were very well controlled (Fig. 11). Some of the chemistries such as Sivanto 200SL, Cobalt advanced, Transform 50WG, Mustang and Endigo provided the best control for the most abundant PA. Malathion & Gamma cyhalothrin and including those mentioned above controlled the less abundant BAA more than other insecticides.

Generally, the populations of PA were low (less than 18 per stem) at any time of the week on untreated plots and never exceed 17 in any of the treated plots at any time of the week. The cumulative pea aphids' population (seasonal total pea aphids) during the experimental period was less than 60 aphids per stem. Each insecticides treatment had significantly fewer seasonal PA than the untreated check. Sivanto at a rate of 10.5 oz./A, Cobalt advanced at a rate of 26 oz./A, Transform at 1 Oz./A, Sivanto at a rate of 7 oz./A, Mustang (4.3 oz./A), microbial insecticides PFR-97 (1 lb./A), and Endigo ZCX (4.5 oz./A) had fewer PA than the other insecticides and untreated check for the entire experimental period (Figure 11).

The population of BAA was generally lower than that of the PA this year too. The seasonal BAA during the experimental period was less than 5 on UTC and less than 1 for Malathion & Gamma cyhalothrin and cobalt advanced treated plots. All insecticides treated plots had fewer seasonal BAA than untreated plots.

The effect of microbial insecticides PFR-97 and BotaniGard appears to be acted differently on PA and BAA. PFR-97 appears to be effective on PA and BotaniGard on BAA.

Treatments were significantly different for dry-matter yield (Figure 11). All insecticide treated plots resulted in higher yields as compared to the untreated check. Plots treated with Malathion & Gamma cyhalothrin, Sivanto 200SL, Cobalt advanced, Endigo, and Mustang exhibited the highest hay yield probably because of the plots treated with them had the lowest numbers of cumulative PA and BAA/stem. The second highest yield was recorded on plots treated with microbial insecticides PFR-97 in spite of higher population of aphids found on this plot. Entomopathogenic fungi such as *Beauveria bassiana* (commercial formulations, BotaniGard) and *Isaria fumosorosea* (Pfr-97) are primarily used for controlling arthropod pests. Research in the recent years evaluated their endophytic (colonizing plant tissues) and mycorrhiza-like (associated with roots) relationship with plants and potential benefits in improving plant growth and health. Studies conducted in California showed the fungus colonized strawberry plants and persisted for up to 9 weeks in various plant tissues (Dara and Dara, 2015a); promoted strawberry plant growth (Dara, 2013); and negatively impacted green peach aphids through endophytic action (Dara 2016). Entomopathogenic fungi also have a positive effect on promoting drought tolerance or plant growth as seen in cabbage (Dara et al, 2016) and strawberry (Dara, 2013) and antagonizing plant pathogens (Dara et al, 2017). Entomopathogenic fungi typically cause infection when spores come in contact with the arthropod host. Under ideal conditions of **moderate temperatures and high relative humidity**, fungal spores germinate and breach the insect cuticle through enzymatic degradation and mechanical pressure to gain entry into the insect body. Once inside the body, the fungi multiply, invade the insect tissues, emerge from the

dead insect, and produce more spores. Fungal pathogens have a broad host range and are especially suitable for controlling pests that have piercing and sucking mouthparts because spores do not have to be ingested. The present higher yield on *Isaria fumosorosea* (Pfr-97) treated plots might have contributed to those all factors that the fungus positively affects in promoting and improving healthy alfalfa plant growth. This is the first field study evaluating the impact of the entomopathogenic fungus on alfalfa aphids control and yield of alfalfa in Arizona. Repeating similar study would result into conclusive remarks.

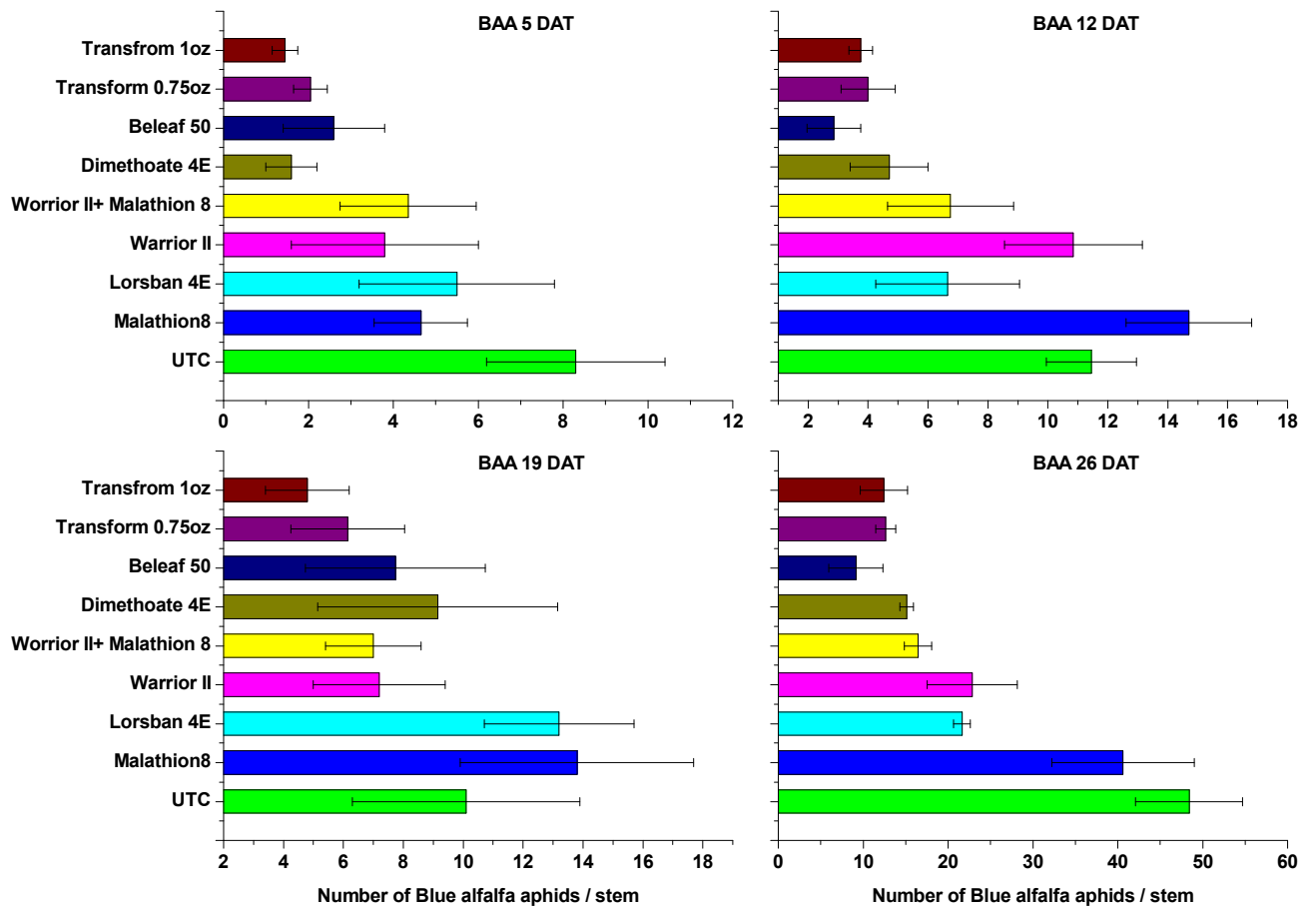


Fig. 1a: Number of blue alfalfa aphids per stem for each treatment 5, 12, 19 and 26 days after treatments (DAT) in the insecticide efficacy trial (I) against aphids in alfalfa at Maricopa Agricultural Center in 2014

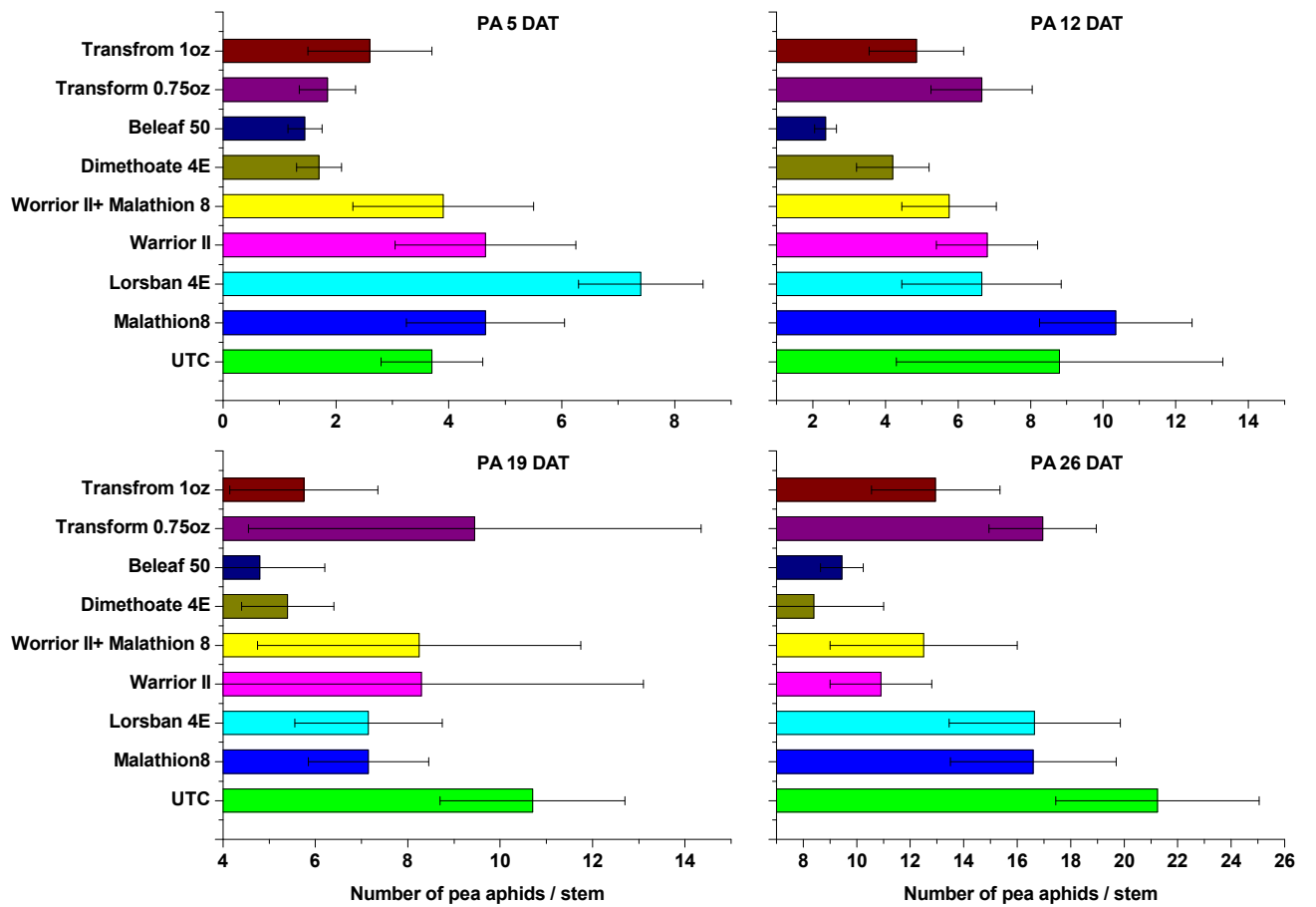


Fig. 1b: Number of Pea aphids per stem for each treatment 5, 12, 19 and 26 days after treatments (DAT) in the insecticide efficacy trial (I) against aphids in alfalfa at Maricopa Agricultural Center in 2014

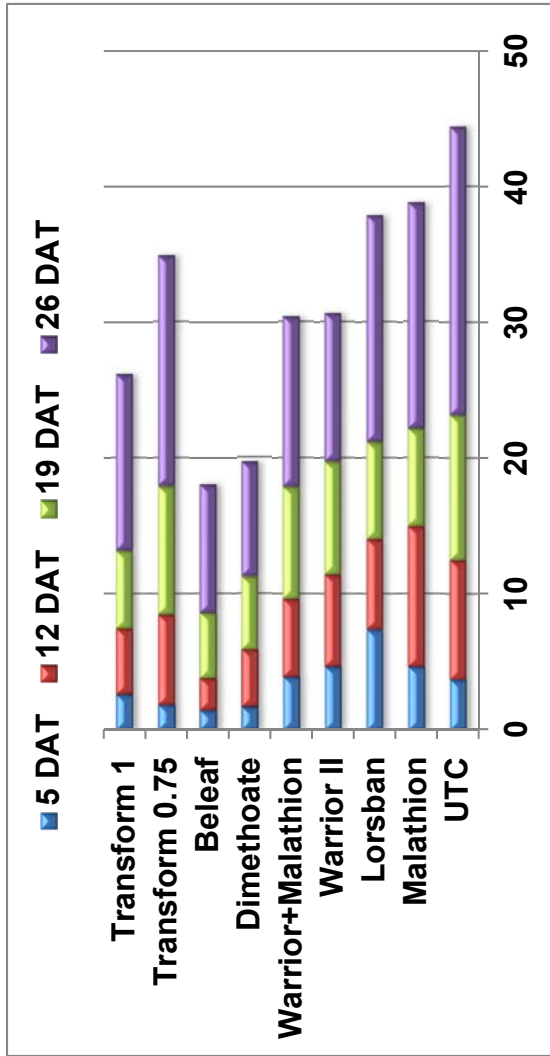
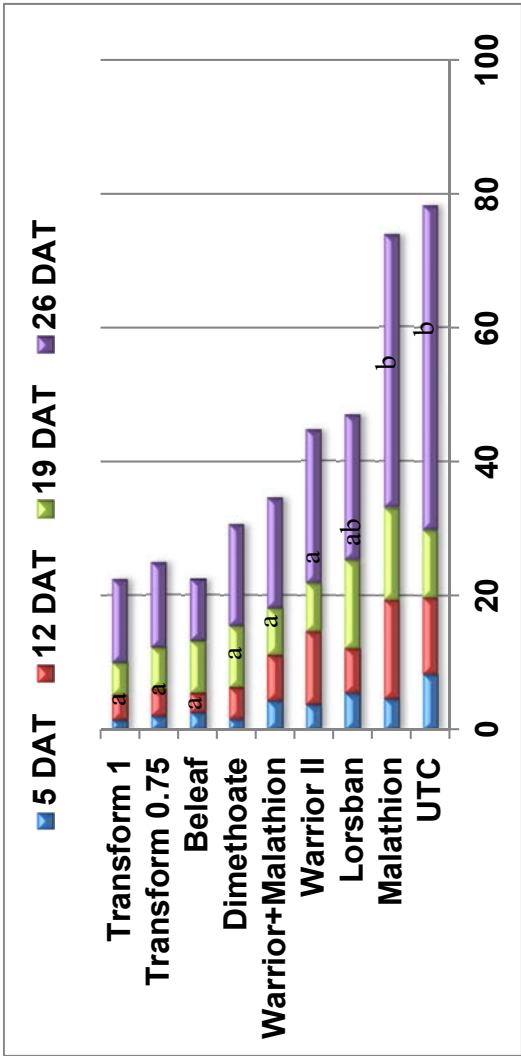


Fig. 3: Seasonal mean (4 post-spray dates) of total aphids per stem (A = BAA, B = PA) for each treatment in the insecticide efficacy trial (I) against aphids in alfalfa at Maricopa Agricultural Center in 2014

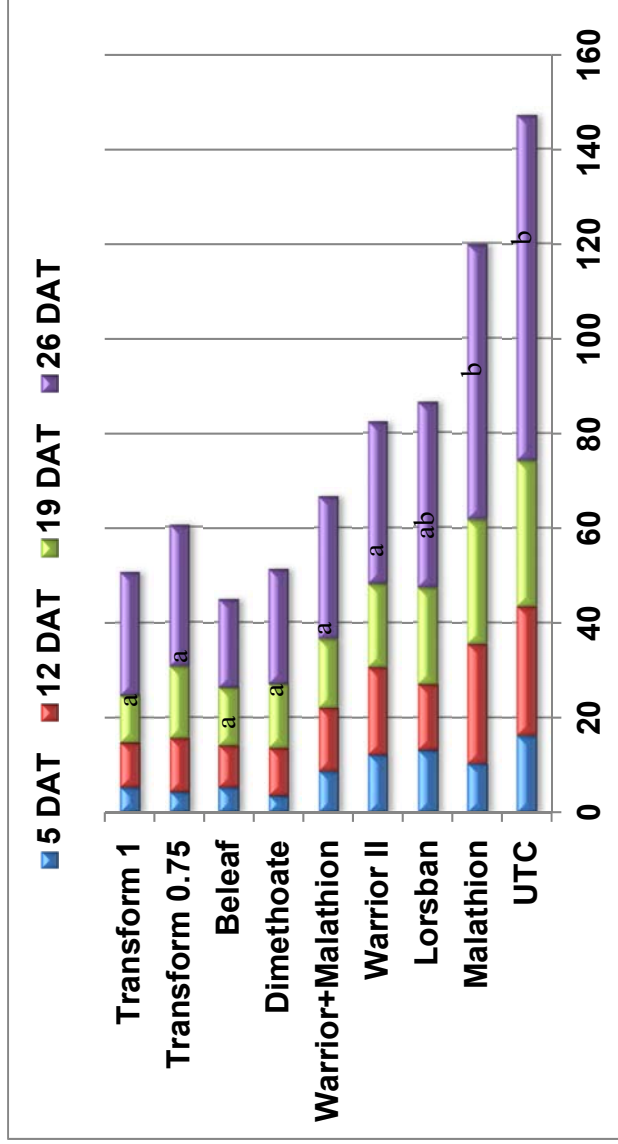


Fig. 4: Number of total aphids per stem for each treatment at 5, 12, 19 and 26 days after treatments (DAT) in the insecticide efficacy trial (I) against aphids in alfalfa at Maricopa Agricultural Center in 2014.

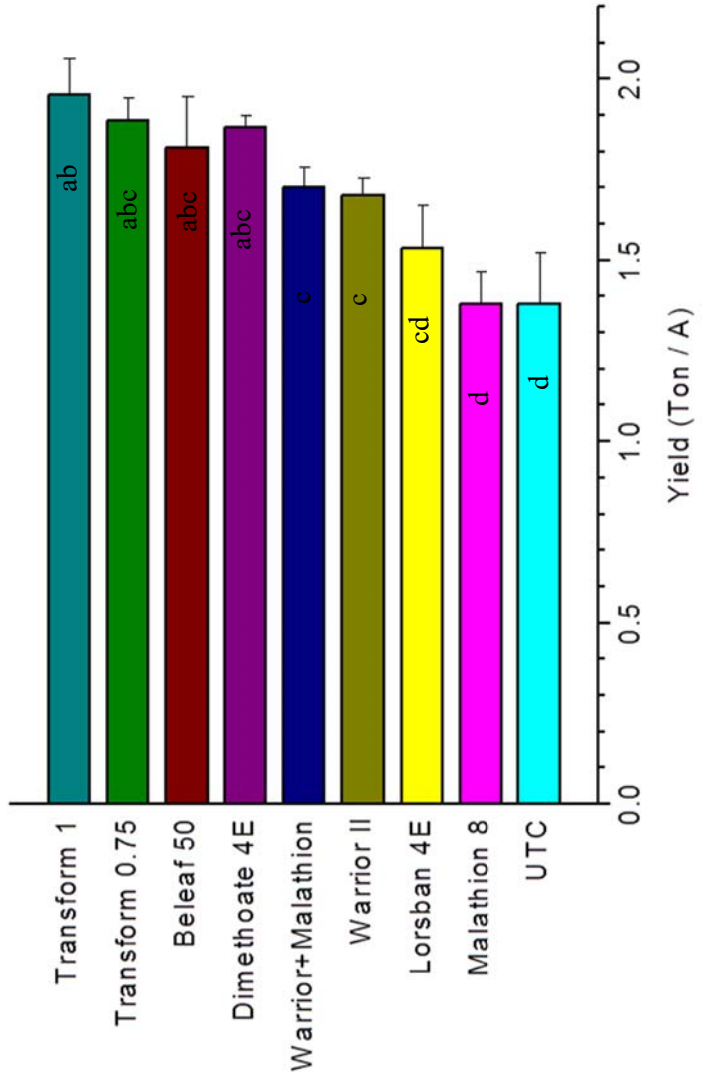


Fig. 5: Mean hay yield (based on 12% moisture) for the insecticide efficacy trial (I) against aphids in alfalfa at Maricopa Agricultural Center in 2014.

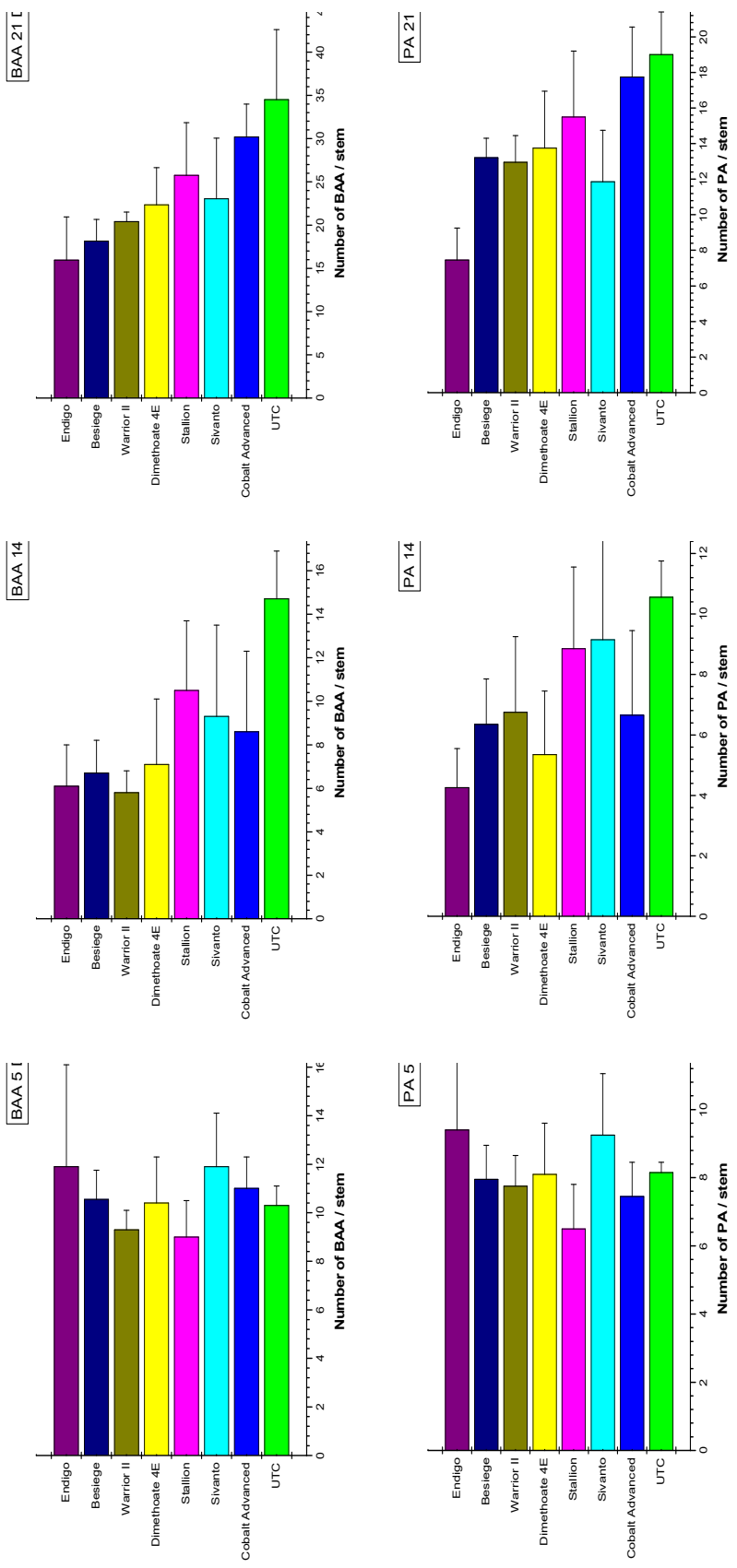


Fig. 6: Number of total aphids per stem for each treatment at 5, 14, and 21 days after treatments (DAT) in the insecticide efficacy trial (II) against PA and BAA in alfalfa at Maricopa Agricultural Center in 2014

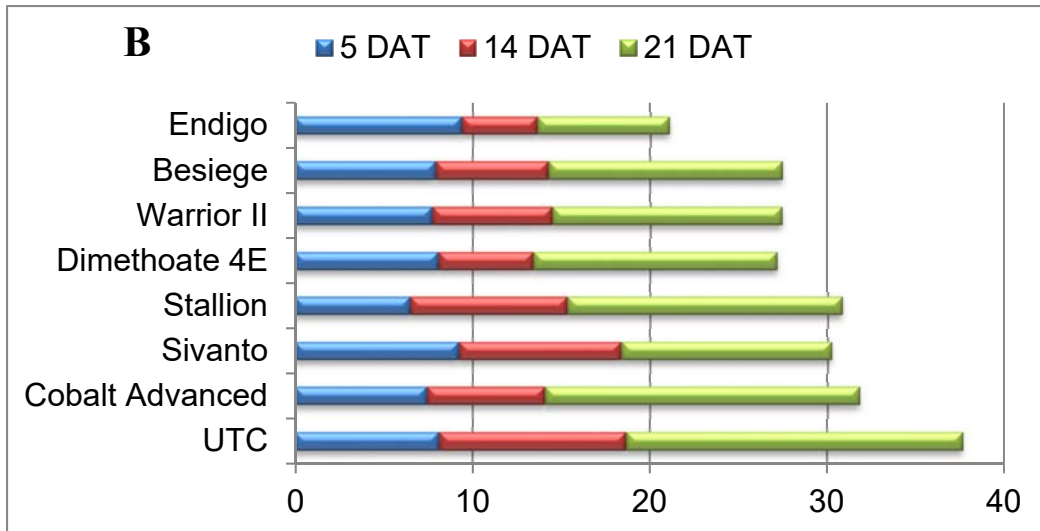
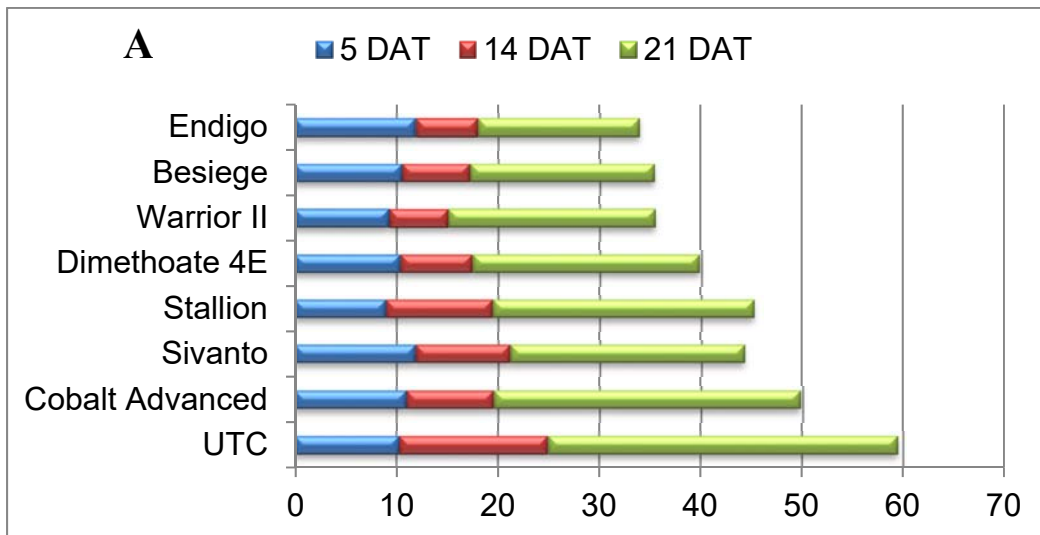


Fig. 7: Seasonal mean (3 post-spray dates) of total aphids per stem (A = BAA, B = PA & C = CPA) for each treatment in the insecticide efficacy trial (II) against aphids in alfalfa at Maricopa Agricultural Center in 2014

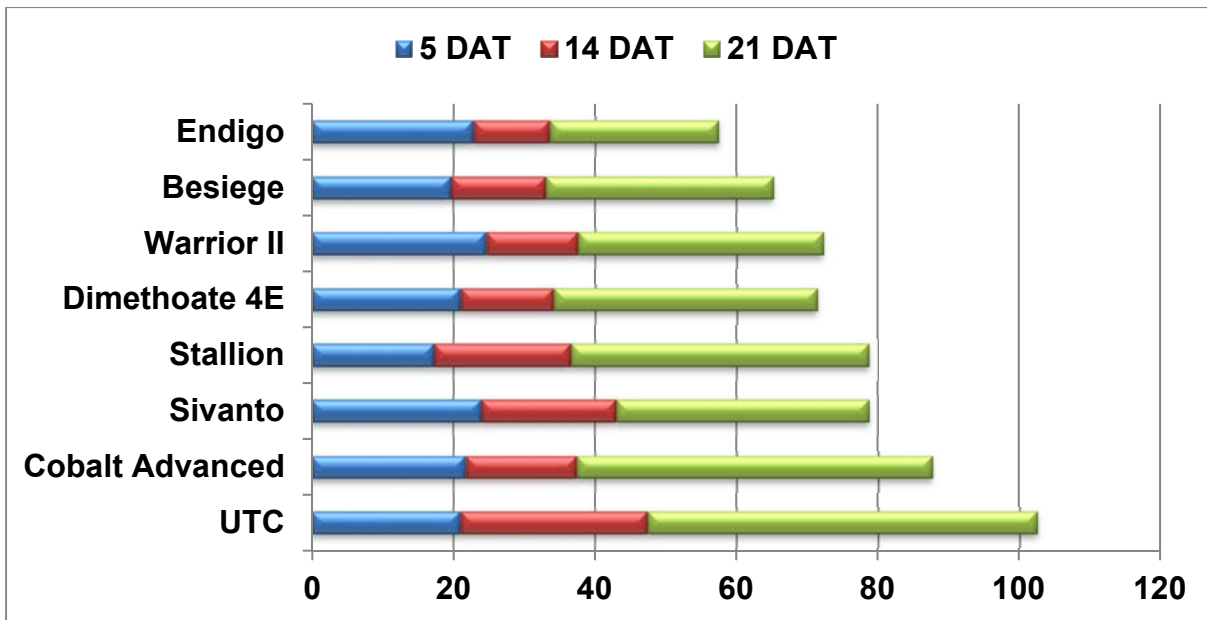


Fig. 8: Number of total aphids per stem for each treatment at 5, 14, and 21 days after treatments (DAT) in the insecticide efficacy trial (II) against aphids in alfalfa at Maricopa Agricultural Center in 2014.

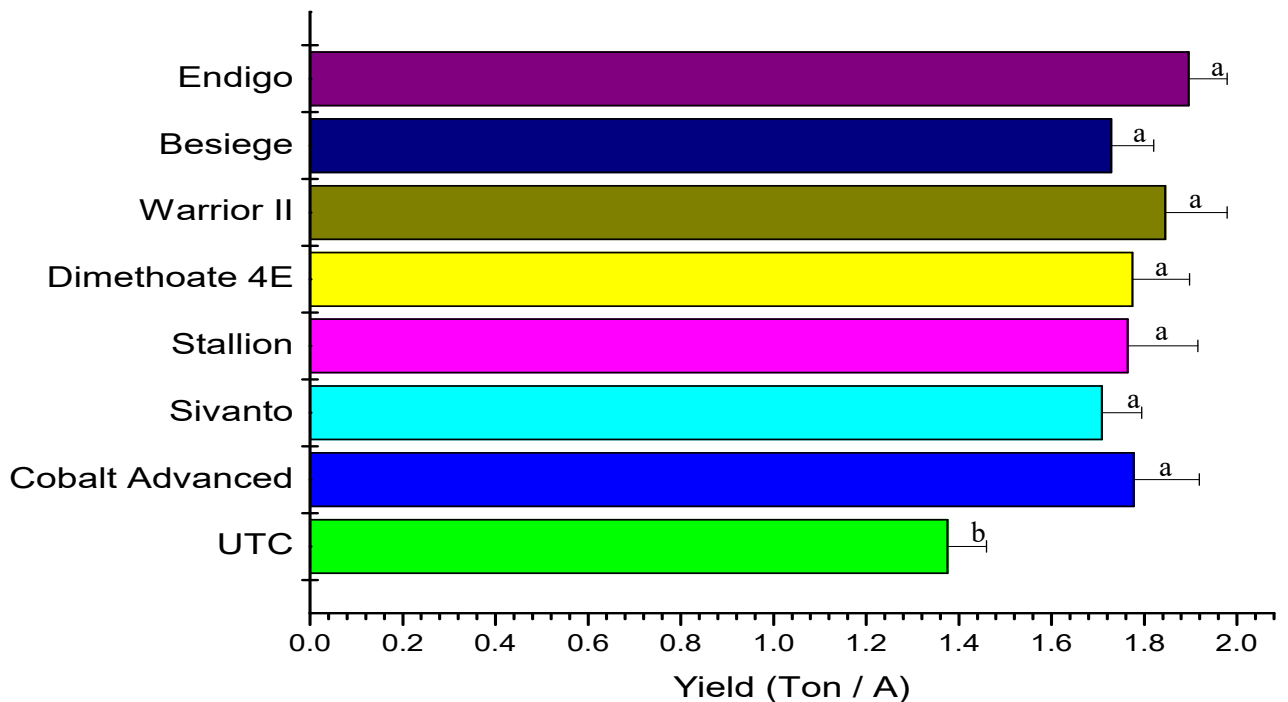


Fig. 9: Mean hay yield (based on 12% moisture) for the insecticide efficacy trial (II) against aphids in alfalfa at Maricopa Agricultural Center in 2014

Table 1. Statistical Analysis for Insecticides Treatment against Blue Alfalfa and Pea Aphids at Different Days of Sampling after Treatment in 2015.

Treatment	Days After Treatment (DAT)											
	5			11			18			25		
	BAA	PA		BAA	PA		BAA	PA		BAA	PA	
Beleaf50(2.85)	4.50 B*	4.50 BC		3.75 AB	14.25 B		1.75 AB	10.00 C		3.50 BC	18.75 BC	
Besiege9	4.00 B	11.00 ABC		7.25 AB	1.00 B		3.00 AB	11.00 C		5.00 BC	19.50 BC	
CobaltAdvanced24	2.00 B	1.25 BC		4.50 AB	7.00 B		1.00 B	19.25 ABC		8.00ABC	28.75 ABC	
Dimethoate4E1	2.00 B	14.00ABC		7.00 AB	27.75 B		3.00 AB	23.00 ABC		4.75 BC	17.00 BC	
Endigo4.5	1.75 B	9.75 ABC		5.00 AB	15.50 B		3.50 AB	24.25 AB		11.00 AB	24.25 ABC	
Pyrifluquinazon1.6	3.50 B	14.00ABC		4.00 AB	14.50 B		1.00 B	11.75 BC		7.75 ABC	22.50 ABC	
Pyrifluquinazon3.2	3.50 B	19.00 AB		4.25 AB	15.00 B		1.25 AB	20.25 ABC		3.50 BC	16.75 BC	
Sivanto10.5	0.75 B	0.50 C		5.50 AB	11.75 B		2.25 AB	17.50 ABC		4.50 BC	9.75 BC	
Sivanto7	2.00 B	2.25 BC		8.50 AB	15.25 B		3.00 AB	16.50 ABC		4.25 BC	16.25 BC	
Transform1	4.00 B	7.00ABC		3.00 B	23.00 B		1.00 B	20.75 ABC		7.50 ABC	41.25 ABC	
WarriorIII.92	1.75 B	3.00 BC		5.00 AB	22.25 B		2.00AB	19.00 ABC		7.25 ABC	33.25 ABC	
UTC	16.75 A	23.00 A		9.50 A	61.00 A		5.00 A	26.25 A		13.00 A	54.00 A	

*Means within columns followed by the same letter are not significantly different, LSD ; P= 0.05



Fig. 10: Aphids naturally affected by an entomopathogenic epizootics fungus in Central Arizona

Table 2. Percent population and infection of alfalfa aphids by fungus during the trial conducted in 2016 at Maricopa Agricultural Center (nd = no data; 10-Feb data were before spray)

Aphids' Population										
Date	BAA		PA		CP		SA		Stem Height	
	% Pop	% Infected	% Pop	% Infected	% Pop	% Infected	% Pop	% Infected	Inches	Inches
10-Feb	87	32	1	0	11	0	1	0	7	7
17-Feb	92	9	2	0	4	0	2	0	8	8
24-Feb	90	34	2	0	8	0	0	0	11	11
2-Mar	94	nd	2	0	3	0	1	0	12	12
9-Mar	96	nd	2	0	1	0	1	0	13	13

Table 3. Statistical Analysis for Insecticides Treatment against Pea aphids, Blue Alfalfa, Cowpea aphids, and spotted alfalfa aphids and seasonal total of aphids per stem in 2016

Treatment	Yield		Seasonal PA		Seasonal BAA ^{ns}		Seasonal CPA ^{ns}		Seasonal SAA ^{ns}		Seasonal Total	
	Tons/A	Seasonal PA	Seasonal PA	Seasonal PA	Seasonal BAA ^{ns}	Seasonal CPA ^{ns}	Seasonal CPA ^{ns}	Seasonal SAA ^{ns}	Seasonal SAA ^{ns}	Seasonal Total	Seasonal Total	
Endigo ZCX 4.5oz/A	1.08 ABC	18 BC	1	1	1	0	0	0	0	20 B	20 B	
Temitry EC 18oz/A	1.25 A	24 BC	1	1	1	0	0	0	0	26 B	26 B	
Beleaf 50SG 2.85oz/A	1.08 ABC	26 BC	0	2	0	0	0	0	0	28 B	28 B	
Transform50WG 1oz/A	0.98 BCD	29 BC	1	1	1	0	0	0	0	31 B	31 B	
Mustang EW 4.3oz/A	1.12 AB	40 ABC	1	2	1	0	0	0	0	43 AB	43 AB	
Dimethoate 4E 16oz/A	1.00 BCD	15 C	1	2	1	0	0	0	0	18 B	18 B	
Sivanto SL 10.5oz/A	0.88 CD	47 AB	1	1	1	0	0	0	0	49 AB	49 AB	
Cobalt Adv'cd 32oz/A	1.25 A	44 ABC	1	1	1	0	0	0	0	46 AB	46 AB	
Untreated Control	0.81 D	61 A	1	1	1	0	0	0	0	63 A	63 A	

*Means within columns followed by the same letter are not significantly different, LSD ; P= 0.05; ns-non significant

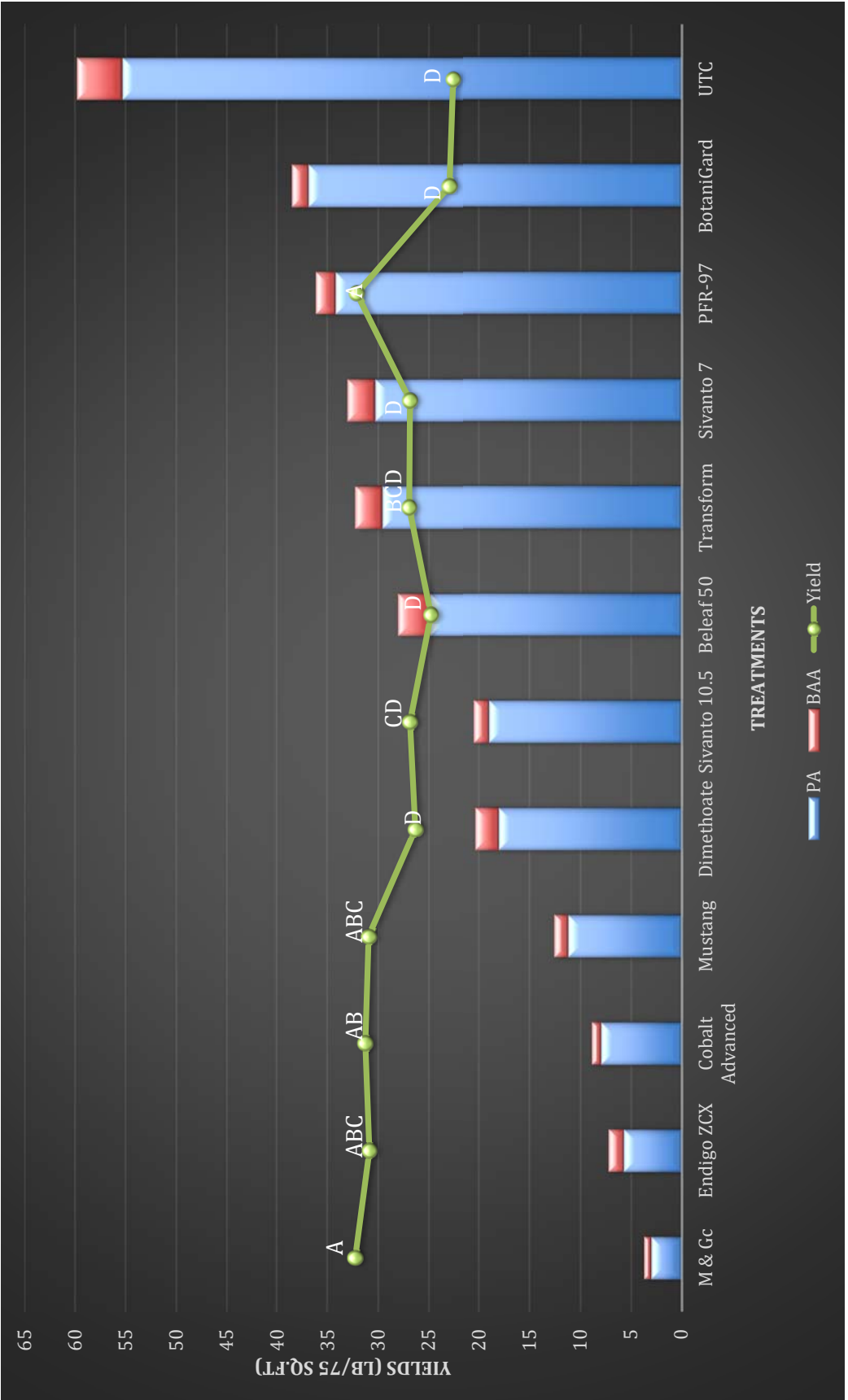


Fig. 11. The effect of total seasonal on hay yield in the 2017 insecticide efficacy trial at Maricopa Agricultural Center (MAC). Means of yield with the same letter are not significantly different from each other at alpha 0.05.

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