TOXICITY OF AQUEOUS FORMULATIONS OF BIFENTHRIN AND MALATHION TO BOLL WEEVIL

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Abstract

Toxicity of technical bifenthrin and malathion against the laboratory reared boll weevil, Anthonomus grandis (Boheman), were greater when topically applied in acetone to dorsum thorax than in solvent-emulsifier concentrate (EC) formulations in aqueous. Toxicity of bifenthrin (EC) formulation (in petroleum solvent) in Milli-Equivilant (MQ) water for dead category were significantly less than for dead + moribund category after 1 and 24 h in solution whether topically applied, tarsi were wiped in aqueous or in dry droplet. Topical application of malathion (in petroleum solvent) after 1 h in solution with MQ or domestic water showed poor toxicity.

Malathion ready-to-use (RTU) EC (in cotton seed oil solvent), was more toxic than EC petroleum solvent formulation when topically applied. A tarsal wipe in droplet or in dry droplet of RTU malathion formulation was less toxic than when the same formulation was topically applied. Malathion EC, in petroleum and cottonseed oil solvent and in pH_{10} solution, was less toxic to boll weevil than in pH_7 buffer topically applied. Both formulations were more toxic in pH_4 buffer than pH_7 buffer.

Bifenthrin and malathion are registered for control of boll weevil, Anthonomus grandis (Boheman) on cotton and their formulations can be diluted in water. Quality of this water used to mix with bifenthrin is not considered to be a major problem. Indeed, cypermethrin, a related pyrethroid insecticide, had a half life of > 8544 h at pH_8 and 25°C (Chapman & Cole, 1982).

Malathion is not considered to be as effective in a water based spray formulation against the boll weevil than as technical without water. It is known that 50% of malathion hydrolyzes in aqueous alkaline (in $pH_{10.79}$ and 0°C) to O, O-dimethyl phosphorodithiodic acid and the mono and di-acids of malathion in 2.4 h (Wolfe et al. 1977). In aqueous acid 50% malathion (pH 2.59) hydrolyzes to the acids in 1560 h; at pH_8 and 36 h, 50% malathion is hydrolyzed at 27°C (Wolfe et al. 1977). Loss in toxicity to boll weevil should follow the degradation of malathion to its hydrolytic metabolites but no data has been found to support this hypothesis

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No information has been found to indicate that toxicity of the boll weevil to bifenthrin and malathion in formulation with water will remain the same when topically applied to dorsum of thorax or their tarsi are wiped in a droplet of water. I propose that reduced contact with the cuticle of the dorsum of thorax and tarsi of the boll weevil and thus reduced toxicity of bifenthrin or malathion can occur in a formulation with surfactant(s) and solvent and water. This is possible when the emulsifiable concentrate (EC) is in a water droplet placed on the cuticle of the dorsum of thorax and a surface for the tarsi to contact, or when the droplet was allowed to dry before the tarsi were contacted.

Materials and Methods

Pure technical bifenthrin (100%) (FMC Corp, Princeton NJ) and malathion (98%) (Cheminova Corp., Copenhagen, Denmark) were diluted in acetone.
Bifenthrin was tested as 240 g/liter (2 lbs/gal) (24.2%) EC. EC malathion was formulated in petroleum solvent as 572 g/l (4.76 lbs./gal) (50%) with emulsifier(s). Ready to use (RTU) EC malathion was formulated in cottonseed oil solvent as 556 g/l (4.63 lbs./gal) (47.6%) with emulsifier(s).

Tests were conducted at 25-29°C. Solutions of $pH_{4, 7}$, and $_{10}$ were standard buffers. Domestic water of 720 ppm total soluble salt ($pH_{6.73}$) was tested. Milli-equivalent (MQ) water had <20 ppm total soluble salts $pH_{6.43}$ and an 18.2 m Ohms resistance /cm^2. Domestic and MQ water were collected once, analyzed for pH and used in all tests. MQ water was considered to be pure enough so it would not hydrolyze either insecticide after 1 h in solution. Bifenthrin was tested in MQ water only because hydrolytic effects of water are unknown. Malathion formulated in petroleum solvent was tested with all buffers and waters: Malathion formulated in cottonseed oil solvent was tested with all the above solutions except domestic water.

Bifenthrin was diluted in one microliter of acetone at 1, 0.5, 0.25, 0.0625, 0.031, 0.0155, 0.00775, 0.003875, 0.0019, 0.00095, and 0.0000475 and malathion at 5, 2.5, 1.25, 0.625, 0.31, 0.155, and 0.0775 µg/boll weevil. Both insecticides in the formulations were treated with most of these same dosages in all experiments.

The laboratory reared strain boll weevil adults were obtained from Gast Rearing Laboratory, Mississippi State University and used in all tests.

Three to seven day old weevils were fed diet and sugar water (5%) before testing. Weevils of both sexes were topically treated on the dorsum of the thorax with bifenthrin and malathion in one microliter acetone to determine LD_{50} value by basic methods of Anonymous (1968). Topical tests were conducted after 1 h and 24, 48 and 72 h with insecticide formulations in solution with the various waters. The 1 h test was conducted to determine if the physical properties of water -surfactant -solvent alone was responsible for loss in toxicity. If no difference in toxicity occurred between 1 h vs. 24, 48 and 72 h with MQ water, then acidity or alkalinity made no difference.

Boll weevil were also treated by wiping tarsi in liquid microliter droplet or in dry area of one microliter droplet with the same solutions at the same times topical applications were made. Tarsi wipes occurred when all tarsi were wiped for 5 seconds in 1 microliter solutions with water of bifenthrin and malathion. Weevils were held upright with forceps during the treatment. The test was conducted to determine if disturbing the meniscus of the water-solvent (petroleum or cotton seed oil)-emulsifier-malathion and bifenthrin in one formulation would allow sufficient contact with the toxicant in the droplet to the tarsi to cause mortalities equal to or greater than technical bifenthrin and malathion in acetone. Another 1 µ1 droplet of water-solvent-emulsifier-malathion solution was placed on a glass surface and allowed to evaporate for 2-3 h. The tarsi was then rubbed back and forth for 5 seconds in the residue of the droplet.

With the three methods ten to 20 weevils were treated with each of four to six dosages in each of two or three replicates. Following treatment weevils were placed individually in 30 ml plastic cups with caps. Determination of moribund, dead and live categories were made 48 h. posttreatment. Live weevils walked upright in their cups. Dead weevils did not move any body part upon probing. Moribund weevils did not walk and moved only legs and proboscis when probed.

LD_{50}, Slope ± SE and 95% confidence interval (C.I.) by the three methods of treating were determined by SAS (1989). Where 95% C.I. overlapped LD_{50} valves were not significantly different. Where ratio of slope/standard error (shown here as slope ± SE) equal $t_{0.05}$, $\leq$ 1.96 for ∞, the regression was not significantly different from zero. When either the dead or dead + moribund category show a nonsignificant regression none or few of the insects were determined to be in the category or large variation in response to the dosage selected occurred for those replicates with that sample of the laboratory reared strain.

Results and Discussion

Topical applications of technical bifenthrin-acetone were equally toxic (95% C.I. overlapped) to test insects in dead or dead + moribund condition (Table 1). Replicates were conducted five times, six days apart, with the same solution and the data were totalled for LD_{50} value. The first test was made 1 h after mixing technical with the acetone. After 1 h of EC bifenthrin in MQ water LD_{50} values for dead + moribund adults were equally toxic with LD_{50} shown for bifenthrin-acetone. LD_{50} values for topical applications by bifenthrin diluted in acetone were significantly less than those shown for bifenthrin EC in MQ for the dead category after 1, 24 and 48 h in solution. Topical applications of bifenthrin-MQ water showed significantly greater LD_{50} values after 1 and 24 h for dead adults than dead + moribund adults. This means that bifenthrin EC in MQ water did not kill enough insects compared to moribund insects for equal LD_{50} values. I also suggest that less bifenthrin EC contacted the cuticle when diluted in aqueous solutions than when technical was diluted in acetone. Bifenthrin-MQ showed a nonsig-nificant regression for dead + moribund after 72 h; it was 1.71 ± 1.15 for 198 insects.

Nonsignificant regression of tarsi wipe in aqueous droplet after 1 h (for 260 insect) as ratio of slope ± SE were 0.47 ± 0.25 and -0.11 ± 0.34 and 48 h, 24 h (for 216 insects) as 0.42 ± 0.40 and 0.56 ± 0.46 for dead + moribund and dead, respectively (data not shown in table). For dead category after 24 h in solution a nonsignificant regression of 0.061 ± 0.53 for 216 insects was determined (data not shown in table). LD_{50} of bifenthrin in MQ water when applied to tarsi after 24 and 72 h for dead + moribund were significantly greater than shown for bifenthrin in acetone. The same was true for dead category after 72 h in aqueous solution compared to bifenthrin in acetone.

LD_{50} values of bifenthrin after 24 and 48 h in solution as tarsi wipe in dry droplet for dead + moribund were statistically similar. LD_{50} values of tarsi wipe in dry droplets for dead + moribund and dead after 72 h in solution were significantly different. Nonsignificant regression of tarsi wipe in dry droplet was also shown for dead + moribund by bifenthrin in 1 hr as slope ± SE were 0.0022 ± 0.61 (for 194 insects) (data not shown in table). When tarsi was wiped over surface of glass of dry droplet a nonsignificant regression was shown for dead category after 1, 24 and 48 h as slope ± SE were -0.21 ± 0.28 (for 92 insects), -0.18 ± 0.41 (for 108 insects) and 0.88 ± 0.45 (for 79 insects), respectively (data not shown in table). The physical properties of the water solvent emulsifier appeared to be the only effect on toxicity with bifenthrin. Results also suggest that bifenthrin causes greater toxicity when the aqueous droplet contacts the body then when it contacts the tarsi.

Technical malathion-acetone topically applied had LD_{50} value of $\leq$ 0.25 (table 2). Six replicates were conducted over 8 weeks with the same acetone solution as described previously for bifenthrin.

Toxicity of malathion EC as a topical application with petroleum solvent in domestic and MQ water 1 h posttreatment showed a nonsignificant regression ($t_{0.05}$, $\leq$ 1.96) for dead + moribund and dead adults was determined (Table 2). With the same domestic water solutions topical applications taken after 24 and 48 h for dead + moribund showed nonsignificant regression of slope ± SE as 0.44 ± 0.98 and 1.03 ± 6.31, respectively, for 119 insects (data not shown in table). Results suggest that not enough malathion contacted the weevil cuticle when the EC formulation was diluted in water(s) because non-significant regression was indicated 1, 24 and 48 h.

Then we evaluated the same EC formulation as a tarsal wipe in a microliter of MQ water after 1 and 24 h in solution (Table 3). Both LD_{50} values for dead weevils were significantly greater than shown for dead of malathion-acetone. Moribund weevil populations were variable among the dosages tested which caused the high SE values for the dead + moribund category. These results suggest that reduced contact toxicity to tarsi is responsible for this difference.
Ratio of t $\leq$ 1.96, in MQ water showed nonsignificant regression for both topical (Table 2) and tarsi wipe (Table 3) for dead + moribund insects; ratio of slope ± SE were 0.28 ± 0.33 (for 155 insects) and 1.89 ± 0.98 (for 120 insects) by topical application and -0.64 ± 45 (for 136 insects) and 0.85 ± 0.60 (for 146 insects) by tarsi wipe 48 and 72 h posttreatment, respectively (data not shown in table).

LD_{50} values of the same formulation applied in pH $_{4}$, $_{7}$ and $_{10}$ buffer solutions were significantly greater (Table 4) at each time than malathion-acetone formulation (Table 2). LD_{50} values were determined after 1 (pH_{10}) and 1 and 24 h (pH_7) in solution except those in pH_4 which showed LD_{50} values after 1, 24 and 72 h (pH_4). Thus, malathion is more toxic in solutions of high hydrogen ion than either neutral or buffer of high hydroxyl ion concentration. LD_{50} values for dead + moribund of pH_4 ranged from a low of 3.17 µg/adult after 72 hr. in solution to a high of 38.86 µg/adult in 1 h for EC malathion with petroleum solvent in pH_4 buffer. Malathion in pH_4 showed nonsignificant regression for dead + moribund as slope ± SE of 11.24 ± 776.4 (for 120 insects) after 24 h (data not shown in table); after 48 h no dead insects were present thus no regression was determined. Malathion in pH_7 showed ratio of t $\leq$ 1.96 after 1 and 48 h of dead + moribund in solution as slope ± SE were 0.53 ± 0.92 (for 207 insects) and 0.43 ± 0.59 (for 210 insects), respectively (data not shown in table). For the dead category of similar pH, formulations and time nonsignificant regressions were determined as slope ± SE were 0.77 ± 1.14 and 0.73 ± 0.75, respectively (data not shown in table). These results show great variation and the greater pH values (both pH_7 and $_{10}$) reduce toxicity of malathion.

These results suggest that when water is mixed with malathion formulation, toxicity is reduced. Hydrolysis is a factor for reduced toxicity of malathion in formulation and diluted in buffers. Quality of water was important but effects were equal or less than equal to physical properties for these solvent-surfactant-toxicant solutions. If hydrolytic factors LD_{50} values would increase at each time period from 1 hr to 72 h.

Then we tested RTU cottonseed oil solvent EC formulation of malathion (Table 5) in MQ water solutions 1, 24, 48 and 72 h after mixing. LD_{50} values of RTU, topically applied, were significantly greater for dead and dead + moribund condition than shown for technical malathion in acetone (Table 2). LD_{50} of weevils in the two categories, determined at the same time were also similar after 1 and 24 h. Topical application of malathion in RTU to boll weevil in solution for 48 h showed dead + moribund condition equal to LD_{50} to topical application of malathion in acetone. This the only example shown for equal LD_{50} values. When in solution for 48 h, topical applications killed no adults. When in solution for 72 h topical application showed nonsignificant regressions for dead + moribund and dead as 0.69 ± 0.45 and 0.58 ± 0.44 (for 135 insects) were determined (data not shown in table).

When tarsi were wiped in MQ liquid or dry droplet, results were variable (Table 5). Tarsi wiped in droplet after 1 and 24 h in solution dead + moribund showed a nonsignificant regression based on ratio from 0.22 ± 0.29 (for 251 insects) and 0.30 ± 0.44, (for 190 insects), respectively (data not shown in table). Topical application and tarsi wipe in dry droplet showed similar LD_{50} values; they ranged from 5.45 to 7.73 µg/adult after 1 h for dead + moribund and dead larvae.

After 24 and 48 h in solution tarsi wipe of RTU malathion in dry droplet showed a nonsignificant regression were determined for dead + moribund insects as ratio of slope ± SE of 0.24 ± 0.19 (for 260 insects) and -0.38 ± 0.41 (for 185 insects), respectively (data not shown in table). For dead category by same treatments nonsignificant regression were 0.22 ± 0.19 and -0.74 ± 0.56 after 24 and 48, respectively, (data not shown in table). After 72 h nonsignificant regression was shown for dead as slope ± SE was 4.88 ± 3.15, for 75 insects, (data not shown in table).

LD_{50} by malathion of both formulations in solution with pH_4 buffer solutions were significantly different (Tables 4 and 6); toxicity of cottonseed oil formulation was greater in solutions with pH_4 than in solutions with petroleum solvent. At pH_7 LD_{50} values for malathion (Tables 4 and 6) in solutions with petroleum solvent showed values after 24 h but not 1, 48 or 72 h. For RTU formulation LD_{50} values in pH_4 were indicated 1, 24, 48 and 72 h (Table 6). At pH_{10} an LD_{50} was indicated at 1 h only with both formulations (Table 4 and 6). LD_{50} values for topical applications of RTU formulation of malathion in pH_4 and $_{7}$, and $_{10}$ after 1 h (Table 6) were greater than shown for technical malathion in acetone (Table 2). However, when topically applied, LD_{50} of buffer solutions of pH_4 and pH_{10} after 1 h were significantly less than shown for malathion in MQ water (Table 5). Results show no trend for greater LD_{50} after 1 to 72 h in solution at pH_4, with RTU formulation compared to LD_{50} values shown for petroleum solvent for the same times.

References

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Table 1. Toxicity after 48 h of technical bifenthrin in acetone and in MQ water topically applied to adult boll weevil, tarsi wipe in droplet and dry droplet.

Condition	Number Treated	Slope ± SE	LD50 (µg/adult)	(95% C. I.)
Topical Application				
In Acetone				
Dead + Moribund	580	0.65 ± 0.21	0.00072	(0.000018-0.0035)
Dead	580	0.75 ± 0.18	0.0092	(0.0032-0.060)
In Aqueous		After 1 h		
Dead + Moribund		0.35 ± 0.14	0.0017	($36x10^{-20}$-0.021)
Dead	664	0.41 ± 0.12	1.38	(0.38-73.62)
		After 24 h		
Dead + Moribund	456	1.13 ± 0.13	0.057	(0.039-0.079)
Dead		0.46 ± 0.098	0.35	(0.14-1.34)
		After 48 h		
Dead + Moribund	387	0.96 ± 0.21	0.021	(0.0056-0.045)
Dead		0.58 ± 0.16	0.045	(0.0089-0.15)
		After 72 h		
Dead	198	3.00 ± 1.24	0.066	(∞ - ∞)
Tarsi in Aqueous Droplet		After 24 h		
Dead + Moribund	252	1.67 ± 0.36	0.010	(0.0039-0.018)
		After 72 h		
Dead + Moribund	270	1.34 ± 0.22	0.077	(0.054-0.12)
Dead		0.69 ± 0.17	0.77	(0.25-14.61)
Tarsi in Dry Droplet		After 24 h		
Dead + Moribund	103	1.10 ± 0.52	0.015	(∞ - ∞)
		After 48 h		
Dead + Moribund	108	0.93 ± 0.28	0.021	(0.0026-0.045)
		After 72 h		
Dead + Moribund	108	2.47 ± 0.41	0.058	(0.04-0.077)
Dead		1.19 ± 0.28	0.25	(0.15-0.56)

Table 2. Toxicity after 48 h of technical malathion in acetone and emulsifiable concentrate (in petroleum solvent) at indicated hours in solution topically applied to dorsum thorax of adult boll weevil.

Condition Weevil	Number Treated	Slope ± SE	LD_{50} (µg/adult)	(95% CI)
		Technical (in Acetone)		
Dead + Moribund	1130	1.22 ± 0.23	0.12	(0.031-0.25)
Dead		1.89 ± 0.45	0.25	(0.093-0.41)
		After 1 h in domestic water		
Dead + Moribund	119	-0.25 ± 0.44[a]		
Dead		-0.046 ± 0.43[a]		
		After 1 h in MQ water		
Dead + Moribund	228	0.88 ± 0.46[a]		
Dead		0.37 ± 0.94[a]		

[a]> Ratio of slope/SE t >0.05, ≤1.96 indicated regression was not significantly different from zero.

Table 3. Toxicity after 48 h of malathion (in petroleum solvent) in MQ water at indicated hrs. in solution as tarsi wipe in 1µl droplet to "ebony" laboratory reared boll weevil.

Condition Weevil	Number Treated	Slope ± SE	LD_{50} (µg/adult)	(95% Confidence Interval)
		After 1 h in MQ water		
Dead + Moribund	264	0.66 ± 0.37[a]		
Dead		0.98 ± 0.49	20.32	(6.52-7.99)
		After 24 h in MQ water		
Dead + Moribund	100	1.84 ± 1.27[a]		
Dead		1.15 ± 0.40	3.39	(2.03-14.91)

[a] Ratio of slope/SE t >0.05, ≤1.96 indicted regression was not significantly different from zero.

Table 4. Toxicity after 48 h of malathion (in petroleum solvent) in solutions topically applied to "ebony" laboratory reared boll weevil at indicated hrs. in solution of different pH equivalents.

pH Buffer and Condition Weevil	Number Treated	Slope ± SE	LD_{50} (µg/weevil)(95% C.I.)
pH_4		After 1 h	
Dead + Moribund	362	0.87 ± 0.32	38.86 (11.29-83247)
Dead		0.71 ± 0.29	76.72 (14.76-470140469)
		After 48 h	
Dead + Moribund	240	0.80 ± 0.38	30.53 (7.99-$1.8x10^{14}$)
		After 72 h	
Dead + Moribund	120	1.40 ± 0.42	3.17 (2.06-7.67)
Dead		1.51 ± 0.40	4.75 (3.09-13.33)
pH_7		After 24 h	
Dead + Moribund	286	1.21 ± 0.29	3.72 (2.77-6.44)
Dead		1.18 ± 0.32	4.19 (3.0-8.65)
pH_{10}		After 1 h	
Dead + Moribund	120	1.14 ± 0.40	8.62 (4.66-137.06)
Dead		1.66 ± 0.49	9.86 (5.81-55.69)

[a]> Ratio of slope/SE t> 0.05, ≤1.96 indicated regression was not significantly different from zero.

Table 5. Toxicity after 48 h of (RTU) malathion at indicated hr in MQ water solution to "Ebony" laboratory reared adult boll weevil.

Condition Weevil	Number Treated	Slope ± SE	LD_{50} (µg/adult)	(95% C.I.)
Topical Application		After 1 h		
Dead + Moribund	185	1.24 ± 0.41	7.52	(4.06-86.74)
Dead		1.06 ± 0.92	7.73	(4.01-63.71)
		After 24 h		
Dead + Moribund	165	1.16 ± 0.34	12.08	(5.77-141.83)
Dead	1.43 ± 0.61		13.67	(∞ - ∞)
		After 48 h		
Dead + Moribund	75	1.44 ± 0.42	0.82	(0.33-1.40)
Tarsi in Droplet		After 48 h		
Dead + Moribund	125	0.85 ± 0.29	6.65	(2.97-181.37)
Dead		1.06 ± 0.31	6.30	(3.21-47.07)
Tarsi in Dry Droplet		After 1 h		
Dead + Moribund	244	0.44 ± 0.19	5.45	(2.07-418.34)
Dead	0.45 ± 0.19		7.27	(2.57-59205)
		After 72 h		
Dead + Moribund	75	5.06 ± 2.24	3.30	(1.15-4.87)

Table 6. Toxicity after 48 h of malathion (RTU) topically applied at indicated hr in solution to "ebony" laboratory reared boll weevil after 48 h.

pH Buffer and Condition Weevil	Number Treated	Slope ± SE	LD_{50} (µg/adult) (95% C.I.)
pH_4		After 1 h	
Dead + Moribund	120	1.71 ± 0.41	0.91 (0.48-1.29)
Dead		2.25 ± 0.43	1.28 (0.94-1.67)
		After 24 h	
Dead + Moribund	120	1.31 ± 0.37	3.25 (2.14-7.85)
Dead		0.99 ± 0.41	13.94 (5.29-47706)
		After 48 h	
Dead + Moribund	135	1.72 ± 0.51	1.15 (0.011-5.59)
Dead		1.45 ± 0.48	1.29 (∞ - ∞)
		After 72 h	
Dead + Moribund	124	0.62 ± 0.34[a]	
Dead		0.79 ± 0.34	4.05 (2.06-522.77)
pH_7		After 1 h	
Dead + Moribund	122	1.26 ± 0.61	1.17 (∞ - ∞)
Dead		1.37 ± 0.38	1.83 (1.14-2.99)
		After 24 h	
Dead + Moribund	195	0.72 ± 0.25	2.64 (1.49-10.33)
Dead		0.89 ± 0.26	5.0 (2.86-24.03)
		After 48 h	
Dead + Moribund	210	1.74 ± 0.35	1.91 (1.34-2.74)
Dead		1.89 ± 0.39	2.56 (1.84-3.78)
		After 72 h	
Dead + Moribund	195	0.80 ± 0.27	3.14 (1.78-14.71)
Dead		0.78 ± 0.28	3.64 (2.01-26.43)
pH_{10}			
Dead+ Moribund	120	1.51 ± 0.71	0.85 (∞ - ∞)
Dead		1.53 ± 0.74	1.81 (∞ - ∞)

[a] > Ratio of slope/SE >0.05, ≤ 1.96 indicated regression was not significantly different from zero.