

Binary Mixture of Neonicotinoid-Pyrethroid Insecticide: Impact on Survival, Cognitive Learning, and Memory in *Apis mellifera jemenitica*

Mohamedazim I. B. Abuagla ¹, Javaid Iqbal ¹, Hael S. A. Raweh ¹, Abdelsalam S. A. Abdelaziz ² and Abdulaziz S. Alqarni ^{1,*}

Supplementary Tables

SUPPLEMENTARY TABLE S1: IM topical exposure and bee mortality (For Figure 1)

Table S1-A: ANOVA for bee morality at individual times

Treatment (IM-Concentrations)*	Time	df	Sum of Squares	Mean Square	F Value	P
Between Concentrations	4 h	4	19430	4857	132.48	<.0001
Between Concentrations	12 h	4	26920	6730	175.57	<.0001
Between Concentrations	24 h	4	29030	7257	435.45	<.0001
Between Concentrations	48 h	4	30020	7505	409	<.0001

*C1(control) = 0 ppm; C2 = 24.60 ppm; C3 = 49.50 ppm; C4 = 98.25 ppm; C5 = 196.50 ppm

Table S1-B: LSD All-Pairwise Comparisons

Time	Treatment (IM-Concentrations)*	Mean Corrected Mortality (%)	Significance ($p \leq 0.05$)
4 h	C1	0.00	d
	C2	57.50	c
	C3	70.00	b
	C4	80.00	a
	C5	88.00	a
12 h	C1	0.00	d
	C2	70.00	c
	C3	88.00	b
	C4	95.00	ab
	C5	100	a
24 h	C1	0.00	d
	C2	75.00	b
	C3	97.50	a
	C4	97.50	a
	C5	100	a
48 h	C1	0.00	d
	C2	78.00	b
	C3	100	a
	C4	100	a
	C5	100	a

*C1(control) = 0 ppm; C2 = 24.60 ppm; C3 = 49.50 ppm; C4 = 98.25 ppm; C5 = 196.50 ppm

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SUPPLEMENTARY TABLE S2: IM oral exposure and bee mortality (For Figure 2)

Table S2-A: ANOVA for bee morality at individual times

Treatment (IM-Concentrations)*	Time	df	Sum of Squares	Mean Square	F Value	P
Between Concentrations	4 h	4	11000	2750	41.25	<.0001
Between Concentrations	12 h	4	21020	5255	34.65	<.0001
Between Concentrations	24 h	4	22480	5620	38.76	<.0001
Between Concentrations	48 h	4	24950	6237.5	35.64	<.0001

*C1 (control) = 0 ppm; C2 = 5.65 ppm; C3 = 11.30 ppm; C4 = 22.65 ppm; C5 = 45.30 ppm

Table S2-B: LSD All-Pairwise Comparisons

Time	Treatment (IM-Concentrations)*	Mean Corrected Mortality (%)	Significance ($p \leq 0.05$)
4 h	C1	0.00	d
	C2	15.00	c
	C3	30.00	b
	C4	35.00	b
	C5	70.00	a
12 h	C1	0.00	d
	C2	22.00	c
	C3	45.00	b
	C4	60.00	b
	C5	95.00	a
24 h	C1	0.00	d
	C2	35.00	c
	C3	52.50	bc
	C4	70.00	b
	C5	100	a
48 h	C1	0.00	d
	C2	37.00	c
	C3	67.50	b
	C4	82.50	ab
	C5	100	a

*C1 (control) = 0 ppm; C2 = 5.65 ppm; C3 = 11.30 ppm; C4 = 22.65 ppm; C5 = 45.30 ppm

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SUPPLEMENTARY TABLE S3: Food consumption after IM oral exposure (For Figure 3)

Table S3-A: ANOVA for food consumption

Treatment (IM-Concentrations)*	df	Sum of Squares	Mean Square	F Value	P
Between Concentrations	4	2332	583	5.40	<.0.0067

*C1 (control) = 0 ppm; C2 = 5.65 ppm; C3 = 11.30 ppm; C4 = 22.65 ppm; C5 = 45.30 ppm

Table S3-B: LSD All-Pairwise Comparisons

Treatment (IM-Concentrations)*	Mean (% Food Consumption)	Significance
C1	100	a
C2	83.00	b
C3	71.00	c
C4	62.00	c
C5	44.00	d

*C1 (control) = 0 ppm; C2 = 5.65 ppm; C3 = 11.30 ppm; C4 = 22.65 ppm; C5 = 45.30 ppm

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SUPPLEMENTARY TABLE S4: IM topical exposure and PER (For Figure 6)

Learning and memory formation			Fisher's exact test/ χ^2 test value	Significance ($p<0.05$)
Learning Trials	2 nd	C1 vs C2	0.512	n.s
		C1 vs C3	0.044	Sig.
		C1 vs C4	0.033	Sig.
		C2 vs C3	0.712	n.s
		C2 vs C4	0.572	n.s
		C3 vs C4	0.844	n.s
	3 rd	C1 vs C2	0.542	n.s
		C1 vs C3	0.042	Sig.
		C1 vs C4	0.041	Sig.
		C2 vs C3	0.081	n.s
		C2 vs C4	0.150	n.s
		C3 vs C4	1.000	n.s
Memory	2 h	C1 vs C2	0.024	Sig.
		C1 vs C3	0.033	Sig.
		C1 vs C4	0.016	Sig.
		C2 vs C3	1.000	n.s
		C2 vs C4	0.856	n.s
		C3 vs C4	0.854	n.s
	12h	C1 vs C2	0.043	Sig.
		C1 vs C3	0.039	Sig.
		C1 vs C4	0.015	Sig.
		C2 vs C3	0.840	n.s
		C2 vs C4	0.680	n.s
		C3 vs C4	0.832	n.s
	24 h	C1 vs C2	0.280	n.s
		C1 vs C3	0.042	Sig.
		C1 vs C4	0.010	Sig.
		C2 vs C3	0.677	n.s
		C2 vs C4	0.675	n.s
		C3 vs C4	1.000	n.s

*C1 (control) = Acetone; C2 = LC₁₀ (3.75 ppm); C3 = LC₂₀ (5.63 ppm); C4 = LC₃₀ (7.54 ppm). Sig. stands for Significant; n.s stands for non-significant.

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SUPPLEMENTARY TABLE S5: IM oral exposure and PER (For Figure 7)

Learning and memory formation			Fisher's exact test/ χ^2 test value	Significance ($p<0.05$)
Learning Trials	2 nd	C1 vs C2	0.558	n.s
		C1 vs C3	0.180	n.s
		C1 vs C4	0.033	Sig.
		C2 vs C3	0.456	n.s
		C2 vs C4	0.117	n.s
		C3 vs C4	0.380	n.s
	3 rd	C1 vs C2	0.270	n.s
		C1 vs C3	0.043	Sig.
		C1 vs C4	0.024	Sig.
		C2 vs C3	0.750	n.s
		C2 vs C4	0.200	n.s
		C3 vs C4	0.340	n.s
Memory	2 h	C1 vs C2	0.161	n.s
		C1 vs C3	0.026	Sig.
		C1 vs C4	0.018	Sig.
		C2 vs C3	0.618	n.s
		C2 vs C4	0.214	n.s
		C3 vs C4	0.457	n.s
	12h	C1 vs C2	0.148	n.s
		C1 vs C3	0.005	Sig.
		C1 vs C4	0.012	Sig.
		C2 vs C3	0.460	n.s
		C2 vs C4	0.289	n.s
		C3 vs C4	0.741	n.s
	24 h	C1 vs C2	0.245	n.s
		C1 vs C3	0.006	Sig.
		C1 vs C4	0.019	Sig.
		C2 vs C3	0.146	n.s
		C2 vs C4	0.104	n.s
		C3 vs C4	0.832	n.s

*C1 (control) = Sucrose; C2 = LC₁₀ (2.45 ppm); C3 = LC₂₀ (4.04 ppm); C4 = LC₃₀ (5.78 ppm). Sig. stands for Significant; n.s stands for non-significant.

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Supplementary Figure

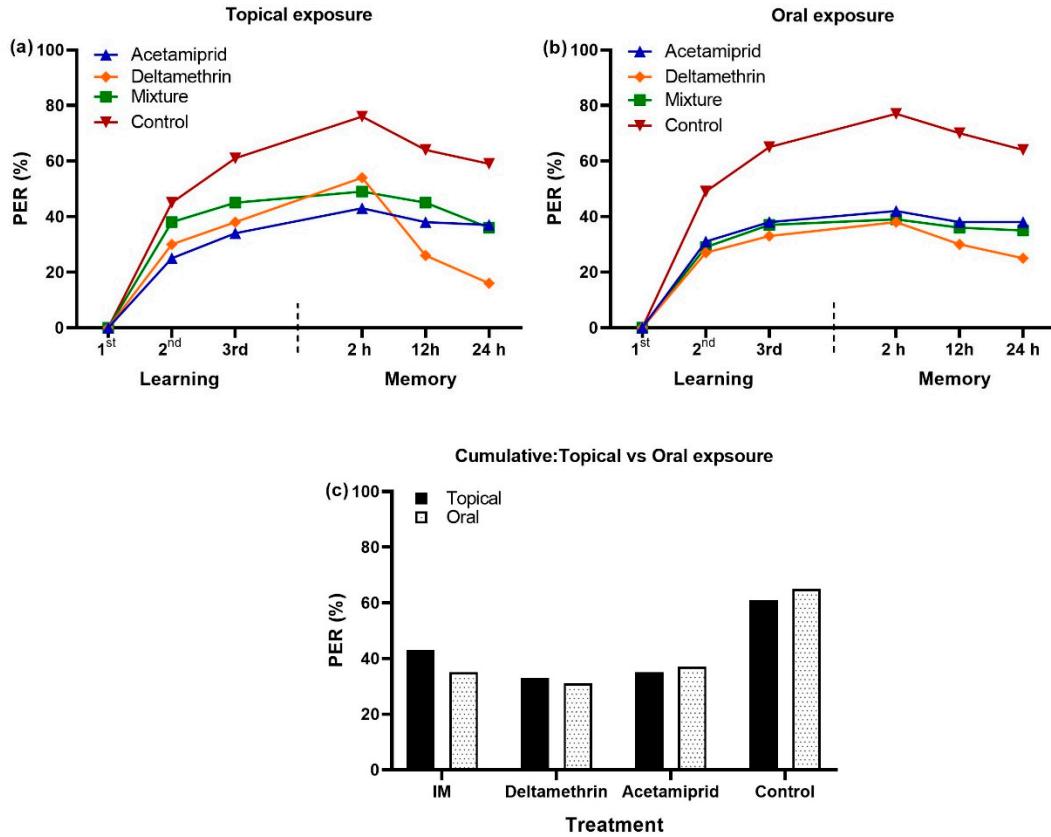


Figure S1. Comparison of PER in response to acetamiprid-deltamethrin mixture and individual insecticides: a) Topical exposure, b) Oral exposure c) Cumulative comparison: Topical Vs. oral exposure.