TOXICITY AND HYPOXIA OF THREE PETROLEUM HYDROCARBONS AND COTTONSEED OIL TO ADULT BOLL WEEVILS[1] AND LARVAE OF TOBACCO BUDWORMS[2]

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ABSTRACT

Topical applications of three petroleum hydrocarbons, at concentrations of 31% to 54% were lethal to 50% of tested boll weevils, Anthonomus grandis Boheman, and concentrations of 3% to 40% of one of the hydrocarbons caused hypoxia (significantly reduced oxygen consumption) in boll weevils. The pyrethroid insecticide, permethrin, was significantly more effective against the boll weevil when diluted in Orchex® 796 than when diluted in acetone or cottonseed oil. The three petroleum materials caused 69% to 96% mortality of neonate and 1-day-old larvae of the tobacco budworm, Heliothis virescens (F.), respectively. None of the petroleum materials caused hypoxia in 5-day-old tobacco budworm larvae, nor did undiluted cottonseed oil cause hypoxia in the boll weevil.

INTRODUCTION

Certain petroleum hydrocarbons are toxic to the beet armyworm, Spodoptera exigua (Hubner), the bollworm, Heliothis zea Boddie, (Wolfenbarger et al. 1970), mites (Chapman 1967), mosquitoes (Micks et al. 1968, Micks 1970), and aphids (Wolfenbarger 1964). Since 1970 modern technologies have made it possible to rearrange the molecular structures of these hydrocarbons. Because the rearranged structures became available we tested certain fractions as toxicants to the boll weevil, Anthonomus grandis Boheman, and the tobacco budworm, H. virescens (I.), in the laboratory. We also wished to determine whether topically applied cottonseed oil was toxic to these two important pests of cotton, and whether this oil or a petroleum hydrocarbon used as a diluent affected the toxicity of the pyrethroid insecticide permethrin.

Hypoxia, the deficiency of oxygen reaching the tissues of an organism, was induced by petroleum hydrocarbon fractions against mosquitoes (Berlin and Micks 1973). However, there is no information on whether these hydrocarbons cause hypoxia in any coleopteran or lepidopteran pest species.

For these reasons tests were conducted to determine (1) toxicity of selected petroleum hydrocarbons and cottonseed oil against adults of the boll weevil and larvae of the tobacco budworm, (2) toxicity of permethrin when diluted with a hydrocarbon and cottonseed oil against the adult boll weevil, and (3) monitor effects of the hydrocarbons and cottonseed oil on consumption of O_2 by the adult weevil and larvae of the tobacco budworm.

1/ Coleoptera: Curculionidae
2/ Lepidoptera: Noctuidae

MATERIALS AND METHODS

A paraffinic petroleum hydrocarbon, Orchex® 796 as described by Wolfenbarger et al. (1970) and Anonymous (1979), and two experimental hydrocarbons, Baytown Petroleum Research Laboratories (BPRL) 7053-1 and 5359-11, were tested. The latter two petroleum hydrocarbons are synthetic with mid-boiling points of 550 ± 50°F and 450 ± 40°F, respectively. Both of the BPRL hydrocarbons are narrow range products with a high level of branching and a spreadability function greater than that of the paraffinic spray oil. The cottonseed oil tested was filtered twice and amber in color; it contained no more than 0.1% oleic acid. Racemic permethrin was obtained in technical grade.

Boll weevils of the ebony strain and tobacco budworms had been reared in the laboratory for at least 5 yr and were considered to be susceptible to insecticides.

Toxicity of Hydrocarbons and Cottonseed Oil. All treatments were made with a microapplicator calibrated to deliver 1 µl to the dorsum of the thorax of 4- to 7-day-old weevils and 0-(neonate) to 5-day-old larvae of the tobacco budworm. Each of the petroleum hydrocarbons used in these studies against the boll weevil were diluted in acetone at 10% increments from 10% to 90% and in 1% increments from 1% to 9%. The cottonseed oil was undiluted. Three replicates (tests conducted on different days) of each dosage of hydrocarbon and oil were made against the adult boll weevil and larvae of the tobacco budworm. Mortalities were recorded 48 h posttreatment against both pest species. Mortalities wre totaled for the dosages against the boll weevil while they were averaged for the single dosage of hydrocarbon tested against the tobacco budworm.

Toxicity of Permethrin in Diluents. Topical applications were made to adult boll weevils to the dorsum of the thorax in concentrations of 0.125, 0.625, 0.031, 0.0156, 0.0078, 0.0039, 0.0019, 0.00098, 0.00049, and 0.000245 µg/µl in acetone, Orchex 796, or cottonseed oil. Mortalities were determined after 48 h. Other details of procedure were described by Wolfenbarger et al. (1970).

Analysis of Toxicity Data. Abbott's formula (Abbott 1925) was used to correct for control mortality of the tobacco budworm. Then analysis of variance (SAS Users Guide 1985) was performed on the means to determine effects to both the oil and age of the larvae by Duncan's multiple range test (SAS Users Guide 1985).

The LD_{50} values and their 95% confidence intervals were determined by log-dose mortality analysis (SAS Users Guide 1985) for hydrocarbon treatments and for the permethrin diluent mixtures against the boll weevil. Also shown is the dosage (%) of hydrocarbon that killed 90% to 100% of the adults. Nonoverlapping confidence intervals were used to indicate significant differences between the LD_{50}'s.

Hypoxia of Insects. We determined the effects of a topically-applied petroleum hydrocarbon and cottonseed oil on hypoxia which is determined by oxygen consumption. An oxygen monitor, Yellow Springs Instrument Company's Model 53, was used at 24- and 48-h posttreatment. Individual male and female boll weevils and larvae of the tobacco budworm were tested with the technique described by Guerra et al. (1983). Measurements, expressed as the respiratory quotient (QO_2) in µl of O_2 consumed/mg body wet weight/hr, were made between 8:00 and 11:30 a.m. Two treated and two untreated insects were measured simultaneously and 1 reading was taken at the end of each 15-min. test period; each 15-min. period was the test interval. Testing continued with the same insect for four 15-min periods until 50 readings were obtained per sex of adult weevils per dose of BPRL 7053-1. Fifty readings were also taken in the same manner for tobacco budworm larvae per petroleum hydrocarbon. Thus, readings were made of 13 different weevils and larvae per mean for each dose and time; 2 readings were made on the last weevil and larvae. Mean O_2 consumption $\pm$ standard deviation of the 50 readings were then determined. Analysis of variance and Duncan's multiple range tests (SAS Users Guide 1985) were determined for representative means of readings of oxygen consumption for both sexes and dosages to the weevil; the same statistical methods were performed for the larvae.

RESULTS AND DISCUSSION

Toxicity to Adult Boll Weevils. The three petroleum hydrocarbons were similar in toxicity to the boll weevil as shown by the LD_{50} values and overlapping confidence intervals (Table 1). Also, compared to the other two hydrocarbons, 10% more BPRL 7053-1 was needed to kill 90 to 100% of the weevils. This is the first report to document the toxicity of petroleum hydrocarbons to the boll weevil, and the results suggest the potential of these petroleum hydrocarbons as control agents for this insect in the future. All petroleum hydrocarbons were more toxic than cottonseed oil; 1 µl of 100% oil did not kill any adult boll weevils.

TABLE 1. Toxicity (48 h Posttreatment) of Petroleum Hydrocarbons to Adult Boll Weevils.

Treatment	No. of boll weevils	% Petroleum hydrocarbons in 1 µl of acetone	
		LD_{50} (95% confidence interval)	LD_{90}-LD_{100}
Orchex® 796	200	41 (26 to 100)	60
BPRL 5359-11	170	54 (39 to 82)	60
BPRL 7053-1	167	31 (20 to 52)	70

Toxicity of Permethrin Diluents Mixtures. The 48 h LD_{50} values and the 95% confidence intervals for permethrin diluted in acetone, cottonseed oil, and Orchex® 796, were 0.082 (0.038 - 0.22), 0.75 (0.15 - 1.06), and 0.00025 (0.00001 - 0.001) µg/weevil, respectively. When permethrin was diluted with the Orchex® 796, the toxicity was significantly different ($P \leq 0.05$) because the confidence intervals did not overlap the other confidence intervals; and the LD_{50} was 328-fold less than when permethrin was diluted in acetone. Although not significantly different permethrin was least toxic when diluted in cottonseed oil. These significant differences in the activity of topically-applied permethrin, attributable to the diluent, indicate the need for testing the combination of petroleum hydrocarbons and pyrethroid in the field.

Hypoxia Effects in Boll Weevils. When BPRL 7053-1 was topically applied at doses $\geq$ 3% of 1 µl, oxygen consumption was significantly reduced, and the reduction was ca. 50% as compared to the untreated check and the acetone check. An increase in dosage did not progressively reduce oxygen consumption (Table 2). The results show that oxygen consumption in males was significantly less than that in females at 24 and 48 h after treatment. Based on these results we suggest that reduced oxygen consumption in boll weevils was caused by petroleum oils and not by cottonseed oil. Oxygen

TABLE 2. Effect of Various Concentrations (in Acetone) of BPRL 7053-1 and Cottonseed Oil on the Oxygen Consumption of Adult Boll Weevils.

Treatment	Mean µl O_2/mg Body wt./h ± Standard Deviation At Indicated Hours Posttreatment[1/]				
	24		48		Mean
	(Male)	(Female)	(Male)	(Female)	
Check (Acetone)	11.4 ± 0.9	12.0 ± 0.8	11.2 ± 0.9	11.9 ± 1.2	11.5a
Check (Untreated)	10.8 ± 0.7	11.5 ± 0.7	9.8 ± 0.9	10.6 ± 1.1	10.7b
BPRL 7053-1 (1%)	10.5 ± 1.0	11.4 ± 0.9	10.3 ± 1.0	10.9 ± 1.2	10.7b
(2%)	10.8 ± 0.9	12.3 ± 0.6	11.6 ± 0.9	12.0 ± 1.2	11.6a
(3%)	5.1 ± 0.6	5.6 ± 0.7	4.4 ± 0.7	5.7 ± 0.8	5.0cd
(5%)	5.8 ± 0.7	6.4 ± 0.7	4.8 ± 0.7	5.6 ± 0.6	5.7c
(10%)	4.4 ± 0.5	5.4 ± 0.5	5.4 ± 0.8	6.0 ± 0.6	5.6c
(20%)	4.2 ± 0.6	5.5 ± 0.9	4.5 ± 0.8	5.4 ± 0.7	4.7d
(40%)	5.6 ± 1.0	6.1 ± 0.7	4.5 ± 0.8	6.4 ± 0.6	5.4cd
Cottonseed Oil (100%)	11.4 ± 1.1	12.9 ± 0.8	10.8 ± 1.1	12.3 ± 0.9	11.6a
Mean	8.0y	8.91x	7.73z	8.68xy	

1/ Means followed by the same letter (a, b, c, d) for the vertical mean and the same letter (x, y, z) for the horizontal mean are not significantly different at P = 0.05 (Duncan's multiple range test).

consumption by the boll weevil when treated with cottonseed oil was statistically equal to the acetone check.

Toxicity to Larvae of the Tobacco Budworm. One µl of all the three hydrocarbon materials was significantly more toxic to neonate than to 1-day-old larvae (Table 3). Larvae 2 days of age were significantly less susceptible when treated with BPRL 7053-1 and Orchex® 796 than when treated at 1 day of age. For BPRL 5359-11, there was no significant difference in the susceptibility of larvae treated when they were 1 and 2 days old. Undiluted BPRL 5359-11 was significantly more toxic to larvae from 3 to 5 days of age than were the other two hydrocarbons.

Oxygen Consumption of Tobacco Budworm Larvae. Applications of the petroleum hydrocarbons did not cause a significant reduction in the oxygen consumption of 5-day-old larvae when measured 24 or 48 hours posttreatment when compared to the untreated check (Table 4).

In conclusion, the impingement of topically-applied droplets of all three petroleum hydrocarbons were lethal to a high percentage of neonate and one-day-old tobacco budworm larvae and > 50% of the adult weevils. Field testing of the hydrocarbons alone, and in combination with other toxicants, is needed.

Reduced O_2 consumption implies a decrease in metabolic activity. The petroleum hydrocarbon tested reduced O_2 of adult boll weevils while the cottonseed oil had no affect on metabolic activity of this insect. We suggest that the reduced oxygen consumption was at least partially responsible for the toxicity of this oil if not wholly responsible.

The permethrin petroleum hydrocarbon mixture was clearly more toxic than the other two mixtures against the boll weevil; these results suggest a synergistic effect by components with completely different modes of action.

TABLE 3. Toxicity After 48 h of Three Petroleum Hydrocarbons Topically Applied to Larvae of the Tobacco Budworm of Various Ages.[1/]

Age of larvae treated (Days)	Percentage Mortality[2/]			
	Treatment			
	Check	BPRL 5359-11	BPRL 7053-1	Orchex® 796
0	4	94 ax	83 ax	86 ax
1	5	73 bx	69 bx	72 bx
2	5	60 bx	42 cy	26 cy
3	22	22 cx	4 ey	0 dy
4	5	23 cx	4 ey	4 dy
5	1	32 cx	20 dy	5 dz

1/ Sixty to 160 larvae of each age treated with each petroleum hydrocarbon; 90 held as checks for each age.
2/ Mortality corrected using Abbott's formula (Abbott 1925). Means followed by the same letter (a, b, c, d, e) for the vertical and by the same letter (x, y, z) for the horizontal are not significantly different at P = 0.05 (Duncan's multiple range test).

TABLE 4. Effect of Topical Applications of Petroleum Hydrocarbons On Oxygen Consumption of 5-Day-Old Tobacco Budworm Larvae in the Laboratory.

Treatment	Mean $\mu l\ O_2$/mg Body wt./h ± Standard Deviation At Indicated Hours Posttreatment[1/]		
	24	48	Mean
Orchex® 796	11.6 ± 0.6	12.7 ± 1.2	12.2a
BPRL 5359-11	11.2 ± 0.3	11.7 ± 1.0	11.5a
BPRL 7053-1	11.8 ± 1.4	11.0 ± 0.9	11.4a
Untreated Check	12.5 ± 0.5	12.9 ± 1.0	12.7a
Mean	11.8x	12.1x	

1/ Means followed by the same letter (a) for the vertical and the same letter (x) for the horizontal are not significantly different at P = 0.05 (Duncan's multiple range test).

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