Pakistan J. Sci. Ind. Res., Vol. 14, No. 3, June 1971

COMPARATIVE EFFECT OF PETKOLIN-M AND DDT ON THE RATE OF OXYGEN CONSUMPTION OF ADULT TRIBOLIUM CASTANEUM H.

REHANA AIJAZ, M.M.H. BAIG, NASIMA KHAN and K.A. LORD*

Department of Plant Protection, Government of Pakistan, Karachi 27

(Received September 23, 1970)

Effect of Petkolin-M and DDT on the respiration of *Tribolium castaneum* H. was compared. Both the insecticides initially increase the rate of oxygen taken up by the test insect but Petkolin-M less than DDT. The rate of respiration also increased faster with DDT. The toxicity tests indicate that petkolin-M is about 5–7 times less effective than DDT.

Petkolin-M is a new insecticide developed by PCSIR by chlorination of a petroleum fraction (mixed with an additive).[1] Nothing is known about its mode of toxic action, therefore, its effects on respiration of *Tribolium castaneum* Herbst. are examined and compared with DDT, a chlorinated insecticide, which many workers[2–6] have shown to increase the metabolism and respiration of insects.

The apparatus available for measurement of oxygen uptake was the Warburg respirometer. Because this apparatus measures changes in gas volumes and pressures in a closed space, some methods by which insecticides may be applied to insects are unsuitable for use in the apparatus, especially if early effects of the insecticides are to be observed. Thus, unless a long time is allowed for evaporation of solvents, dipping and spraying techniques are likely to introduce solvent vapours into the manometer vessels and make measurements of respiration unreliable, and also to modify the insect metabolism. Equally with small insects, topical application is time consuming so that initial measurements are delayed. Residual films were not used because a few manometer flasks were available for preliminary tests and it is difficult to produce similar deposits of insecticides in dissimilar vessels. These problems were avoided by applying the insecticides as dusts, which did not contain volatile constituents and which were capable of being applied rapidly to the insects in the manometer flasks. Also the shape of the vessels is not so critical as for residual films tests, so preliminary toxicity tests may be done in any convenient vessels.

Experimental

The test subjects were adult *T. castaneum* Herbst. reared on whole meal flour at 85–90°C and 60–70% humidity.

Preparation of Dust.—A weighed amount of DDT was ground by hand in a glass mortar and powdered soapstone† (a variety of talc), 200 BS mesh, was gradually added while the grinding continued to give 5% of insecticide in the finished dust.

Petkolin-M which is a liquid insecticide, was weighed into a beaker and absorbed on a small portion of soapstone and transferred to the mortar for grinding and the rest of the soapstone powder was added gradually.

Smaller concentrations of both insecticides were made by diluting the 5% dust with soapstone and mixing thoroughly in an end-over-end mixer.

Selection of Concentrations of Insecticides and Amounts of Dust for Tests.—Because little is known of the effect of Petkolin-M, its effects on respiration were compared with those of DDT at doses having comparable toxic effects.

Two series of experiments were done to study the effects of amounts of dust and concentrations of insecticide on toxicity. The tests were made at a constant temperature of 30°C in tablet tubes about $2.5 \times 1''$ in size. A small spatula holding about 30 powder was used to measure approximately equal mg amounts of insecticide powder.

Portions of DDT and Petkolin-M dusts were placed in tubes and groups of 15 adult beetles were added. The tubes were shaken gently to distribute the dust over the insects. Each treatment was done in triplicate. The tubes were kept in big jars which were kept in a constant temperature bath. The insects were inspected on a hot-plate[7] at constant temperature of 40–50°C to make the insects more active and facilitate the examination. The insects were examined after 2, 4 and 6 days to determine the toxic end-point. Moribund insects were counted as dead.

The effects of concentration on toxicity were observed by treating batches of 15 insects with 300 mg portions of dusts containing a range of concentrations of each insecticide—0.00625, 0.0125, 0.025, 0.05, 0.1% for DDT and 0.05, 0.1, 0.2, 0.4 and 0.8% for Petkolin-M. The control mortality was estimated using insects treated with powdered soapstone alone. The results (Table 1) were examined by the method of Probits.[8]

Doses of 15, 30, 60, 120, 300 and 600 mg of dust were applied to batches of 15 insects to determine

* From Rothamstead Experimental Station, Harpenden, U.K., on secondment to the Department of Plant Protection, Karachi 27, under U.K. Colombo Plan arrangements.

† Supplied by Industrial Grinding Ltd., Karachi, from Jumrud, Peshawar Division, N.W.F.P., Pakistan.

the effect of different amounts of dust on toxicity using three concentrations of each insecticide—0.00625, 0.0125 and 0.025% for DDT and 0.125, 0.25 and 0.5% for Petkolin-M (Table 2).

Method for Measurement of Effects of Insecticides on Respiration of T. castaneum *H.*—For each test 40 insects were removed from the culture into clean tubes ($2\frac{1}{2} \times 1$ in). The manometers were prepared by placing 0.2 ml of 10% KOH on a one-inch strip of filter paper in the central well and 300 mg insecticide-dust or soapstone powder in the main part of the flask. For untreated controls the powder was omitted. Then in quick succession the insects were placed in each flask in contact with the poison and the flasks were attached with the manometers. Readings were begun as soon as the manometers had equilibrated, 30 min after poisoning the insects. Observations were made daily for 6 hr. Each test was repeated at least three times. Fig. 1 shows oxygen uptake in each of 12 manometers during the first 5 hr after treating insects. The daily rate of oxygen uptake following four different treatments is given in Fig. 2, each point represents the average daily rate from at least 3 manometers.

TABLE 1.—TOXICITY OF DDT AND PETKOLIN-MAS. DUSTS TO *T. Castaneum* H.

Days after teratment	Heterogeneity	Regression equation	log lc 50
(i) DDT			
2	$\chi^2=(4)=2.12$	$Y=1.58x+4.54$	0.28 ± 0.08
3	$\chi^2=(4)=2.94$	$Y=1.65x+4.68$	0.19 ± 0.09
4	$\chi^2=(4)=4.99$	$Y=1.82x+83$	0.093 ± 0.01
(ii) Petkolin-M			
2	$\chi^2=(6)=4.66$	$Y=2.72\ x\pm1.16$	1.41 ± 0.011
3	$\chi^2=(6)=6.51$	$Y=2.74\ x\pm1.27$	1.36 ± 0.036
4	$\chi^2=(6)=6.16$	$Y=2.116x\pm2.197$	1.33 ± 0.04

χ^2 = Value of random variation between theory and obesrvation. X = Lograthemic values of concentration.
Y = Expected probits — value of ordinates to the provisional line. LC = Letnal concentration.

Comparison of the Toxicity of Petkolin-M and DDT.—DDT is about 5–7 times more toxic to *T. castaneum* than Petkolin-M when both are formulated as dusts with soapstone (Table 1).

Both the concentration of poison and the amount of dust used affected the mortality of *T. castaneum* but increased concentration of poison caused a greater increase in kill than did increases in the amount of dust. When the amount of dust used was doubled it had little effect on the final kill, although sometimes the insects did appear to die more rapidly when more dust was used. Within the limits of the tests (6 days) kill usually increased with the time of exposure (Table 2).

The Effect of Petkolin-M and DDT on the Rate of Oxygen Consumption of T. castaneum *H.*—Both DDT and Petkolin-M caused an increase in the rate of

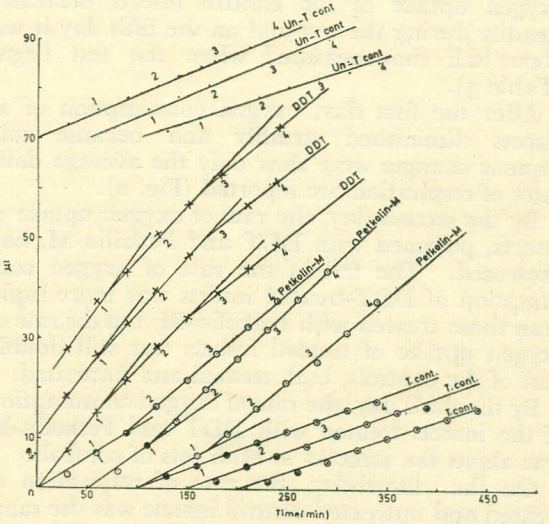

Fig. 1.—comparison of the effects of Petkolin-M and DDT on the oxygen uptake of *T. castaneum*. 1st day=24.4 68. % conc—10—LC 50; DDT—0.3%; Petkolin-M=2.3%.

TABLE 2.—EFFECTS OF AMOUNT OF DUST APPLIED AND CONCENTRATION OF DDT AND PETKOLIN-M USED.

Wt. of dust applied mg	Conc. of DDT (days after treatment)									Conc. of Petkolin-M (days after treatment)								
	0 025%			0 0125%			0.00625%			0.5%			0.25%			0.125%		
	2	4	6	2	4	6	2	4	6	2	4	6	2	4	6	2	4	6
	% kill																	
15	5.2	5.2	12.7	—	—	—	—	—	2.7	11.9	17.7	39.5	5.1	8.8	11.9	11.9	—	12.3
30	51.0	58.0	73.3	—	9.3	17.0	—	—	—	48.3	51.1	51.1	12.5	19.6	25.8	—	—	—
60	46.8	69.7	72.1	41.6	45.8	53.6	3.1	5.2	12.3	77.0	77.0	81.0	46.5	39.4	69.1	9.3	10.4	28.9
120	59.3	81.2	92.8	23.9	45.8	59.7	20.8	39.5	35.1	79.0	76.0	81.2	52.3	59.3	74.1	21.3	33.3	—
150	57.3	87.5	87.5	53.5	—	54.0	21.1	33.5	33.5	75.0	77.0	78.6	51.0	64.5	69.7	20.8	20.8	32.8
300	82.0	92.4	100	49.7	62.5	—	23.5	21.5	35.3	70.0	70.0	70.0	49.4	61.3	70.0	40.0	53.2	66.5
600	81.0	24.8	96.5	49.3	54.0	62.5	42.4	42.4	62.3	82.0	100	100	57.5	73.1	89.0	—	—	—

Untreated control % kill = 0 0
Treated control % kill = 4 0
(corrected for treated control mortality by Abbot's formula)

oxygen consumption of *T. castaneum* within 2 hr of the insects being treated by the insecticide (Fig. 1).

The effect of DDT was both greater and more rapid than that of Petkolin-M. With DDT the maximum effect was attained by the time the first measurements could be made and oxygen uptake decreased progressively. In contrast the rate of respiration of the Petkolin-M-treated insects increased during the period of measurements made on the first day. During this time DDT had increased respiration about threefold and Petkolin-M about two fold.

Treatment of insects with powdered soapstone appear to have little effect on their oxygen consumption, although throughout the test the respiration of the treated control insects was about 10% less than the untreated control insects. The oxygen uptake of the control insects decreased steadily during the test and on the fifth day it was about half that measured when the test began (Table 3).

After the first day, oxygen consumption of all insects diminished steadily and because subsequent changes were slow only the average daily rates of respiration are reported (Fig. 2).

By the second day, the rate of oxygen uptake of insects, poisoned with DDT and Petkolin-M, had decreased. The fall in the rate of oxygen consumption of DDT-treated insects was more rapid than those treated with Petkolin-M, but the rate of oxygen uptake of treated insects was still double that of the controls, both treated and untreated.

By the third day, the rate of oxygen consumption of the insects treated with DDT and Petkolin-M was about the same as in both sets of controls.

On the fourthday, the rate of respiration of treated and untreated control insects was the same as on the previous day. The rate of oxygen uptake of Petkolin-M and DDT-treated insects was still decreasing and was less than the control.

On the fifth day, the rate of respiration of insects treated with both of the insecticides was observed to be very small and eventually ceased. The control insects continued to respire.

Finally the insects were inspected visually and all the control insects were found to be alive, although they were sluggish. Between 90–100% of each batch of insects treated with DDT and 85 to 95% of the insects treated with Petkolin-M were dead.

Discussion

Both the insecticides—Petkolin-M and DDT—initially increased the rate of oxygen taken up by *T. castaneum* at 30°C but Petkolin-M less than DDT. The rate of respiration also increased faster with DDT. Treated and untreated control initially showed a steady rate of respiration followed by a gradual slowing. On the fifth and final day, the oxygen consumption of all the insects treated with both poisons ceased and a visual inspection showed that almost all the insects were dead, whereas the control insects were alive, though not very active.

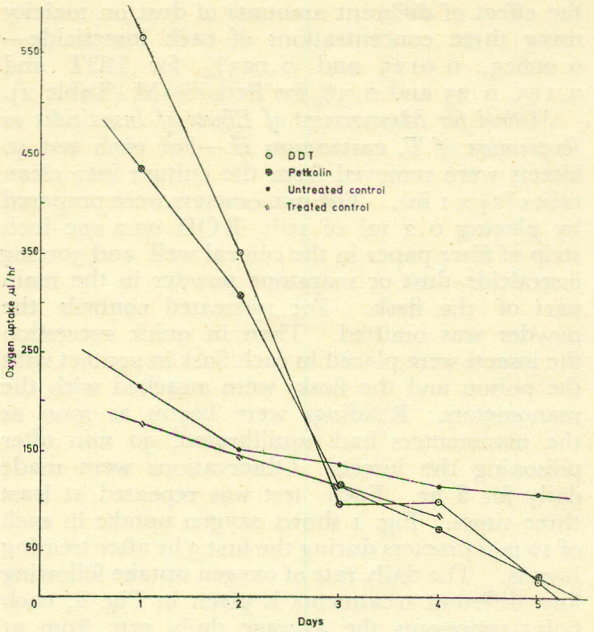

Fig. 2.—Comparison of the effects of Petkolin-M and DDT and of soapstone powder on the rate of oxygen uptake of *T. castancum* at daily intervals.

TABLE 3.—RATE OF CONSUMPTION (µ l/hr).

Day	Time (hr)	DDT	Petkolin-M	Treated control	Untreated control
1st	1	723.7	341.8	189.8	223.7
	2	588.9	431.6	194.1	220.5
	3	558.1	483.8	124.1	213.4
	4	430.0	483.8	192.7	205.6
	5	—	—	—	—
	6	—	—	—	—
2nd	1	509.4	366.4	133.3	152.5
	2	499.2	356.1	179.7	165.9
	3	385.7	332.5	139.2	149.6
	4	293.9	282.8	136.3	142.7
	5	210.9	250.9	133.4	142.6
	6	180.4	244.2	134.8	144.9
3rd	1	164.3	137.6	120.7	135.4
	2	164.0	142.1	114.9	133.2
	3	95.3	118.5	119.2	139.9
	4	83.6	106.7	119.2	126.4
	5	19.3	90.8	104.7	135.4
	6	33.4	84.9	120.9	137.9
4th	1	129.9	83.8	84.3	117.7
	2	134.3	76.1	81.4	113.2
	3	80.9	64.4	81.3	108.7
	4	73.1	58.4	88.6	108.7
	5	68.9	64.3	84.3	117.7
	6	—	—	—	—
5th	1	24.6	45.8	91.3	102.1
	2	13.1	10.5	69.5	106.0
	3	—	4.35	78.3	106.0

These results indicate that although the effects of Petkolin-M resemble those of DDT, they are smaller and Petkolin-M is about 5–7 times less toxic than DDT. However, both insecticides increased insect respiration, which follows a pattern similar to that previously reported by numerous other workers for DDT and for other chlorinated insecticides. Thus, the course of poisoning and death caused by Petkolin-M resembles other chlorinated insecticides. Unfortunately, too little is known about their mode of action to draw any definite conclusions, especially as in these tests Petkolin-M and DDT seem to take approximately the same length of time to kill, although DDT affects respiration sooner and more.

Acknowledgement.—The authors are grateful to Dr. Heshamul Huque, T.I., Plant Protection Adviser and Director of Plant Protection, for encouragement in carrying out these studies.

References

1. M. Anwarullah and T. Akhtar, Pakistan J. Sci. Ind. Res., **9,** 181 (1966).
2. D. Ludwig, Ann. Entomol. Soc. Am., **39,** 496(1946).
3. R.S. Mereill, J. Savit and M. Tobias, J. Cellular Comp. Physiol., **28,** 465(1946).
4. J.B. Buck and M.L. Keister, Biol. Bull., **97,** 64(1946).
5. K.A. Lord, Ann. Appl. Biol., **36,** 113 (1949).
6. K.A. Lord, Ann. Appl. Biol., **37,** 105 (1950).
7. F. Tattersfield and C. Potter, Ann. Appl. Biol., **30,** 2 (1943).
8. D. J. Finney, *Probit Analysis. A Statistical Treatment of the Sigmoid Response Curve* (Cambridge University Press, Cambridge, England, 1947), p. 256.