Pakistan J. Sci. Ind. Res., Vol. 16, Nos. 3-4, June–August 1973

THE RELATIONSHIP BETWEEN THE DENSITY AND MORTALITY OF FLOUR BEETLES EXPOSED TO PETKOLIN-TREATED SURFACES

S.M. MURTUZA, QAMAR H. EFFENDI and SHAHID H. ASHRAFI

PCSIR Laboratories, Karachi 39

(Received November 21, 1972; revised July 2, 1973)

Abstract. When three or more beetles per cm² were exposed to Petkolin-treated filter papers in closed petri dishes the resulting mortality was much higher than when one beetle or less per cm² was exposed. Similar results were obtained with Petkolin dust. However, when the exposures were made in uncovered dishes the density–mortality relationship did not indicate the above trend. In case of Dimecron, though the exposures were made in covered dishes, the mortality did not increase with higher densities.

The probability of critical level of oxygen need in covered dishes has been reported by many workers in such studies. The same was the case with Petkolin in the present study.

The relationship between the density of insects and the relative toxicity of insecticides against two species of flour-beetles has been reported earlier.[1] Kruse *et. al.*,[2] Schmidt and Weidhaas[3] and Wattal and Cutkomp[4] have also reported the correlation between the toxicity of DDT and different densities of mosquito larvae being tested in a given vessel.

In order to establish such a correlation the above workers have studied a number of possible factors such as the pick up of insecticides, the rate of movement of the insects, accumulation of carbon dioxide and the effect of secretions of quinone compounds by the insects besides studying the possibility of critical need of oxygen consumption. They have reported that of all the factors studied, probably the last factor appeared to have an indirect effect on the rate of mortality.

On the basis of the above results, an attempt has been made in the present study to establish a relationship, that may exist, between the number of beetles, *Tribolium castaneum* (Hbst) and the toxicity of Petkolin[5] under similar conditions and only the last factor mentioned above as a possible cause has been studied. The results have been compared with those of DDT and Dimecron.

Materials and Methods

The red flour beetles, *Tribolium castaneum* (Hbst), 3–4 weeks old were used throughout the experiment. The beetles were reared from established cultures in nutrient medium consisting of branless wheat flour+ 5% dried brewer's yeast at a temperature ranging between 25–27°C as described by Shepard.[6]

The technical grades of Petkolin (100% a.i.) and DDT (77.2% a.i.) were used for making the test dilutions in acetone whereas in case of Dimecron the emulsifiable concentrate (100% E.C.) was used. Tests were also conducted with the dusts of Petkolin (25% a.i.) and DDT (50% a.i.).

Procedure. The experiments were conducted in Pyrex petri dishes measuring 9-cm dia in which the Whatman's filter paper No. 31 of the same diameter were fitted tightly.

The stock solutions of the three insecticides were made in acetone separately to give 0.5% concentration of Petkolin and 0.1% concentrations of DDT and Dimecron. One ml of each stock solutions was pipetted out each time and spread on one filter paper already fitted in marked petri dishes. The treated filter papers were allowed to dry at the laboratory temperature for one hr. The insecticides thus spread uniformly on an area of 63.6 cm² contained approximately 0.11 mg/cm² residues of Petkolin, 0.02 mg/cm² DDT and 0.015 mg/cm² of Dimecron.

Two replications were kept for each insecticide in one set of the experiment. Each replication consisted of five dishes for exposing 50, 100, 200, 400 and 600 beetles per dish. Acetone check and untreated control each having 100 beetles were also kept with each set of the experiment.

The beetles were exposed to the treated surfaces of the filter papers for 24 hr. The observations were recorded at intervals of 72 hr and one week after transferring the insects to the food medium. During the period of exposure to the treated surfaces and during the subsequent observations the beetles were kept in dark at a temperature of 25±2°C. At the end of the exposure period, the beetles were transferred to the food medium kept in 250 ml beakers. To avoid crowding and to ensure uniform recovery conditions the beetles were reduced to batches of 100 in high density group.

The dust applications were made with the help of a modified vacuum duster adapted from the design of Farrar *et al.*[7] One half gram of dust was applied each time resulting in approximately 7.7 mg/cm² for Petkolin dust and 8.25 mg/cm² for DDT dust. The pre-and post-treatment techniques were same as in the case of technical grades of the insecticides.

For those pesticides that caused increased mortality with higher densities, in the covered dishes the rate of oxygen consumption was determined by using a Warburg's apparatus. The rate of respiration of beetles was measured in batches comprising of 50 individuals per batch. The time of respiration in each case was 40 min at a temperature of 25°C. KOH solution was used to absorb the CO_2 evolved in the process of respiration. After the measurement of the rate of

respiration, the beetles were returned to the food medium for recording the observations.

At the same time exposures were also made with Petkolin and DDT in uncovered dishes to determine the behaviour of the two insecticides when free oxygen was made available to the insects during the exposure period.

Results

The results of toxicity density relationship as expressed by per cent mortality are presented as follows:

Study with Petkolin (Technical). In the toxicity test of Petkolin to *Tribolium castaneum* (Hbst) a progressive increase in the mortality of beetles was observed with increasing densities in covered dishes. This trend was quite consistent ranging from 64% when 100 insects were exposed to 95% when 600 specimens were used in one replicate. However, when comparative tests for Petkolin were made in uncovered dishes, the trend of increased mortality with higher densities was not significant as the mortality ranged between 30–38% at the minimum and the maximum densities (Fig. 1).

Study with DDT (Technical). In case of DDT like that of Petkolin a decided increase was noted with higher densities in covered dishes. The per cent mortality ranged from 47, 69, 73 and 90% when 100, 200, 400, and 600 beetles respectively were used. When the tests with DDT were made in uncovered dishes, the range of per cent mortality values was the same with all the density groups (Fig. 2).

Study with Petkolin (Dust). Experiments with Petkolin dust were carried out both in covered and uncovered dishes. In both cases the dust was applied directly on the filter papers before releasing the insects. The results showed that the rate of mortality was on the increase with higher densities in covered dishes only whereas no differences were noted in the per cent mortality when the exposures were made in uncovered dishes.

Experiments with DDT dust were conducted in covered dishes only as it was observed that the rate of mortality at the minimum and the maximum densities was the same (Fig. 3).

Study with Dimecron. The tests with Dimecron were conducted in covered dishes only as the mortality values showed a decreasing tendency with higher density groups. In this case 35% mortality was recorded with the lowest density whereas only 24–26% mortality was noted with the greater number of beetles.

Discussion

Since the mortality values were found to increase with higher densities in covered dishes only, the rate of respiration of insects was measured to evaluate the critical need of oxygen consumption in different density groups.

In case of Petkolin it was noted that the amount of oxygen consumed was 0.05 µl/min/beetle when the density of beetles was 50. It was further observed that the rate of oxygen consumption increased significantly with higher density groups. When the rate of respiration was measured with the highest density group, it

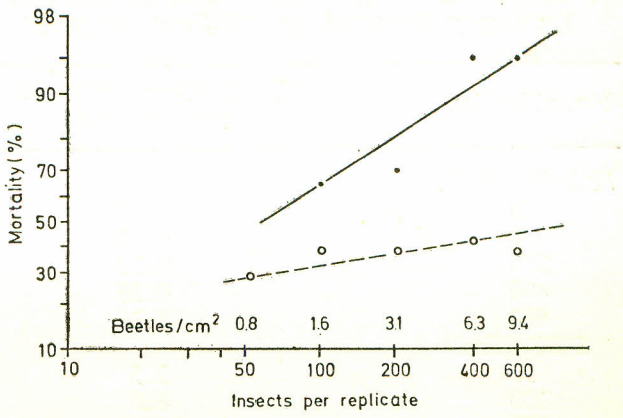

Fig. 1. Comparative mortality of flour beetles resulting from the residual application of Petkolin in covered and uncovered dishes. Petkolin: ●—● covered; o—o uncovered.

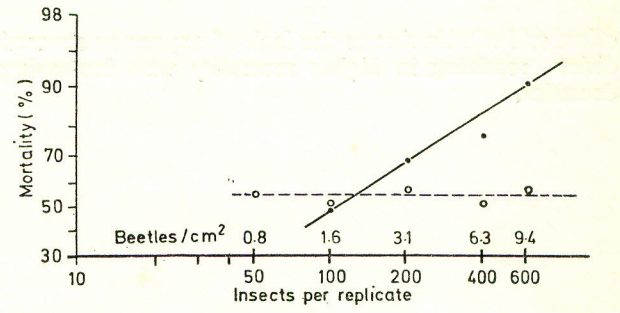

Fig. 2. Comparative mortality of flour beetles resulting from the residual application of DDT in covered and uncovered dishes. DDT: ●—● covered; o—o uncovered.

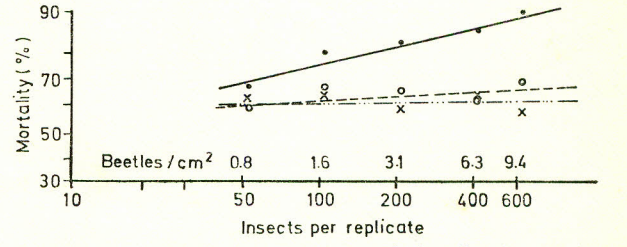

Fig. 3. Comparative mortality of flour beetles resulting from the dust application of Petkolin and DDT. ●—● Petkolin (dust 25% a.i) covered; o—o Petkolin (dust 25% a.i.) uncovered; ×—···—× (dust 50% a.i) covered.

was noted that 1.3 µl/min/beetle oxygen was being consumed by the beetles resulting in acute depletion of oxygen in the petri dish which contained approx. 15,300 µl oxygen.

Similarly in case of DDT also the rate of oxygen consumption increased progressively from the lower to the higher densities (Table 1).

The results obtained in this study for Petkolin and DDT and those reported for DDT by Wattal *et al.* showed a similar mode of action.

It was also noted that Dimecron did not cause any increase in the rate of consumption of oxygen and its mode of action was probably similar to that of a thiocyanate Lethane-384(R).

The present findings also showed that the insects were exposed to the full extent of the fumigation

TABLE 1. COMPARATIVE RATE OF RESPIRATION IN DIFFERENT DENSITIES AT 25°C FOR 40 MIN.

No. of beetles	Total oxygen consumed/ min/beetle (μl)
Petkolin (Tech).	
50	0.05
100	0.95
200	0.25
400	0.55
600	1.3
DDT (Tech)	
50	2.5
100	6.0
200	17.32
400	68.1
600	105.2

effect of Petkolin due to the lack of oxygen in covered dishes, resulting in higher mortality with increasing densities.

It was, therefore, concluded from the above results that the higher mortality of beetles with higher densities resulted due to the critical need of oxygen in covered dishes.

Acknowledgements. The authors are thankful to Mr. M. Saghir for assistance in the collection and rearing of insects.

References

1. B.L. Wattal and L. K. Cutkomp, J. Econ. Entomol., **52,** 6 (1959).
2. C.W. Kruse, G.F. Ludvik and W.B. Hawkins, J. Econ. Entomol., **45,** 598 (1952).
3. C.H. Schmidt and D.E. Weidhaas, J. Econ. Entomol., **51,** 640 (1958).
4. B.L. Wattal and L.K. Cutkomp, unpublished data (1959).
5. S. Siddiqui, S.A. Qureshi and S.H. Ashrafi, Pakistan Patent 114302 (January 14, 1964).
6. H.H. Shepard, Am. Assoc. Adv. Sci., **20,** 41(1943).
7. M.D. Farrar, W.C. O'Kane and H.W. Smith, J. Econ. Entomol., **41,** 647 (1949).