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EFFECTS OF CHEMICAL PESTICIDES ON THE GRAVID FEMALES AND STADIA OF THE WOODLOUSE, *PORCELLIO SCABER* (LATREILLE) (ISOPODA, ONISCIDEA)

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ABSTRACT

Studies were made on the mortality rates of the gravid females, and on the survival and growth rates of the stadia of the woodlouse *Porcellio scaber* (Latreille) exposed for a period of 21 days to sub-lethal concentrations of five different pesticides *viz:* abate, sumithion, pesguard (insecticides), benlate (fungicide) and gramaxon (herbicide). The survival of gravid females was severely affected when they were exposed to gramaxon or sumithion, but only marginally so following exposure to the other pesticides. Marked variation occurred in the number of broods liberated by females exposed to different pesticides. A delay in liberating the young from the brood pouch by the females exposed to pesguard was noted. High mortality of the young born to the females exposed to gramaxon or sumithion was evident. A significant difference was observed in the rate of growth of stadia born and exposed from birth to different pesticides. The ecological implications of these findings are discussed.

INTRODUCTION

The widespread use of pesticides to control pests is a recent development. Many pesticides are toxic to non-target organisms and create considerable ecological disturbances. Their impacts are usually widespread and may even reduce or exterminate the useful species as well (Brown, 1978). Since the pesticides are non-specific in their action, they have radical effects on the ecosystem and affect the soil more severely than the freshwater bodies (Edwards, 1965). Woodlice form a major group among the non-target animals living on the soil (Wallwork, 1970). These crustaceans have colonised a variety of land habitants, and are considered

primary decomposers that ingest a wide variety of dead and living plant material and feed on other animals (Paris and Sikora, 1967, Edney *et al.*, 1974, Sutton, 1980, Wieser, 1984). Except for the work of Mohamed and Nair (1989), there has been little research on the effects of chemical pesticides on woodlice. However, a good deal of information is available on the accumulation of heavy metals and on the effects of pollution from smelting and mining operations on the distribution of land and freshwater isopods (Wieser, 1961, Wieser *et al.*, 1976, Coughtrey *et al.*, 1977, Martin and Coughtrey, 1982, Hopkin and Martin, 1982, 1984, Giudice *et al.*, 1986, 1987, 1988).

The present study was undertaken to examine the effects of different commonly used pesticides in the Benghazi areas of Libya, on the survival, growth and development of the gravid females and various stadia of the woodlouse, *Porcellio scaber* (Latreille), the dominant species of terrestrial isopods in that region. Casual collections made from the field made it clear that the isopods occurred in large numbers in agricultural farmlands where humus and moisture content of the soil are moderate.

MATERIALS AND METHODS

For the present study, the gravid females of *P. scaber* containing embryos in their brood pouches were used. These females, which were collected in large numbers from Benena farms near the Benghazi airport, were maintained for 3 to 4 days in the laboratory in Petri dishes (9 cm dia) with wet filter papers at the bottom to maintain moisture levels equivalent to their natural habitats. The mean body weight ± S.E. of females was 101.5 ± 9 mg.

The sub-lethal concentrations of different pesticides used, were selected after initial trial experiments. All pesticides were initially screened for such dosages that were considered sub-lethal and would therefore, result in the marginal mortality of the woodlice during the initial exposure periods. Thus, the pesticide dosages were selected based on trial tests where the initial high mortality was not the goal. The names and the descriptions of the pesticides and the concentrations of the dosages used are:

1. Gramaxon is a herbicide with an active ingredient of 20 EC. Dosage used was 500 ppm.
2. Benlate 50 WP is a systemic fungicide. Dosage used was 2500 ppm.
3. Abate 500 EC is an organophosphate insecticide. Dosage used was 1000 ppm.
4. Sumithion — NP 50/5 Oil is a new formulation by Sumitomo Chemical Company, Japan, containing sumithion, an organophosphate insecticide and

neo-pynamin — a pyrethroid. Dosage used was 40 ppm.

5. Pesguard — NSB 5/5/15 Oil is a new compound by Sumitomo Chemical Company, Japan, consisting of two pyrethroid insecticides — neo-pynamin and sumithrin synergised by piperonyl butaxid. Dosage used was 20 ppm.

The first two compounds have been used in and around Benghazi farming systems for a decade or so, while the last three insecticides are relatively new and are now extensively used for the control of public health insects, especially mosquitoes.

The filter papers and fresh carrots (which were sliced into small pieces and given as food for the animals) were dipped in the designated doses of the different pesticides for 5 min, then dried for 10 to 15 min before the woodlice were exposed to them. To avoid mortality due to desiccation, a small cotton plug soaked in distilled water was kept in the corner of the Petri dish. Five gravid females were exposed to the treated food and filter papers in a single Petri dish and there were 6 replicates for each pesticide. Controls in which the gravid females were exposed to the untreated food and filter papers moistened with distilled water were maintained. During every 24 hours after the experiment was started, observations were made of:-

a) the mortality rates of the gravid females, b) their feeding and behavioural responses, c) the rates of liberation of the broods from the maternal brood-pouches, d) the total number of broods liberated by each female, e) the mortality and the growth (body length was measured from the front of the cephalon to the tip of the telson) rates of the different stadia.

The observations lasted for 21 days except that growth rates were studied for 28 days. The filter papers were not changed for the whole duration of the experiment, but fresh food was substituted after every 5 days. After the experimental period, the remaining females and the different stadia were maintained in normal culture media to record their recovery and survival rates.

RESULTS

1. **Mortality rates of gravid females exposed to different pesticides**

The mortality rates of the control and of the experimental gravid females of *P. scaber* are presented in Table 1. Analysis of variance of the results showed that pesticide-specific differences in mortality were highly significant ($P < 0.01$). Subsequent analysis using Dunnett's (1964) test indicated that the mortality of woodlice exposed to sumithion or to gramaxon was significantly higher than that of control, whereas such a difference was not discernible in those exposed to the

remaining pesticides. Duncan's (1955) multiple range test showed that mortality of animals exposed to abate, pesguard or benlate was lower than that of animals exposed to sumithion or gramaxon.

Table 1

P. scaber: Week-wise mortality (total number) (mean ± S.E.) of the gravid females exposed in different pesticides.

Duration of the exposure period: 21 days (3 weeks)
Total animals exposed in each pesticide: 30

Weeks	Control	Pesticides				
		Sumithion	Pesguard	Abate	Benlate	Gramaxon
1	0.57±0.30	3.00±1.41	1.14±0.40	0.43±0.30	0.57±0.37	2.43±0.81
2	0.57±0.30	0.57±0.37	0.14±0.14	0.29±0.18	0.57±0.30	0.86±0.14
3	0.14±0.14	0.00±0.00	0.29±0.18	0.29±0.28	0.00±0.00	0.29±0.19
Mean	0.43±0.15	1.19±0.94	0.52±0.31	0.33±0.25	0.38±0.28	1.19±0.59

Regressions showing pesticide-specific mortality of *P. scaber* over time are presented in Fig. 1. The resulting equations, correlation coefficients and level of significance are shown in Table 2. The results showed that the mortality rates of

Table 2

P. scaber: Linear regression equations of pesticide-specific mortality on time

Pesticides	Intercept a	Regression coefficient b	Correlation coefficient r	P-values
Control	1.17	0.33	0.94	< 0.005
Sumithion	15.94	0.58	0.74	+4 0.005
Pesguard	4.34	0.37	0.94	< 0.005
Abate	0.13	0.36	0.95	< 0.005
Benlate	2.25	0.35	0.90	< 0.005
Gramaxon	2.90	1.31*	0.91	< 0.005

*Significantly different ($P < 0.01$) from others

these isopods exposed to the control or to the different pesticides over time were significant ($P < 0.005$). The covariance technique in regression analysis (Johnston, 1970) showed that the linear regression equations of pesticide-specific mortality on time and their corresponding intercepts were significantly different ($P < 0.01$).

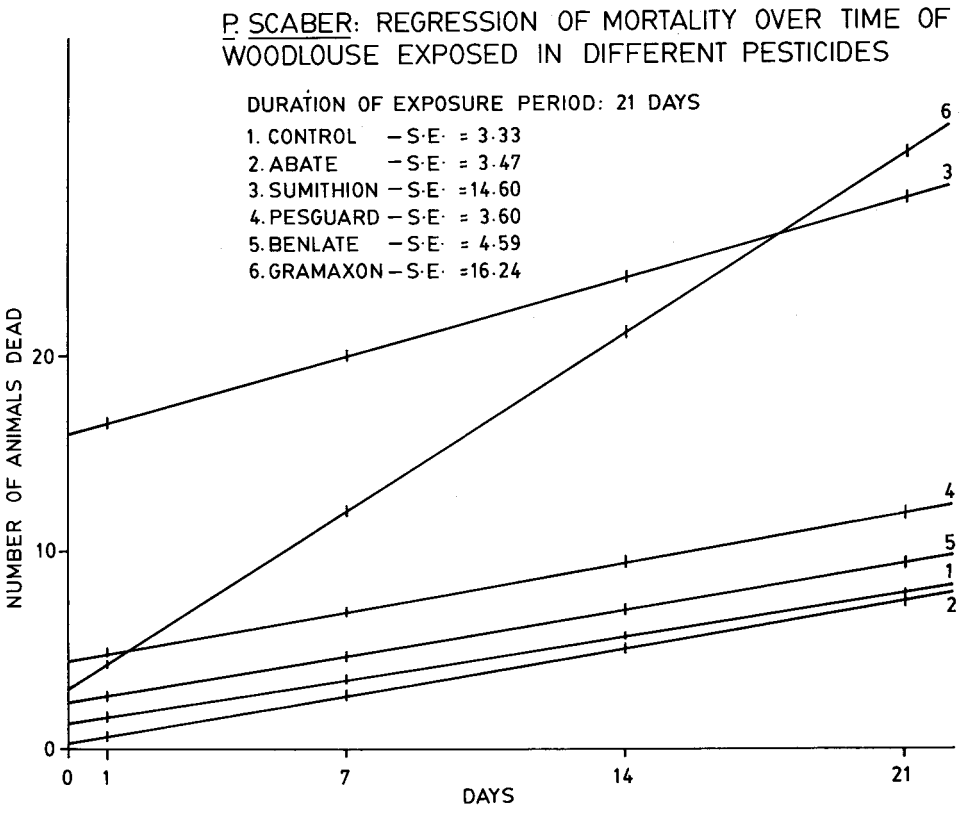

Fig. 1: *P. scaber:* Relationship of pesticide-specific mortality to time in a 21-day exposure. 1 = control, 2 = abate, 3 = sumithion, 4 = pesguard, 5 = benlate, 6 = gramaxon. Vertical bars are meant as visual aids for estimating mortality at weekly intervals.

2. Effects of pesticides on the liberation of the broods and on the total broods liberated by the females

The weekly average number of gravid females that liberated the broods is presented in Table 3 and the average number of broods liberated per female is presented in Table 4. In all, 117 gravid females liberated 1712 broods. Most of the females, except those exposed to pesguard, liberated their broods during the first

week of the observational period. The highest number of females liberating the broods did so after exposure to benlate, while the lowest did so after exposure to sumithion. The lowest broods liberated per female also came from females exposed to sumithion. However, none of these differences were found to be statistically significant following analysis by Anova and Ancova.

3. Growth of Stadia

At hatching, all individuals showed the same body length (1.8 mm) (Table 5). Later on, differences in the average growth of body lengths of control and experimental animals became apparent. Dunnett's test was used to compare the rates of growth, where $d_{0.05} = 0.08$. From this test, the different pesticides tried and the control could be grouped into three categories based on their intensity of inhibition on the rates of growth of these animals. Thus, gramaxon was grouped under category I,

Table 3

P. scaber: Weekly average number of gravid females (Mean ± S.E.) which liberated broods

Duration of observational period: 21 days
Total animals exposed in each pesticide: 30

Weeks	Control	Pesticides				
		Sumithion	Pesguard	Abate	Benlate	Gramaxon
1	1.71±0.61	0.57±0.30	0.14±0.14	1.57±0.37	3.14±1.39	2.86±1.16
2	0.71±0.36	0.43±0.30	0.43±0.30	0.86±0.55	0.71±0.56	0.00±0.00
3	0.57±0.37	0.00±0.00	1.71±0.99	1.29±0.84	0.00±0.00	0.00±0.00
Mean	1.00±0.48	0.33±0.14	0.76±0.36	1.24±0.60	1.29±0.56	0.95±0.82

Table 4

P. scaber: Weekly average number of broods (Mean ± S.E.) liberated per female.

Duration of observational period: 21 days
Total animals exposed in each pesticide: 30

Weeks	Control	Pesticides				
		Sumithion	Pesguard	Abate	Benlate	Gramaxon
1	15.6±0.5	8.0±0.9	15.0±0.0	17.1±0.7	13.1±0.5	14.7±2.5
2	18.0±1.2	14.6±3.9	11.7±2.7	7.8±1.1	20.6±1.9	0.0±0.00
3	16.7±1.1	0.0±0.0	13.8±0.5	17.8±0.2	0.0±0.0	0.0±0.0
Mean	16.1±0.6	10.8±2.2	13.5±0.7	15.2±0.9	14.5±0.8	14.7±2.5

where the growth of the young was severely affected when exposed to it, category II included abate, sumithion and pesguard where the growth of the young was moderately affected and category III included control and benlate where the growth rates were unaffected.

The linear regression analysis confirmed that the differences in growth over time was highly significant (P 0.01) (Fig. 2). The 't' values for the six regression lines were 17.49 (control), 4.83 (abate), 12.25 (sumithion), 10.97 (pesguard), 17.00 (benlate), 7.00 (gramaxon). When compared with the control, the mean growth rates of juveniles exposed to gramaaxon were significantly low, whereas the mean growth rate was higher for those exposed to benlate (Table 6).

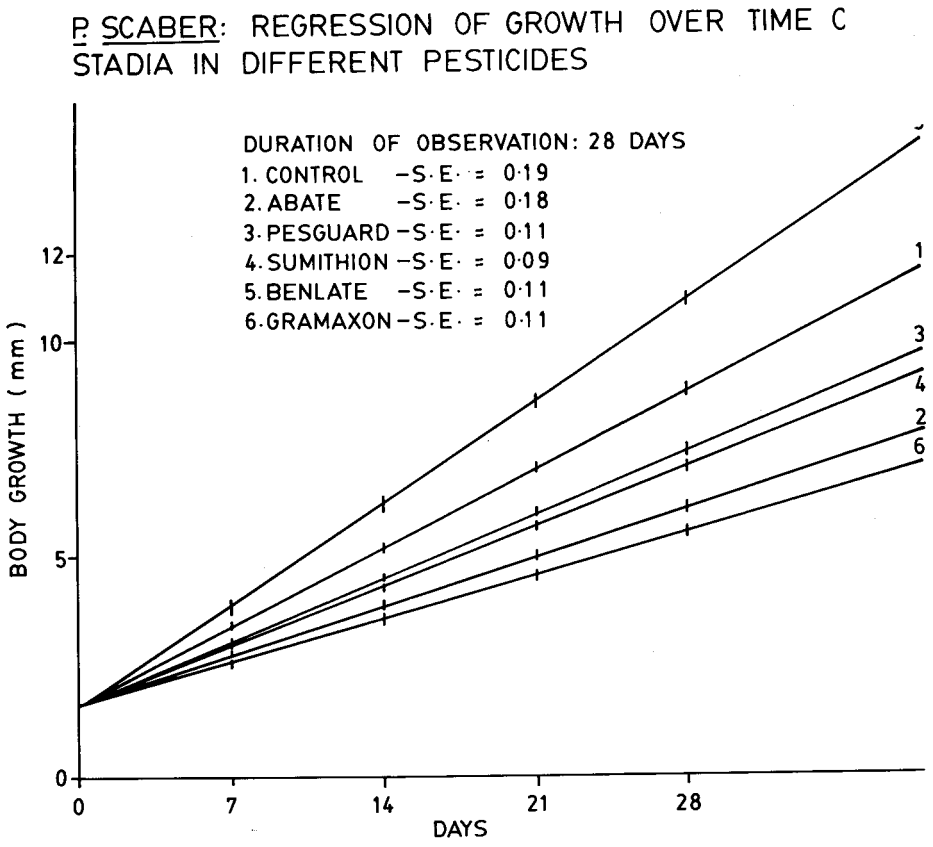

Fig. 2: *P. scaber:* Relationship of pesticide-specific growth of stadia to time in a 28-day exposure. 1. Control S.E. = 0.19, 2. Abate S.E. = 0.18, 3. Pesguard S.E. = 0.11, 4. Sumithion S.E. = 0.09, 5. Benlate S.E. = 0.11, 6. Gramaxon S.E. = 0.11Vertical bars are meant as visual aids for estimating growth at weekly intervals.

Table 5

P. scaber: The growth (mean body length + S.D.) of stadia
Duration of observation: 28 days
Total animals exposed in each medium: 60

Days	Control	Pesticides				
		Sumithion	Pesguard	Abate	Benlate	Gramaxon
1	1.8±0.1	1.8±0.2	1.8±0.1	1.8±0.2	1.8±0.2	1.8±0.2
7	2.3±0.1	2.0±0.1	2.1±0.2	2.2±0.2	2.1±0.1	2.0±0.1
14	2.5±0.2	2.3±0.1	2.3±0.1	2.2±0.1	2.5±0.2	2.2±0.1
21	2.7±0.1	2.4±0.2	2.4±0.1	2.4±0.2	2.9±0.2	2.2±0.2
28	2.9±0.1	2.6±0.1	2.7±0.2	2.5±0.1	3.1±0.2	2.4±0.2
Mean	2.4±0.4	2.2±0.3	2.3±0.3	2.2±0.3	2.5±0.5	2.1±0.3

Table 6

P. scaber: Linear regression equations of pesticide-specific growth on time for different stadia

Pesticides	Intercept a	Regression coefficient b	Correlation coefficient r	P-values
Control	1.66	0.26	0.97	< 0.005
Sumithion	1.62	0.20	0.99	< 0.001
Pesguard	1.63	0.21	0.99	< 0.001
Abate	1.74	0.16	0.94	< 0.01
Benlate	1.46	0.34	0.99	< 0.001
Gramaxon	1.70	0.14	0.97	< 0.005

4. **Behavioural responses**

The gravid females of control *P. scaber* showed slight tendencies of cannibalism after 12 days of exposure. The filter paper and the food were partly affected by fungal growth.

A tendency for aggression within 24 hours of exposure was shown by the animals kept in the abate-treated medium. The young stadia which were hatched from the maternal brood pouches were inactive for some days. Cannibalism was also observed among the adults after 8 to 9 days of exposure, and scraping the filter papers with the thoracic appendage was a common phenomenon.

P. scaber exposed to sumithion were mostly inactive during the initial 5 to 6 days of exposure to it. Mortality of the adults and the young was very high throughout. Cannibalistic tendencies were also observed after 13 days of exposure. One gravid female could not shed its anterior moult and in this condition it remained alive for 6 days before it died.

Animals exposed to pesguard were also affected by showing symtptoms of slow movements. Tendency for aggression and high cannibalism were observed among the adults. The antennae of 2 females turned white in colour after 12 days of exposure in it. A delay of about 7 to 10 days in the hatching of the young was observed.

Almost normal behaviour was shown by the animals exposed to benlate. The tendency to liberate broods by the females was high during the initial exposure period in this medium.

Females kept in the gramaxon-treated medium showed normal behaviour during the early exposure periods in it, but later the mortality of the adults and the newly hatched young began to increase rapidly. The food and the filter papers were partly affected by fungal growth.

The physical reactions of the gravid females and of the juvenile stadia to all the pesticides were more or less similar. The initial symptoms of the toxic effect was evident by the significant decrease in their feeding rates followed by erratic movements. Later, tendencies for aggregation and occasional cannibalism were observed. Following that, they remained motionless with their bodies upside down with occasional feeble movements of their walking appendages and pleopods. Death occurred soon after this stage.

DISCUSSION

The toxic effects of five different pesticides on the gravid females and on the stadia of *P. scaber* showed wide variations, with the herbicide gramaxon and insecticide sumithion producing higher mortality in these animals than the fungicide benlate and insecticides abate and pesguard. Mohamed and Nair (1989) also reported high mortality of the woodlice *Procellionides pruinosus* (Brandt) and *Porcellio laevis* (Latreille) exposed to leaves treated with gramaxon whereas *Armadillo officinalis*

Dumeril, the pillbug seen commonly in the Benghazi area, showed more tolerance to this herbicide. The effect of herbicides on the soil fauna mainly result from alterations in ground cover that they induce (Fox, 1964). Of the several herbicides tested on soil fauna, some produced reduction in numbers of predatory mites, hemiedaphic collembola and to a lesser extent of detritivorous mites, earthworms and insect larvae (Edwards, 1965, Curry, 1970). Thus herbicides in general seem to have adverse effects on the survival rates of many soil invertebrates.

The toxic effect of sumithion, a newly introduced organophosphate insecticide used in the present study, was much faster with a rapid knock-down during the initial exposure period; the recovery rates were also minimal. This insecticide also interfered with moulting to the extent that some gravid females died in the process. The other insecticides tried, abate and pesguard, were comparatively less harmful as far as mortality of the gravid females and stadia were concerned, but there was a considerable delay in the emergence of the young from the maternal brood pouches in those exposed to pesguard. The fungicide benlate, however, proved to be the least toxic to the animals and almost normal behaviour of the females and stadia were discernible.

The pesticides directly affected stadial growth in different ways. Thus, juveniles exposed from birth to the herbicide gramaxon showed strong inhibitions whereas those exposed to the fungicide benlate showed slight stimulations in their growth rates, as compared with control juveniles. The other pesticides had only marginally adverse effects on the growth rates of the stadia.

Our results indicate that the various chemical pesticides sprayed on the farms of Benghazi area in large quantities affect adversely the non-target oranisms living in the soil such as woodlice. The higher reates of mortality occurring in these organisms as a result of pesticide spraying results in an imbalance in the soil ecosystem which in turn affects the process of humification and aeration of the soil by the activities of these organisms in due course of time (Edwards, 1965, Brown, 1978). Accordingly, a well planned screening programme on the optimum dosage of the different pesticides to be used is vitally needed for a better conservation of these beneficial organisms.

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تأثير المبيدات الكيميائية على الإناث الحاملة وصغار قمل الخشب *Porcellio scaber*

انشوتان نايـر و عبد الله ابراهيم محمد و ك.س. بويان

لقد استهدفت الدراسة تقيم أثر المبيدات الكيميائية الزراعية على لافقاريات التربة النافعة وذلك من خلال بحث أستخدم فيه الإناث الحاملة وصغار قملة الخشب *porcellio scaber* كحيوان للتجربة و ٥ مبيدات كيميائية وهي الأبيت والسوموثيون والبيسجارد كمبيدات حشرية والبنسليت كمبيد الفطريات والجرامكسون كمبيد الأعشاب .

وقد دلت النتائج على أن الجرامكسون والسومثيون لهما تأثيرات بالغة على معيشة الإناث ، بينما كانت المبيدات الثلاثة الباقية طفيفة التأثير .

كما دلت النتائج أيضاً على اختلافات متباينة في أعداد المواليد الناتجة من الإناث المعرضة للمبيدات المختلفة ، بالإضافة إلى ذلك فإن هناك تأثير في التولد للإناث المعرضة لمركب البيسجارد .

ودلت النتائج أيضاً على ارتفاع في معدل موت الصغار المتولدة من الإناث المعرضة لكل من الجرامكسون والسوميثيون .

كما أفادت التحاليل الإحصائية على وجود اختلافات في معدل النمو للصغار المتولدة من الإناث عند إستخدام المبيدات المختلفة .