Appl. Ent. Zool. 11 (2): 94-99 (1976)

Antifeeding Activity of Chlordimeform for Plant-Sucking Insects

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(Received March 8, 1976)

The antifeeding effect of chlordimeform was tested on five different species of plantsucking insects using radioactive ³²P-H₃PO₄. Chlordimeform exhibited a non-specific antifeeding action, and the sucking rates of all the insect species tested were reduced to 40 to 60% at the concentration as low as 10 ppm.

INTRODUCTION

If chemical inhibition of insect feeding is valid, then it is easily conceivable that such chemicals can be substitute for varietal resistance, and/or augment its utility value of the agricultural crop, and play one of the important role as a desirable partner in the integrated control program for crop pests.

It was shown recently (Fujiwara, 1971) that chlordimeform, N'-(4-chloro-o-tolyl)-N, N-dimethylformanidine (Fig. 1-1), so far as developed as an acaricide, has antifeeding activity for the phytophagous insects. The present investigation was carried out to determine the effect of chlordimeform on feeding of the five species of plant-sucking insects.

MATERIALS AND METHODS

Insects: Female adults of the following five species were used.

Delphacidae

Nilaparvata lugens, the brown planthopper

Laodelphax striatellus, the smaller brown planthopper

Deltocephalidae

Nephotettix cincticeps, the green rice leafhopper

Aphididae

Myzus persicae, the green peach aphid

Lipaphis pseudobrassicae, the turnip aphid

N. lugens and L. striatellus were collected from the stock culture maintained on rice seedlings at 25°C, and 16 hr photoperiod in the laboratory. N. cincticeps was supplied from the Aburabi Laboratory, Shionogi & Co., Ltd. The two species of

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aphids were collected from the natural colony on the potted cabbage in the green-house.

Test solution: Chlordimeform hydrochloride was dissolved in 20% sucrose solution, contained 1% aspartic acid, at a series of concentrations ranging from 0.1 to 1000 ppm. Sucrose solution of 5% was prepared in the case of N. cincticeps. Radioactive $^{32}\text{P-H}_3\text{PO}_4$ was added to thus prepared chlordimeform hydrochloride solution at 100 to 500 cpm/ $10~\mu l$ for the respective plant- and leaf hopper test and about 4000 cpm/ $10~\mu l$ for the aphid. Sucrose solution without chlordimeform hydrochloride was used as a control.

Sucking cage: Experiments were conducted by means of two different-sized artificial sucking cages made of glass rings. One was 20 mm in inner diameter with 15-mm height, and the other was 25 mm in inner diameter with 20-mm height, respectively. The top of each cage was closed with tetoron gause remaining a small hole at the center, through which the insects were introduced into it. The bottom opening was covered with a parafilm sachet containing 0.1 to 0.2 ml of the test solution.

Bioassay: Ten planthoppers or five leafhoppers and ten aphids were confined in each sucking cage of 25×20 mm and smaller cage of 20×15 mm, respectively. These sucking cages were kept at 27°C with 100% RH under the continuous lighting. N. cincticeps, N. lugesns—L. striatellus, L. pseudobrassicae, and M. persicae were allowed to suck the test solution freely for 3, 6, 9, and 24 hr, respectively. The amount of fluid intake was determined by measuring the radioactivity both of the insect body and the excrement. For this purpose, the test insects fed the radioactive solutions were oxidized with concentrated nitric acid by heating under an infra-red lamp, and the honeydews excreted were recovered by washing the surface of parafilm membrane of the cage with a small amount of distilled water. The washed-out honeydews were dried up under the infra-red lamp after adding a drop of KOH alcoholic solu-The radioactivities in the dried up residues of the oxidized insect bodies and in the honeydews were measured by means of Aloka SC-1C automatic 2π thin window gas flow G.M. counter or Aloka TDC-1 G.M. counter. A preliminary test showed 80% recovery of 32P-H3PO4 deposited on the parafilm membrane. Therefore, an estimation of the total amount of fluid imbibition during the given period is possible by the following formula:

$$X=10 (A + B / 0.8) / C$$

where X=total amount of imbibition (μl), A=cpm in the insect body, B=cpm in the honeydew recovered, and C=cpm in 10 μl of each test solution.

The insect mortalities were also recorded at the end of the test period. In another experiments, mortality trends of *N. lugens* and *L. striatellus* were compared between the insects sucked the test solution containing 100 ppm chlordimeform and those given no dietary solution.

RESULTS

The amount of fluid intake by the said five different species of plant-sucking insects could be quantitatively estimated by measuring the radioactivity of the insect body and the honeydew excreted after the test insect fed on the test solution incorporated with a small quantity of the radioactive ³²P-H₃PO₄.

Table 1. Sucking-Inhibitory Effect of Chlordimeform for N. lugens

Concentration (ppm)	Radioactivity, cpm/cage		Total	Rate of sucking	Insect
	Insect body	Honeydew	$\begin{array}{c} {\rm imbibition} \\ (\mu l/{\rm cage}) \end{array}$	inhibition (%)	mortality (%)
0	177	19	3.9	0	4
0.1	152	23	3.5	10	4
1	138	12	3.0	23	22
10	116	9	2.5	36	16
0	171	21	5.8	0	18
10	112	10	3.7	36	40
100	27	3	0.9	84	60
1000	3	0	0.1	98	93

Note: Radioactivities in the dietary solutions were 499 (upper) and 331 cpm/10 μl (low), respectively.

Table 2. Sucking-Inhibitory Effect of Chlordimeform for L. striatellus

Concentration	Radioactivity, cpm/cage		Total imbibition	Rate of sucking	Insect	
(ppm)	Insect body	Honeydew	$(\mu l/\text{cage})$	inhibition (%)	mortality (%)	
0	78	14	3.2	0	2	
0.1	119	22	4.9	-53	4	
1	91	13	3.6	-13	2	
10	35	3	1.3	59	22	
100	25	4	1.0	69	30	
1000	7	0	0.2	94	52	

Note: Radioactivity in the dietary solution was 289 cpm/10 μl .

Table 3. Sucking-Inhibitory Effect of Chlordimeform for N. cincticeps

Concentration	Radioactivity, cpm/cage		Total imbibition	Rate of sucking inhibition	Insect
(ppm)	Insect body	Honeydew	$(\mu l/\text{cage})$	(%)	mortality (%)
0	31	77	8.8	0	0
0.1	34	90	10.1	-15	0
1	26	64	7.3	17	0
10	19	35	3.7	58	0
100	11	15	2.1	76	0
1000	5	7	1.0	89	8

Note: Radioactivity in the dietary solution was 123 cpm/10 μl .

It was found out that the leafhopper, N. cincticeps, excreted much more radiotracer than that detected from the insect body. On the other hand, the radioactivity in the insect body was apparently higher than that in the honeydew in the cases of both species of planthoppers, N. lugens and L. striatellus. In the case of an aphid, L. pseudobrassicae, most of the radiotracer was accumulated in the insect body.

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Table 4. Sucking-Inhibitory Effect of Chlordimeform for L. pseudobrassicae

Concentration	Radioactivity, cpm/cage		Total	Rate of sucking	Insect	
(ppm)	Insect body	Honeydew	$rac{\mathrm{imbibition}}{(\mu l/\mathrm{cage})}$	$\begin{array}{c} \text{inhibition} \\ (\%) \end{array}$	mortality (%)	
0	64	9	0.18	0	0	
0.1	59	6	0.16	11	0	
1	54	7	0.15	17	0	
10	28	6	0.08	56	0	
100	5	6	0.03	83	0	
1000	3	6	0.02	89	0	

Note: Radioactivity in the dietary solution was 4100 cpm/10 μl .

Table 5. Sucking-Inhibitory Effect of Chlordimeform for M. persicae

Concentration (ppm)	Radioactivity in insect body (cpm/cage)	Total imbibition ^a $(\mu l/\text{cage})$	Rate of sucking inhibition (%)	Insect mortality (%)
0	73	0.20	0	0
0.1	33	0.09	55	0
1	35	0.09	55	10
10	4	0.01	85	3
100	5	0.01	85	4

Note: Radioactivity in the dietary solution was $3700 \text{ cpm}/10 \mu l$.

Table 6. Comparative Mortality Trend of N. lugens and L. striatellus between Starvation and Feeding on the Dietary Solution Contained 100 ppm Chlordimeform

Species	Treatment	Mortalities (%) at the indicated period				periods
		2	4	6	8	10hr
N. lugens	Starvation	0	3	73	90	97
	Chlordimeform	0	3	50	80	97
		3	6	9	18	24hr
L. striatellus	Starvation	0	0	10	70	90
	Chlordimeform	0	0	7	50	90

Note: Thirty female adults were used in each treatment.

As were recognized from Tables 1 to 5, chlordimeform suppressed effectively the rate of imbibition of all the insect species tested. At the concentration of 10 ppm, the amount of imbibition was reduced by 36% in N. lugens, and 55% or more in the case of the others. The sucking-inhibitory activity of chlordimeform increased as the concentration increase, and 70 to 85% and 90% inhibition were recorded at 100 ppm and 1000 ppm, respectively. Sucking by L. striatellus and N. cincticeps was tended to enhance in some extent by chlordimeform below 1 ppm.

The mortalities of N. lugens and L. striatellus increased with increase of the chlordi-

a Based on the radioactivity in insect body only.

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meform concentration, although few or no mortality was observed in the rest during the test period. However, the mortality of the both species of planthoppers on the dietary solution contained 100 ppm chlordimeform showed a similar trend to those induced by a starvation (Table 6).

DISCUSSION

Fujiwara (1971) has reported that chlordimeform exhibited antifeeding activity of the phytophagous insect, especially in the case of larvae of lepidopterous species, such as Pryeria sinica and Calopilos miranda. NAKAYAMA et al. (1972) have found out that chlordimeform inhibited the feeding and consequently disturbed the aggregation of newly hatched larvae of the tabacco cutworm, Spodoptera litura, as low concentration as 5 ppm. Similarly, Hirao et al. (1972) and Koyama (1975) have demonstrated that the newly hatched larvae of the rice stem borer, Chilo suppressalis, were prevented to bore into rice stems treated with 5 ppm chlordimeform. These evidences seemingly indicate a practical significance that chlordimeform has induced an artificial insect resistance in appropriate crops. However, such antifeeding property of chlordimeform against the plant-sucking insects have been remained obscure. In the present experiments it was demonstrated that chlordimeform reduced effectively the sucking rates of the five different species of Homoptera at the concentrations above 10 ppm. Relatively higher mortalities were recorded in N. lugens and L. striatellus being fed on the dietary solutions contained higher concentration of chlordimeform, but the mortalities were considered to be induced by starvation, as well, as a result of the strong antifeeding effect of this chemical. The obtained result was confirmed, as quite similar mortality trends were shown when the both species of planthoppers were kept in starvation and on the dietary solution contained 100 ppm chlordimeform. Fujiwara (1971), and Nagata and Moriya (1975) have reported that the number of eggs laid on the rice seedlings dipped in the chlordimeform solution were reduced in the case of L. striatellus and N. lugens. It seems possible that these phenomena are attributed to the antifeeding activity of chlordimeform. Although the mode of antifeeding action of this chemical is not known yet at present, it is of particular interest that the chemical structure of chlordimeform is very similar to that of the aromatic amines which have previously demonstrated to act as a potential antifeeding chemical for the plant- and leafhoppers (Sōgawa, 1971; Kurata and Sōgawa, 1976) (Fig. 1).

Fig. 1. Similarlity of chemical structures of chlordimeform (1), tyramine (2) and hordenine (3).

ACKNOWLEDGEMENT

The authors wish to express their sincere thanks to Prof. T. Saito, Faculty of Agriculture, Nagoya University, and Dr. M. Yoshimeki, Tropical Agriculture Research Center, for their critical readings of this manuscript, and to Mr. H. Honda for his assistance in rearing the test insects.

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