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Toxicity of Nicotinyl Insecticides on the Haemocytes of Red Cotton Bug, *Dysdercus koenigii* (Fb.) (Pyrrhocoridae: Hemiptera)

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ABSTRACT

The toxicity of acetamiprid 20% SL and imidacloprid 25WP on adult *Dysdercus koenigii* Fb. was studied. Five types of haemocytes i.e, plasmatocytes, granulocytes, prohaemocytes, oenocytoids, spherulocytes were identified as 35, 23, 18. 25, 7.5 and 3.5% after treatment with acetamiprid, and 28.25, 24.50, 32.75, 2.25 and 1.25% after treatment with imidacloprid, respectively. After the application of acetamiprid, the total haemocyte count (THC) increased (19865 cells/mm^3) soon after the application of insecticide, while decreased (11010 cells/mm^3) after half an hour and increased considerably again (22855 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3). In case of imidacloprid, THC decreased (14488 cells/mm^3) soon after the application of insecticide, while it increased (16163 cells/mm^3) after half an hour and increased again (21983 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3).

Key Words: Toxicity; Nicotinyl insecticides; Haemocytes; *Dysdercus koenigii;* Acetamiprid; Imidacloprid

INTRODUCTION

Cotton (*Gossypium hirsutum* L) is known as lifeblood and white gold for the economy of Pakistan. It contributes a major part in our foreign exchange, which is up to 68% (Khan & Khan, 1995) and shares about 62.3% in the total exports (Anonymous, 2003).

Chemical control is the most commonly used method against insect pests of cotton in Pakistan. These insecticides greatly affect the different systems of the insects. In this sense the haematological studies are very important in the field of insect physiology because certain vital functions are performed by the haemocytes as they are responsible for supplying nutrients and oxygen to the various tissues and are phagocytic in action (Richards & Davies, 1977). Some haemocytes are engaged to direct the function of food storage and connective tissue formation (Wigglesworth, 1956).

Insects have only extra-cellular fluid, haemolymph, in contrast to vertebrates, which have two such fluids, blood and lymph. As the only extra-cellular fluid, it is perhaps not surprising that the haemolymph, in general, serves the functions of both blood and lymph of vertebrates. Thus, the fluid fraction (plasma) is the transport system for nutrition, hormones and metabolic wastes and contains elements of the immune system while the cellular components are haemocytes (Gillot, 1995). The number of the cells present in the circulating blood of various adult species varied from 1000 per cubic mm to about 100 times of this number (Tauber & Yeager, 1936). The number of various types of haemocytes under various conditions and different time intervals provide useful information about the resistance and virulence of various species against different insecticides. The percentage of phagocytes in differential counting of haemocytes may provide useful information about the immunity of insects against insecticides. The knowledge of effects of insecticides on the insect haemocytes can be useful in determining the control recommendations against any species.

The aim of the present study was to examine the effect of acetamiprid 20% SL and imidacloprid 25 WP, on the total haemocyte count, differential haemocyte count, and to observe the abnormalities caused by these insecticides in the haemocytes of red cotton bug (*Dysdercus koeningii* Fb.)

MATERIALS AND METHODS

The adults of red cotton bug were collected from cotton field located at University of Agriculture Faisalabad and brought to the laboratory for the present research.

Nicotinyl insecticides, acetamiprid 20% SL and imidacloprid 25WP, were applied with 0.125 and 0.25% concentrations, respectively on the thorax, with the help of micro-applicator at the rate of 8µl per application and after each application of insecticide, a new tip was replaced. Effect of insecticide was noted just after application, after 30 and 60 min and in terms of abnormalities found in the haemocytes and the changes in the total haemocyte count. Experiment was repeated four times.

RESULTS

Total haemocyte count in the untreated specimen of red cotton bug (*Dysdercus koenigii* Fb.). The results obtained from the present study (Table I) indicate that on an average there are 17000 cells/mm^3 in the haemolymph of untreated adult of *Dysdercus koenigii* Fb.

Differential haemocyte count in the untreated specimen

of red cotton bug (*Dysdercus koenigii* Fb.). The differential haemocyte count was done with the object to find out the percentage of various classes of haemocytes in the haemolymph of adult of red cotton bug Dysdercus koenigii Fb. under normal conditions. The data (Table I) reveal that the percentage of plasmatocytes was the highest (39.75%) followed by granulocytes (32%), prohaemocytes (22%), oenocytoids (4.25%) and spherulocytes (2%).

Effect of acetamiprid 20% SL on the total haemocyte count (THC). The total haemocyte count (THC) increased (19865 cells/mm^3) soon after the application of insecticide, while decreased (11010 cells/mm^3) after half an hour and increased considerably again (22855 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3) as shown in Table II.

Effect of acetamiprid 20% SL on the differential haemocyte count (DHC). The percentages of plasmatocytes, granulocytes, prohaemocytes decreased from normal by 39.75, 32, and 22% to 35, 23, and 18.25%; while the percentages of oenocytoids and spherulocytes increased from normal by 4.25 and 2% to 7.5 and 3.5%, respectively after the application of acetamiprid 20% SL as shown in Table II.

Effect of imidacloprid 25 WP on the total haemocyte count (THC). The data given in Table III reveals that the total haemocyte count (THC) decreased (14488 cells/mm^3) soon after the application of insecticide, while it increased (16163 cells/mm^3) after half an hour and increased again (21983 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3).

Effect of imidacloprid 25 WP on the differential haemocyte count (DHC). The data (Table III) shows that percentages of plasmatocytes, granulocytes, oenocytoids, spherulocytes decreased from normal by 39.75, 32, 4.25 and 2% to 28.25, 24.50, 2.25 and 1.25%, respectively; whereas, the percentage of prohaemocyte increased from normal by 22 to 32.75%.

Table I. Total and differential haemocyte count in the untreated adult of *Dysdercus koenigii* Fb.

No. of observations	Total no. of haemocytes/mm^3	Differential haemocyte count PL %	GR%	PR%	OE%	SP%
1	17375	43	37	27	5	1
2	18785	37	33	20	8	4
3	16740	41	36	26	3	2
4	15100	38	22	15	1	1
Average	17000	39.75	32	22	4.25	2

Table II. Effect of acetamiprid 20% SL on the total and differential haemocyte count of adult of *Dysdercus koenigii* Fb.

No. of observations	Total no. of haemocytes Just after application	After 30 minutes	After 60 minutes	Differential haemocyte count PL%	GR%	PR%	OE%	SP%
1	19660	10550	22860	40	24	17	9	3
2	18375	13423	24375	37	27	20	6	4
3	21530	8297	22305	39	22	26	10	2
4	19900	11770	21880	24	19	10	5	5
Average	19865	11010	22855	35	23	18.25	7.5	3.5

Table III. Effect of imidacloprid 25 WP on the total and differential haemocyte count of adult of *Dysdercus koenigii* Fb.

No. of observations	Total no. of haemocytes Just after application	After 30 minutes	After 60 minutes	Differential haemocyte count PL%	GR%	PR%	OE%	SP%
1	16000	13555	22322	33	22	40	3	3
2	12353	17070	24175	30	29	27	1	2
3	15275	16517	21780	28	27	29	3	0
4	14324	17510	19655	22	20	35	2	0
Average	14488	16163	21983	28.25	24.50	32.75	2.25	1.25

DISCUSSION

Effect of acetamiprid 20% SL on the total and differential haemocyte count. The data (Table II) revealed that the total haemocyte count (THC) increased (19865 cells/mm^3) soon after the application of insecticide, while decreased (11010 cells/mm^3) after half an hour and increased considerably again (22855 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3).

These findings are in conformity with those of Mahmood and Yousaf (1985), Khan (1994), who applied Dipterex, Curacron 500 EC, on *Gryllus bimaculatus* de Geer, *Leucinodes orbonalis* Guen, respectively and observed that the total haemocyte count increased just after application of these insecticides.

Regarding differential haemocyte count (DHC), the data given in Table II indicates that the percentages of plasmatocytes, granulocytes, prohaemocytes decreased from normal by 39.75, 32, and 22% to 35, 23, and 18.25% while the percentages of oenocytoids and spherulocytes increased from normal by 4.25 and 2% to 7.5 and 3.5%, respectively after the application of acetamiprid 20% SL.

These results are in contradiction with those of Khan (1994) and Alhariri (2000), who studied the effect of Curacron 500 EC on *Leucinodes orbonalis* Guen, Lorsban 10 Ec and Basudin 60 EC on *Schistocera gregaria* Forsk, and noted that the percentage of plasmatocytes increased.

Effect of imidacloprid 25 WP on the total and differential haemocyte count. The data given in Table III revealed that the total haemocyte count (THC) decreased (14488 cells/mm^3) soon after the application of insecticide, while it increased (16163 cells/mm^3) after half an hour and increased again (21983 cells/mm^3) after one hour of application from the normal count (17000 cells/mm^3).

These findings differ from those of Hassan (1985) and Alhariri (2000) who studied the effect of carbamates, pyrethroids, Decis 2.5 EC, Karate 2.5 EC, Lorsban 40 EC and Basudin 60 EC, respectively on haemocyte count *Tryporyza* sp., and *Schistocera gregaria* Forsk., respectively

and noted that the total haemocyte count increased just after application of insecticides.

Regarding DHC, the data shown in Table III revealed that percentages of plasmatocytes, granulocytes, oenocytoids, spherulocytes decreased from normal by 39.75, 32, 4.25, and 2% to 28.25, 24.50, 2.25, and 1.25%, respectively; whereas, the percentage of prohaemocyte increased from normal 22 to 32.75%.

These results are in partial accordance with those of Ambrose and George (1996), Ayub (1996) who studied the effect of Quinalphos and endosulfan, Fyfanon 50 EC and Perfekthion 40 EC and Methyl parathion 50 EC on the differential haemocyte count of *Acanthaspis pedestris* Stal., and *Drosicha stebbingi* Green., respectively and observed that the percentage of prohaemocyte increased from normal.

Abnormalities caused to haemocytes. The following abnormalities were recorded in the haemocytes of red cotton bug after the application of acetamiprid 20% SL, imidacloprid 25 WP: enlargement of haemocytes, distortion of the shape of haemocytes, rupturing of wall of haemocytes, denucleation of cells, abnormal stainging of the cells, displacement of nuclei on one side of the cell. The present results are in conformity with the findings of previous workers (Hassan, 1985; Mahmood & Yousaf, 1985; Ali, 1986).

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