



Insecticide induced alterations in the mid gut of Red Cotton Stainer, *Dysdercus koenigii* (Fabricius) (Hemiptera: Pyrrhocoridae) treated with Deltamethrin

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Abstract

Dysdercus koenigii, the Red Cotton Stainer is a well-known destructive pest of cotton and also act as mechanical vector in transmitting fungus, *Nematospora gossypii* that cause internal boll disease of cotton. Deltamethrin, in the concentrations 0.0001% and 0.0005% were provided in feed of adult red cotton bugs to observe alteration in the mid gut after 24 hours. Mid gut is lined with enteric epithelium which rest upon a basement membrane and is covered by inner circular and outer longitudinal muscles. Inner cavity is filled with lumen. Histopathological studies on mid gut resulted in degradation of basement membrane and degeneration in the epithelial lining in both concentrations (0.0001% and 0.0005%). Higher dose caused more disruption in longitudinal and circular muscles as compared to lower dose. In lower concentration, slight constrictions were observed in lumen while at higher concentration, shrinkage became more pronounced.

Keywords: *Dysdercus koenigii*, deltamethrin, mid gut, histopathology, necrosis

1. Introduction

Dysdercus koenigii also known as the Red Cotton Bug belongs to the family Pyrrhocoridae of the order Hemiptera. It is a polyphagous insect which has a wide range of hosts belonging to family Malvaceae and Bombaceae [1]. It is a well-known destructive pest of cotton in India, and many parts of the world [2]. Cotton is a premier cash crop that influences the economic development of many countries including India. India is the largest producer of cotton in the world accounting for about 26% of the world cotton production. It has the distinction of having the largest area under cotton cultivation in the world ranging between 10.9 million hectares to 12.8 million hectares and constituting about 38% to 41% of the world area under cotton cultivation [3]. The nymphs and adults of *D. koenigii* suck up the juice from the seed of either green or ripe cotton bolls. Both are voracious feeders and cause extensive damage to the crop. The damaged bolls remain shrivelled and vitality of plant and the quality of the lint is also severely affected in case of heavy infestation. *D. koenigii* is also responsible for the transmission of internal boll disease of cotton, caused by a fungus, *Nematospora gossypii* and related fungi, which are probably transmitted from contaminated boll to uncontaminated boll mechanically through mouth parts of the bug [4]. The advent of pest problem leads to the development of chemical control method. Insecticides are currently the key to insect-pests management in almost all cropping systems around the world [5]. Deltamethrin is an insecticide belonging to the pyrethroid family. Pyrethroids are the man-made versions of pyrethrins, natural insecticides from *Chrysanthemum* flowers. Pyrethroids mode of action is to modify the gating kinetics of voltage-sensitive sodium channels. It keeps the sodium channels open for a prolonged period of time resulting in hyper excitation. They inhibit sodium channel deactivation and causing uncontrolled nerve firing. Deltamethrin poisoning occurs through cuticular penetration or oral uptake. It has a

half-life of 5.9-17 days on plant surfaces [6]. It controls numerous insect pests of field crops. A wide variety of household pests, especially spiders, fleas, ticks, carpenter ants, carpenter bees, cockroaches and bed bugs can be prevented and eliminated with the help of deltamethrin. Treatment with various doses of insecticides has damaged the alimentary canal, in *Spodoptera litura* treated with organophosphate insecticides [7], in *Oxya nitidula* treated with monocrotophos [8], in *Blattella germanica* treated with boric acid [9], in *Mythimna separata* treated with fraxinellone [10], in *Periplaneta americana* treated with *Datura alba* leaf extract [11], in *P. americana* treated with N-nitroso-N-methylurea [12], in red palm weevil *Rhyncophorus ferrugineus* treated with the zinc sulphate [13], in *P. americana* treated with deltamethrin and chlorpyrifos [14, 15] and in the midgut of oriental latrine fly, *Chrysomya megacephala* treated with deltamethrin [16]. Present study deals with histopathological effect of insecticide Deltamethrin on the midgut of red cotton stainer, *Dysdercus koenigii*.

2. Materials and Methods

2.1 Breeding and Maintenance of *Dysdercus koenigii*

Red Cotton Stainers (*Dysdercus koenigii*) were collected during September-October, 2017 from the okra field located at near the Aligarh Muslim University campus, Aligarh and cultured at room temperature.

2.2 Application of Insecticide

Cotton bugs were kept in the jars containing 2 cm thick layer of sterilized sandy soil at the bottom of the jar. This soil layer was kept moist to provide the suitable environment for cotton bugs. Cotton bugs were fed with cotton seeds soaked with 2 ml of each insecticide concentrations (0.0001% and 0.0005%) in each petri dish which were kept in jars 1 and 2 separately and 10 cotton bugs were released in each jar. Finally, glass jars were

covered with muslin cloth. In addition to this, one controlled setup was also run parallelly. The cotton bugs were then examined and dissected in insect Ringer's solution after 24 hours to obtain mid gut for further studies.

2.3 Preservation and Histological Preparation

The alimentary canal was dissected out from cotton bugs and was immediately fixed in Bouin's solution for 18-20 hours. After removing the tissue from the fixative, they are washed in tap water and distilled water and then after this dehydration proceeded in ascending grades of alcohol i.e. 30%, 50%, 70%, 80%, 90%, 96% and 100% followed by mixture of 100% and xylene solution (1:1), then incubating the tissue in xylene and paraffin (1:1) at 60° C for 15 minutes and subsequent incubation of tissue in pure wax for 2 hours. Mid gut was embedded in paraffin wax. After this, 5 µm sections were cut using microtome from each prepared block. The ribbons were then placed on the glass slide lubricated with a solution of albumin and glycerine (1:1). The slides containing section were warmed slightly to straighten the creases.

The slides were processed in xylene and then descending grades of alcohol, then in distilled water for 5 minutes. Slides were stained in Delafield's haematoxylin and counter stained with Eosin followed by upgrade dehydration of alcohol and then 2 changes of xylene for 5 minutes each. After air drying, slides were mounted in DPX to observe under the compound light microscope. Photographs were taken using Nikon Eclipse compound microscope by using appropriate magnification.

3. Results and Discussion

3.1 Normal histology of the Mid gut of *D. koenigii* (Figs.1-3)

The alimentary canal of *D. koenigii* is a long, muscular and tubular structure extending from mouth to anus. It is differentiated into three regions viz., Foregut (stomodaeum), Mid gut (mesenteron) and Hindgut (proctodaeum) with sphincters controlling food movements between regions.

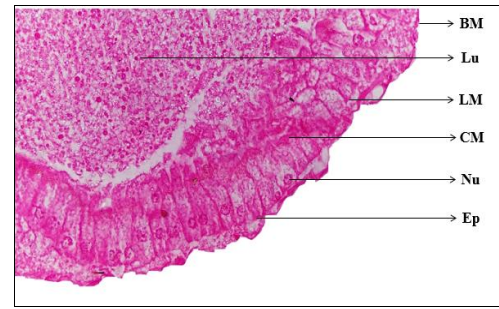


Fig 3: T.S. of control Midgut of *D. koenigii* (400X)

Foregut or stomodaeum is ectodermal in origin. Internal cuticular lining is present. Terminal mouth parts leads into a preoral cavity. Behind the mouth a well muscled organ called pharynx is present which pushes the food into oesophagus. Pharynx act as a sucking pump in sap feeders. Oesophagus is a narrow tube which conducts food into crop. Transfer of the food from foregut to midgut is regulated through Cardiac valve or Oesophageal valve.

Mid gut or mesenteron is endodermal in origin. This part contains no cuticular lining. It is further divided into anterior, middle and posterior part. Anterior mid gut arises from junction of gastric caeca. A muscular sphincter is present at posterior part of mesenteron which prevents entry of undigested food and uric acid from hindgut into mid gut. Internally, the mid gut is lined by the stratum of enteric epithelium and cells rest upon a basement membrane, the latter is followed by an inner layer circular muscles and an outer layer of longitudinal muscles. Epithelium possesses a striated border and these cells have well defined nucleus.

Hindgut or proctodaeum is ectodermal in origin. Internal cuticular lining is present. It is differentiated into three regions viz., ileum, colon and rectum. Malpighian tubules are present at the junction of mesenteron and ileum. Ileum is short and narrow tube and colon is highly coiled. Rectum opens out through anus.

3.2 Effect of 0.0001% and 0.0005% concentrations of deltamethrin on midgut of *D. koenigii* (Figs. 4-7)

Histological changes in the mid gut treated with 0.0001% and 0.0005% concentration of deltamethrin were examined. Present study deals with basic histology of the mid gut which has been distorted after the application of 2 concentrations of insecticide. At lower concentration, it depicts slight changes in the histology of the mid gut while at higher concentration, the damage became more prominent.

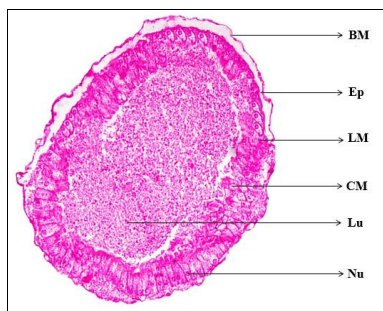


Fig 1: T.S. of control Midgut of *D. koenigii* (100X)

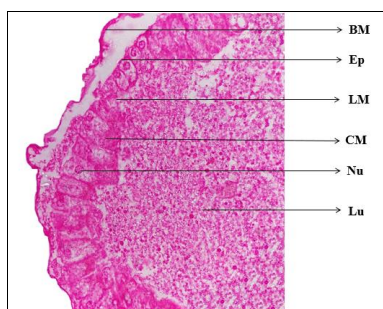


Fig 2: T.S. of control Midgut of *D. koenigii* (400X)

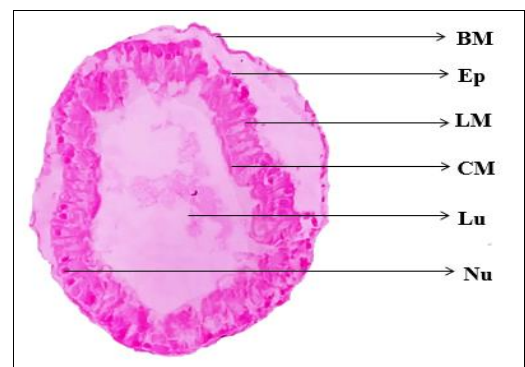


Fig 4: T.S. of Midgut of *D. koenigii* treated with 0.0001% Deltamethrin (100X)

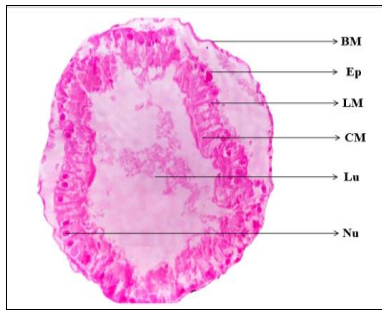


Fig 5: T.S. of Midgut of *D. koenigii* treated with 0.0001% Deltamethrin (400X)

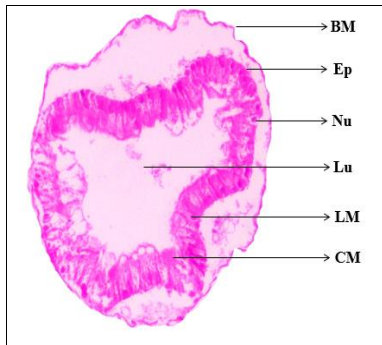


Fig 6: T.S. of Midgut of *D. koenigii* treated with 0.0005% Deltamethrin (100X)

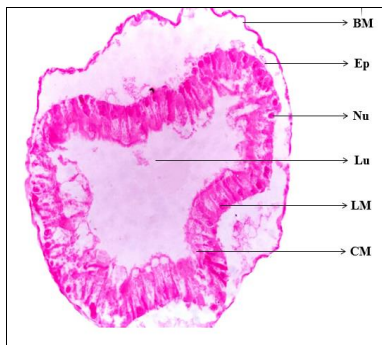


Fig 7: T.S. of Midgut of *D. koenigii* treated with 0.0005% Deltamethrin (400X)

Marked changes in the conformation of the epithelium were observed at both the concentrations of insecticide. Degeneration, distortion, thinning and number of folding were observed in the epithelium. At 0.0001% concentration of insecticide, detachment of the epithelium with only few folds were visible after application while at 0.0005% concentration, thinning in the epithelium was initiated, disruption increased and distortion became more prominent with further increase in the folding's of epithelium which showed more damage in the epithelium in comparison to the lower dose. The cells were faintly visible at both the concentration of insecticide. Necroses in the cells were quite distinctly evident in the present findings at different concentrations. Similar histopathological changes were reported in *Schistocerca gregaria* treated with various synthetic insecticides by Mukherji & Haridas [17], in *Periplaneta americana* by Sutherland [18] and Bearwlad & Boush [19], in *Spodoptera litura* treated with organophosphate insecticides by Lal *et al.* [7], in *Oxya nitidula* treated with monocrotophos by Amutha *et al.* [8], in *Blattella germanica* treated with boric acid by Habes *et al.* [9], in *Mythimna separata* treated with fraxinellone by Lu *et*

al. [10], in *P. americana* treated with *Datura alba* leaf extract by Khan *et al.* [11], in *P. americana* treated with N-nitroso-N-methylurea by Jain & Ahi [12] and in *P. americana* treated with deltamethrin and chlorpyrifos by Majumdar *et al.* [14, 15].

Degradation of the basement membrane is prominently observed at both the concentrations. Similar observations have been demonstrated in *B. germanica* treated with boric acid by Habes *et al.* [9], in red palm weevil *Rhynchophorus ferrugineus* treated with the zinc sulphate by Al-Dhafar & Sharaby [13], in *P. americana* treated with *D. alba* leaf extract by Khan *et al.* [11] in *P. americana* treated with N-nitroso-N-methylurea by Jain & Ahi [12] and in the midgut of oriental latrine fly, *Chrysomya megacephala* treated with deltamethrin by Yasmeen & Amir [16].

After application of both the concentrations of insecticide, disorganisation and disintegration of the epithelial cells were observed. Degenerated cytoplasm along with distorted circular and longitudinal muscles, as well as disappearance of cell boundaries of the epithelial cells of the midgut became clearly evident. Similar findings were also made by Mukherji & Haridas [17] in *S. gregaria* treated with various synthetic insecticides, by Sutherland [18] and Bearwlad & Boush [19] in *P. americana*, by Lal *et al.* [7] in *S. litura* treated with organophosphate insecticides, by Amutha *et al.* [8] in *O. nitidula* treated with monocrotophos, by Habes *et al.* [9] in *B. germanica* treated with boric acid, by Lu *et al.* [10] on the midgut of *M. separata* treated with fraxinellone, by Khan *et al.* [11] on the midgut of *P. americana* treated with *D. alba* leaf extract, by Jain and Ahi [12] on the midgut of *P. americana* treated with N-nitroso-N-methylurea and by Majumdar *et al.* [14, 15] on the mid gut of *P. americana* treated with deltamethrin and chlorpyrifos.

At lower concentration, decrease in the content of the lumen was observed while at higher concentration lumen has clustered towards the centre of the mid gut. Similar results have been demonstrated in *B. germanica* treated with boric acid by Habes *et al.* [9], in red palm weevil *R. ferrugineus* treated with the zinc sulphate by Al-Dhafar & Sharaby [13], in *P. americana* treated with *D. alba* leaf extract by Khan *et al.* [11], in *P. americana* treated with N-nitroso-N-methylurea by Jain & Ahi [12] and by Yasmeen & Amir [16] on the mid gut of oriental latrine fly, *C. megacephala* treated with deltamethrin.

In conclusion, deltamethrin caused cellular deformation in the mid gut which is dose dependent and time dependent. So, the present study elucidates significant damage in the mid gut accounts for the positive signs towards management in the population of *D. koenigii*.

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