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Histopathological effect of deltamethrin on the midgut of American cockroach, *Periplaneta americana* (Linn.) (Dictyoptera: Blattidae)

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Abstract

Periplaneta americana, the American cockroach is a major nuisance and also act as mechanical vector in transmitting bacteria that cause diarrheal diseases, nosocomial infections and allergic symptoms. Deltamethrin, in the concentrations 0.005% and 0.01% were provided in feed of adult cockroaches to observe histopathological changes in midgut after 24 hours. Midgut is lined with enteric epithelium which rest upon a basement membrane and is covered by inner circular and outer longitudinal muscles. Midgut contains a thin peritrophic membrane as innermost layer. Histopathological studies on midgut resulted in degradation of basement membrane and degeneration in epithelial lining in both concentration (0.01% and 0.005%). Higher dose caused more disruption in longitudinal and circular muscles as compared to low dose. Peritrophic membrane was detached from epithelial lining and is degenerated to great extent in high dose. In lower concentration slight constriction is observed in lumen while at higher concentration shrinkage is more pronounced.

Keywords: *Periplaneta americana*, deltamethrin, midgut, histopathology

Introduction

Periplaneta americana are insects of the order Blattoidea. *P. americana*, also known as the American cockroach, is an urban cockroach found in places such as homes and shops [1]. Cockroaches are generally omnivorous; and feed on a great variety of foodstuffs. They also serve as potential carriers of the causes of bacterial diarrhoea and nosocomial infections in hospitals [2, 3]. Cockroaches, especially the large American cockroach (*P. americana*) and the smaller German cockroach (*Blattella germanica*) are a major nuisance in a large number of Indian homes. Cockroaches can also passively transport microbes on their body surfaces including pathogens that are potentially dangerous to humans. The main source of nourishment for cockroaches in mines and sewers is human faeces, which can be 80% bacterial. German cockroaches have been observed feeding on mouth secretions of corpses riddled with lung disease; these secretions were almost 100% infectious bacteria. There is ample evidence that substances produced by cockroaches are involved in producing allergic symptoms [4, 5]. Besides that, cockroaches also secrete a mixture of Xanthurenic acid, kiturenic acid and 8-hydroxyquinoline, which are tryptophan derivatives that have mutagenic and carcinogenic properties [6]. Cockroaches, especially species that come in contact with faeces like German cockroaches may transmit bacteria responsible for food poisoning. Cockroaches act as mechanical vector in transmitting *Salmonella*, *Shigella* and *Cryptosporidium parvum* bacteria that cause diarrheal diseases. Antibiotic resistant strain of *Klebsiella pneumoniae* bacteria that cause pneumonia has been found in patients and cockroaches in hospitals. In addition, evidence suggests that cockroaches spread typhoid, dysentery and leprosy organisms. Cockroach allergy was first reported in 1943, when it was noted that certain patients developed skin rashes immediately after the insects crawled over their skin [7]. Cockroach allergy is an important health problem associated with the development of asthma, as a consequence of chronic exposure to low levels of allergens in susceptible individuals. At least eleven proteins isolated from German and American cockroaches can cause allergic reactions and contribute to asthma in humans. The allergens are heat-stable and persistent in the environment even after the insect death. Cockroach allergen sensitization is one of the greatest risk factors for increased asthma-related poor health among the low-income population in the United States

(Referred to as the inner-city asthma problem in the US). Deltamethrin is a pyrethroid ester insecticide. This material is a member of one of the safest classes of pesticides, synthetic pyrethroids. It is helpful in eliminating and preventing a wide variety of household pests, especially spiders, fleas, ticks, carpenter ants, carpenter bees, cockroaches and bed bugs. Deltamethrin is also one of the primary ingredients in ant chalk. Treatment with various doses of insecticides has damaged the alimentary canal, in *Plebiogryllus guttiventris* treated with Fenitrothion [8], in *Spodoptera litura* treated with organophosphorus insecticides [9], in *Periplaneta americana* treated with N-Nitroso-N-Methylurea [10], in *Oxya nitidula* treated with monocrotophos [11], in *Periplaneta americana* treated with Cypermethrin, carbaryl and monocrotophos [12], in *Schistocerca gregaria* treated with various synthetic insecticides [13], in *Chrotogonus trachypterus* treated with some insecticides [14]. Present study deals with histopathological effect of insecticide Deltamethrin on the midgut of cockroach *P. americana*.

Material and Method

Cockroaches (*Periplaneta americana*) were collected during September-November, 2015 from sewer manhole in and around the campus of Aligarh Muslim University, Aligarh and cultured at room temperature.

Preparation of insecticidal concentrates

0.1% stock solution of deltamethrin was prepared in distilled water. Then this concentration was diluted 10 times to get desired concentration of 0.01%. After that, 0.005% concentration was prepared by mixing 2.5 ml of 0.01% with 2.5 ml of distilled water.

Application of insecticide

Bread crumbs were mixed with 3ml of each insecticide concentrations (0.005% and 0.01%) in each petri dish which were kept in jars 1 and 2 separately and 5 cockroaches were released in each jar. Parallel to these control set up was also maintained. Cockroaches were then dissected in insect Ringer's solution after 24 hours to obtain midgut for further studies.

Preservation and histological preparation

Alimentary canal was excised from junction of midgut and hindgut containing Malpighian tubule and fixed immediately in Bouin's solution for 24 hours. After 24 hours, washing was done 2-3 times in tap water to remove excess picric acid and dehydration proceeded in ascending grades of alcohol i.e. 30%, 50%, 70%, 80%, 90% for 5 minutes each while in 96% and 100% for half an hour each followed by mixture off 100% and xylene solution (1:1) for 10 minutes. Incubation was done in 60°C in xylene and paraffin was (1:1) for 15 minutes and then in pure wax for 2 hours.

The gut section was embedded in paraffin wax. 5 micrometer microtome sections were cut from the prepared block. Then the ribbons were placed in glass slide which was lubricated by albumin solution having 5 drops glycerin. Slides were then stretched on warming table to remove creases.

Slides were processed in 2 changes of xylene, then descending grades of alcohol series 100%, 96%, 90%, 80%, 70%, 50%, 30% for 5 minutes each and in distilled water for 5 minutes each. Slides were stained in hematoxylin for 10 seconds, then washed in tap water and counter stained with eosin for 25 minutes followed by upgrade dehydration of alcohol for 5 minutes each and then 2 changes of xylene for 10 minutes.

After air drying slides were mounted using D.P.X to observe under compound microscope. Photographs were taken using LEICA compound microscope using appropriate magnification.

Results and Discussion

Normal histology of the midgut of *P. americana* (Figs. 1&2)

The gut of *P. americana* consists of foregut (stomodaeum), midgut (mesenteron) and hindgut (proctodaeum) with sphincters controlling food movement between regions. Foregut or stomodaeum includes pharynx, oesophagus, crop and gizzard. Mouth leads into muscular tube called pharynx. Pharynx leads into a narrow and short tube known as oesophagus. Oesophagus opens into crop which is a sac like structure. Gizzard is highly muscular and bulb like structure. Stomodaeal valve is present at junction of gizzard and mesenteron. Hepatic caecae are finger shaped structure present at the junction of gizzard and mesenteron.

Midgut is further divided into anterior, middle and posterior part. Anterior midgut 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 midgut.

Proctodaeum includes 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.

Internally the midgut 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. Midgut contains peritrophic membrane within its central lumen and is present as a thin transparent membrane.

Effect of 0.005% and 0.01% concentrations of deltamethrin on midgut of *P. americana* (Figs. 3-6)

At 0.01% concentration deltamethrin cause progressive destructive changes in the midgut, whereas, in treated cockroaches fed on 0.005% deltamethrin shows slight change in histology of midgut. Degeneration and distortion in epithelial lining was observed in both concentrations. Degraded basement membrane and decrease in midgut lumen was observed in both concentrations. Circular and longitudinal muscles get more disrupted at higher concentration (0.01%) than at low concentration. Striated border was affected to some extent at 0.005% concentration while at 0.01% concentration it is almost lost. Conformation of epithelium has changed. Peritrophic membrane was detached from epithelial lining and get constricted at low concentration (0.005%) and highly degenerated and clustered at centre of gut cavity at high concentration (0.01%).

In present study, after application of insecticide disorganization and disintegration of epithelial cells was observed. The gut epithelium was seen with degenerated cytoplasm along with distorted circular and longitudinal muscle shrinkage, disappearance of cell boundaries of the epithelial cells of midgut were observed in *P. americana*. Similar histopathological changes were reported in *P. americana* [15-19], *Spodoptera litura* treated with endosulfan, diazinon and dichlorvos [9], and in *Plebiogryllus guttiventris* treated with fenitrothion [8].

Present study reveals marked disruption of peritrophic membrane and striated border and same histopathological

changes was observed in the midgut of *Culex pipiens* larvae and *P. americana* where oil extract of chamomile plant produced destruction of the peritrophic membrane [20, 21, 10]. In

conclusion, results obtained from this investigation showed that deltamethrin caused cellular deformation in the midgut.

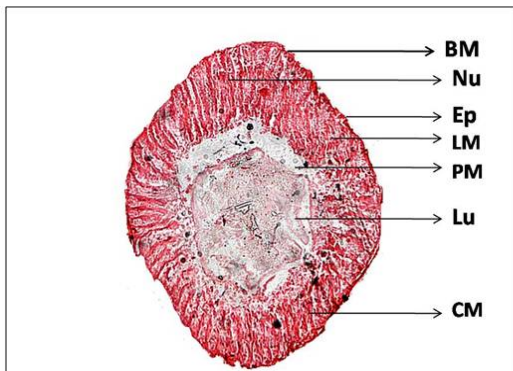


Fig. 1: T.S. of midgut of *P. americana*: Control (4X)

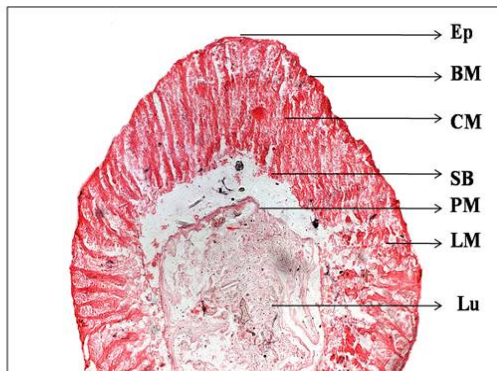


Fig. 2: T.S. of midgut of *P. americana*: Control (10X)

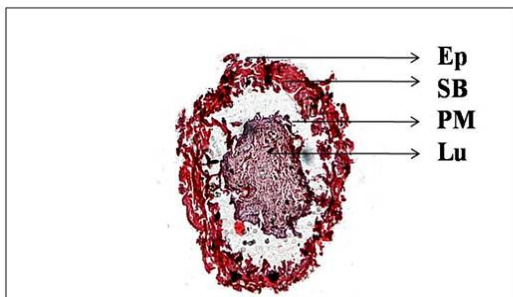


Fig. 3: T. S. of midgut of *P. americana* treated with 0.005% deltamethrin (4X)

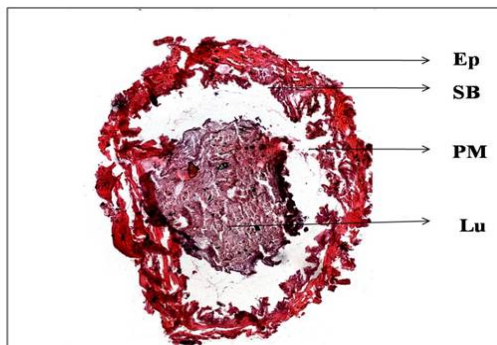


Fig. 4: T. S. of midgut of *P. americana* treated with 0.005% deltamethrin (10X)

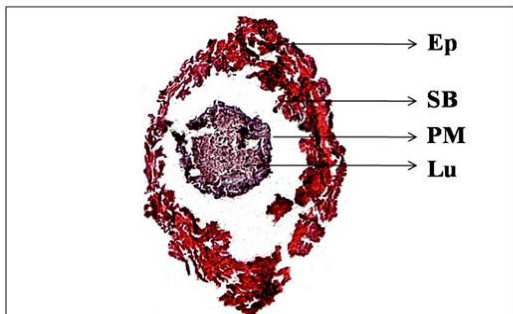


Fig. 5: T. S. of midgut of *P. americana* treated with 0.01% deltamethrin (4X)

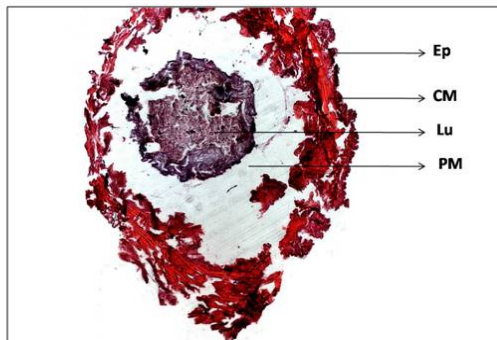


Fig.6: T. S. of midgut of *P. americana* treated with 0.01% deltamethrin (10X)

Abbreviations: BM- Basement Membrane, CM-Circular Muscle, Ep-Epithelium, LM-Longitudinal Muscle, Lu-Lumen, Nu-Nucleus, PM-Peritrophic Membrane, SB-Striated Border.

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