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Sublethal Stress of Pyrethroids on Biochemical Contents in Prostate Gland of a Freshwater Snail, *Bellamya bengalensis*

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ABSTRACT: Effect of sub lethal concentration of pyrethroid pesticides Fenvulrate (Milfen) and Cypermethrin (Cyperkil) on prostate gland of aquatic snail, *Bellamya bengalensis* was evaluated. The histological, histochemical and biochemical analysis for 15 days exposed snail was made in the present work. 24 hrs treatment of both pesticides caused cellular swelling. 7 days treatment caused distinct alterations in size, displacement of secretary cells and their arrangement in the globules. 15 days treatment resulted in damage to the prostatic acini and its tubules as well as its cells. The amount of biochemical components was greatly influenced by fenvulrate which causes depletion in glycogen, protein level, whereas cypermethrin stimulated slightly decline in metabolic contents. The ascorbic acid level decreased insignificantly in exposure to both pesticides.

Key words: Fenvulrate Cypermethrin, *Bellamya bengalensis*, Glycogen, Protein, Lipid and ascorbic acid

Introduction

The structure and function of the reproductive tract of snails and slugs have been studied with increasing interest in recent years. The functions of different reproductive organs have also been investigated. Hubendia (1948) studied on the anatomy of Bulinus with a discussion of the term prostate and its sense in the Basommatophora. Bretchneider (1948) on the mechanism of ovipostion in *Lymnea stagnalis* and Lavioletta (1954) on the role of gonad in the maturation of the reproductive tract of Arionidae. Quattizini (1967) studied on the structure and ultrastructure of the molluscan prostate gland. Plesch et.al. (1971) showed histological and histochemical changes in the reproductive organs of *Lymnea stagnalis*. Nanaware (1975) repeated the data on histochemical and biochemical changes in some gastropod snails. Bhatlawande (1989) studied the effect of hepatopancreas of a terrestrial snail, *Cerastus moussonianus*. Magare (1993) studied effect of pesticide Hygro in prostate gland of a snail, *Cerastus moussonianus*. Recently few investigations have been made on histochemical changes of snail due to various pesticides in reproductive organs of snails.

Most of the snails are serve as an intermediate hosts for certain parasitic worms of man and his domestic animals. The predilection of snails for fungal foods increases the attractiveness of diseased plant and possibility of spreading of the disease by these snails.

Histochemical nature of the reproductive tract of pulmonates is mainly limited to the exploration of functional aspects of various reproductive organs. The histochemical studies are limited to Ariophanta and Lymnea. Histochemistry of the reproductive tract of *Laevicaulis alte* revealed that the glycogen and alkaline phosphates were found throughout the epithelium of reproductive tract. Utilization of carbohydrate and protein in *Bullimus delbatus* have been studied by Horne (1973). Nanaware (1974) has made a brief review of the histochemical work on different metabolites and enzymes in the reproductive tract of different gastropods.

As there is paucity of information on this aquatic snail, regarding prostate gland in male, the present work was undertaken to investigate its histological, histochemical and biochemical changes due pyrethroid pesticides fenvulrate and cypemethrin in a snail, *Bellamya bengalensis*.

Material and Methods

Male *Bellamya bengalensis* were collected from Aner Dam near Shirpur and maintained in the laboratory condition for acclimation. LC_{50} values for 24 hr were determined by exposing the snails to pesticides fenvulrate and cypermethrin during breeding season. Exposure for lethal concentration was extended upto 15 day for sub lethal concentration. At the end of experimented snails were sacrificed and prostate gland was separated.

For histological studies prostate gland was fixed in Bouins fixative for 24 hrs. Fixed tissues were dehydrated in alcohol, cleaned in xylol and embedded in paraffin wax. Sections of 6-7 u thickness were cut and stained with Heildelihains iron Haematoxylene with eosin as a counter stain.

The staining technique used for histochemical localization are Best's carmine (Glick, 1949) for glycogen detection, Mercuric Bromophenol blue (Mazia et.al 1953) for proteins and Sudan Black B (Mc. Manus, 1946) for lipids. For biochemical estimations dry powder was used and its weight was kept practically constant through the experimental work. Glycogen was estimated by Kemp et.al. (1954) method, proteins by Lowry et.al. (1951) method, lipids by Barns and Blackstock (1975) method and ascorbic acid by Chinnoy (1969) method. Each value given here is the mean and SD of three different preparations and each preparation was assayed three times. A variation was considered significant at 5% level of probability.

Results

LC_{50} values for pesticide fenvulrate and cypermethrin have been calculated by simple graphical method which corresponds to 0.008 ppm for fenvulrate and 0.005 ppm for cypermethrin. For the above result compel as to come to universally accepted conclusion that the pesticides are more unspecific and unpredictable as far as their action on the animal body is concerned. In the present case the pesticidal stress of fenvulrate affects the general structure of the gland and its secretary cells more effectively than that of the cypermethrin. The lobules lined with these secretary cells called the prostatic acini. Effect of 1 day exposure of both these pesticides caused cellular swelling. Seven days treatment resulted in alterations of the acini and its capsule cells were also found to be damaged.

Histochemical studies revealed that the utilization of glycogen and protein was intense in exposure to fenvulrate and cypermethrin while it was steady during cypermethrin stress after 1 and 7 days exposure period whereas very slight and steady decline in lipid and ascorbic acid was recorded.

In *Bellamya bengalensis* the effect of fenvulrate and cypermethrin revealed that mobilization and utilization of glycogen, protein, lipid and ascorbic acid took place during (male) maturation cycle and this mobilization resulted in the decrease in glycogen and protein contents after 1 and 7 days exposure of both pesticides. While the lipid and ascorbic acid level was found to be declined insignificantly. Effective decline in metabolites was found in exposure to fenvulrate whereas steady utilization of metabolites was observed in exposure to cypermethrin pesticide. (Table-1)

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Table 1 : Effect of pyrethroid Fenvulrate on biochemical contents of Prostate gland of a snail, *B.bengalensis*.

Metabolites	Control	1 Day	7 Day	15 Day
Glycogen	26.38± 1.00	29.53± 1.23	22.00± 1.92	19.66± 1.30
Protein	20.77± 0.88	17.80± 1.86	17.14± 1.11	12.00± 1.22
Lipid	20.08± 0.96	1.89± 0.85	1.66± 0.31	1.40± 0.28
Ascorbic Acid	21.76± 1.00	20.61± 1.92	19.34± 1.90	12.62± 1.32

Fig 1: Effect of pyrethroid Fenvulrate on biochemical contents of Prostate gland of a snail, *B.bengalensis*

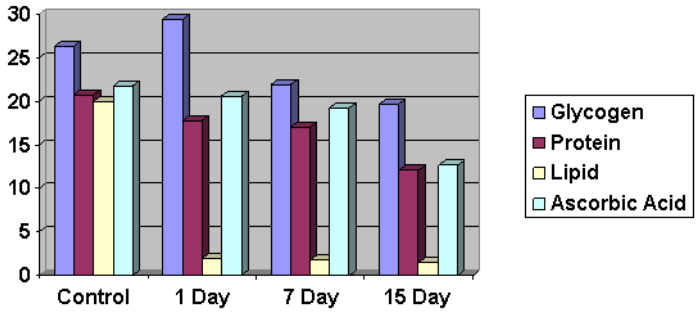
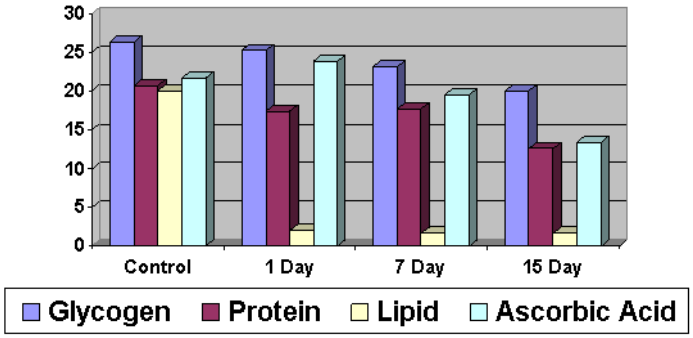


Table 2 : Effect of pyrethroid Cypermethrin on biochemical contents of Prostate gland of a snail, *B.bengalensis*

Metabolites	Control	1 Day	7 Day	15 Day
Glycogen	26.38± 1.00	25.28± 1.36	23.21± 1.60	20.00± 1.00
Protein	20.77± 0.88	17.40± 1.53	17.67± 1.73	12.66± 1.33
Lipid	20.08± 0.96	2.03± 0.05	1.67± 0.66	1.60± 0.80
Ascorbic Acid	21.76± 1.00	23.80± 2.30	19.62± 1.39	13.36± 1.26

Fig. 2: Effect of pyrethroid Cypermethrin on biochemical contents of Prostate gland of a snail, *B.bengalensis*.



Discussion

The metabolic needs are dependant on nutritional and reproductive state and some phylogenetic factors. For maximizing the power of reproduction, individuals naturally devoted its more energy to the reproduction. In the present investigation the histological, histochemical and biochemical studied were carried out on prostate gland of a snail, *Bellamya bengalensis* is exposure to pyrethroid pesticides fenvulrate and cypermethrin. In which the pesticidal stress of fenvulrate was stood effective than that of cypermethrin. During fenvulrate exposure the general structure of the prostate gland and its secretary cells were found to be displaced. Swellings of prostatic acini were found due to 1 day exposure while rearrangement and damage of prostatic acini and capsule cells was found after 7 days exposure. The present results are co-related with the findings of Magare (1993).

Pesticides are widely used to combat agricultural pest and their application has greatly contributed to stopping up to the agricultural production. But none of these pesticides employed are specific and due to their indiscriminate and wide spread use, several non-target organisms like snails, fishes, crabs etc. of the ecosystem are adversely affected.

The effect of fenvulrate on prostate gland of B. bengalensis showed intense staining activity which indicates significant decline in glycogen and protein levels. Glycogen is available for instant energy source while proteins are important organic constituents of the animal cells playing a vital role in the process of interactions between intra and extracellular media. The active depletion of these metabolites might be due to their mobilization to liberate energy during pesticidal stress. Umminger (1970) observed that the aquatic inhibitants exposed to toxic conditions, utilized proteins as the energy source. Similar observations were also noted in *Pila globosa* (Ramana Rao and Ramamurthy, 1980). These results partly correlate the present findings.

The main organic reserve shows variations in their contents due to pesticidal stress. A great deal of energy is to be channelized during reproduction. This reflects the depletion in nutrient depot which the advent or departure of the reproductive season (Webber, 1970). The present work has been carried out during breeding season. The significant decline in glycogen and protein was more pronounced due to fenvulrate exposure while lipid and ascorbic acid level was declined insignificantly due to pesticides. In the present findings the histochemical results correlates the biochemical changes. Results of the present investigation correlates with the findings of Magare

(1993), Laviolette (1954), Nanaware (1975) and Bhatlawande (1989).

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