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Effect of an organophosphate pesticide in the ovulation of air breathing fish *Anabas testudineus* (Bloch, 1792)

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Abstract : The pollution of environment due to use of pesticides has become an increasing problem over the last century with the development of industry, agriculture and increase in population. The organophosphate pesticides are widely used because of their rapid biodegradability and non-persistent nature. Changes occurring in the haematological characteristics provide a sensitive measure to assess the fish health. Pesticide is a major factor for fish mortality along with water temperatures. The main purpose of the study was to show the vital effect of organophosphate pesticides in the ovulation of air breathing fish.

Keywords:- Organophosphate pesticides, *Anabas testudineus,* biodegradability.

INTRODUCTION

Pesticides as a group of environmental contaminants cause severe toxicity, both acute and chronic in non-target species. Pesticides are a group of environmental contaminants causing severe toxicity, both acute and chronic in non-target species. The word "organophosphates", when appearing in communications (e.g., from the press or the government), in areas such as agriculture, the environment, and human and animal health, very often refers to a group of insecticides (pesticides) that act on the enzyme acetylcholinesterase. Today, organophosphates make up about 50% of the killing agents in chemical pesticides.[1] Organophosphate pesticides (OPPs), like some nerve agents, inhibit this neuromuscular enzyme, which is broadly essential for normal function in insects, but also in humans and many other animals. Literature abounds with reports on the impact of pesticides on various fish tissues but studies on reproductive toxicity in fish are not as many. Studies on pesticidal effects on female reproduction are abortion in *Gambusia affinis* (Boyd 1964)[1], reduction in reproductive efficiency in brown trout, *Salmo trutta,* and brook charr, *Salvelinus fontinalis* (Burdick *et al.* 1972)[2], prevention of reproduction by Carbaryl (Carlson 1972)[3], reduced hatching of embryos by PCB in brook charr (Freeman & Idler 1975)[4], retardation of ovarian development by Fenitrothion and Carbaryl (Saxena & Garg 1978)[5] and Phenthoate (Dey & Bhattacharya 1989)[6] in *Channa punctatus* and by Malathion and Endrin in *Heteropneustes fossilis* (Singh & Singh 1980)[7]. Recently we reported the reproductive toxicity of Metacid-50 pituitary GtH content and hypothalamic GnRH activity in *C. punctatus* (Ghosh *et al.* 1990)[8]. The present work is a follow up of our earlier observation to evaluate

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the effects of non lethal concentrations of metacid- 50 (Methyl-parathion 50%) and carbaryl on the ovarian weights (GSI) and levels of estrogen in the ovary and plasma of the Indian climbing perch, *Anabas testudineus* (Bloch) during an exposure regimen of 90 days extending over the pre-spawning and spawning phases of the annual reproductive cycle.

MATERIALS & METHODS

Pesticides

Metacid-50 is a commercial organophosphate containing 50% methyl-parathion as the active ingredient. Carbaryl is also a commercial formulation containing 50% n-methyl naphthyl-1-carbamate as the active ingredient.

Healthy fishes were procured from local fish market and were kept in cemented tank. The water depth of 0.5-0.75 m was maintained in the tank. Fish were fed twice daily, i.e., 11 am and 6 pm. Perfectly mature female fish have swollen, reddish vent and ooze eggs while the males ooze milky milt upon slight pressure in the abdomen.

Treatment

The tolerance limit for Metacid 50 was 5.33 ppb and that for Carbaryl was 15.83ppm. Depending on the mortality rate & phusiological distress the exposure doses was selected. Since the experiment was carried out for longer duration the fishes were once daily. As per Anon, (1960)[9] regulations the water was changed daily and 0.106ppb of Metacid-50 and 1.66 ppm of Carbaryl were at first dissolved in 20 ml of the aquarium water and then added freshly to negate the effect of metabolites on the toxicity of the poison. the experimental was done for the determination of GSI and hormonal profiles on day 3,7,15,20.

The gonads were dissected out and weighed on a single pan balance. The GSI was calculated on 100g body weight on the basis of given by Abidin (1986)[10].

Collectiojn of blood was done from caudal region and by the process of centrifugation plasma was separated at 2500 g at 4° C for 10 min. The ovary was disected very carefully and immediately homogenized in 0.6% saline; after centrifugation the supernatant was decanted and stored at 4°C for future use.

Radioimmunoassay of l7P-Estradiol

Extraction efficiency was more than 85%. Anti-Estradiol-17/3 was diluted 1:25000 in phosphate buffered saline-gelatin (PBSG). 100p1 of antiserum were added to the standards which ranged from 10 to 1000pg. After the incubation of the samples for 1 h, of 3H-E2 were added to each tube. Samples were incubated at 4°C for a night and then placed in an ice bath for 15min. Samples were then centrifuged, decanted into scintillation fluid. The results were expressed as mg per ml plasma and pg per mg of ovarian protein. Samples were then prepared and stored at -24°C til the end of the sampling period.

RESULT AND DISCUSSION

When the fish *Anabas testudineus* was treated with the carbaryl 15 days after exposure there was no deviation from the control value of GSI (Gonadosomatic index). But however when it was treated from 20 days onwards until the termination of the experiment there was great variation in GSI level from the respective control value. The profiles of the plasma and ovarian estradiol-17P (E,) of the control fish showed a gradual increase until 60 days of observation. When the ovarian E_2 level was observed 90 days after exposure showed decrease in the level as compared to those experimented for 60b days. In the case of plasma1 & ovarian E, levels in the organophosphate treated fish a gradual increase was observed from day 3 to 15 days of treatment, thereafter showing a sharp decline until day 60 of treatment. However, at 90 days of sampling both plasma and ovarian E level, it showed little improved 87% decline of plasma E_2 and 29% in the ovarian E, level over the control. After 15 days of carbaryl treatment the plasma E_2 titre showed significant increase (18%-128%) while in the later period from 20-60 days of exposure the hormone level decreased (19%-83%) significantly. However, at 90 days, the degree of depletion was not very remarkable. Ovarian E_2 level, on the other hand, showed a small decline on day 3rd day leading to 25-45% increment on day 7 and 15 day, respectively. This increase subsided to only an 8% increase on day 20 which was followed by significant depletion from day 30 to 90.

The exposure of the two different classes of pesticides caused no mortality of the fish or sign of physiological distress. However, they were both potent enough to cause significant reproductive impairment to ovarian tissue. Histological examination revealed a

preponderance of stage I and destruction of stage 11 and stage 111 oocytes in the fish treated with the pesticides (unpublished observation). As reported by Shukla (1982)[11], GSI is greatly affected by DDT, BHC, endosulfan, chlordane and toxaphane. Dey & Bhattacharya (1989) observed the preponderance of stage I and destruction of stage 11 and stage 111 oocytes in association with decreased ovarian weight in phenthoate exposed *C. punctatus*. Previous workers have suggested an impairment of steroidogenic activity in the gonad of pesticide treated fish (Freeman & Idler 1975, Saxena *et al*. 1986, Kapur *et al*. 1978)[4,12,13]. GtH in *Channa punctatus* exposed to sublethal doses of metacid-50 and carbaryl for 30 days. It may, therefore, be suggested that the decreased steroidogenic activity in *Anabas testudineus* during this period may be due to the decreased release of GtH during a long term exposure.

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