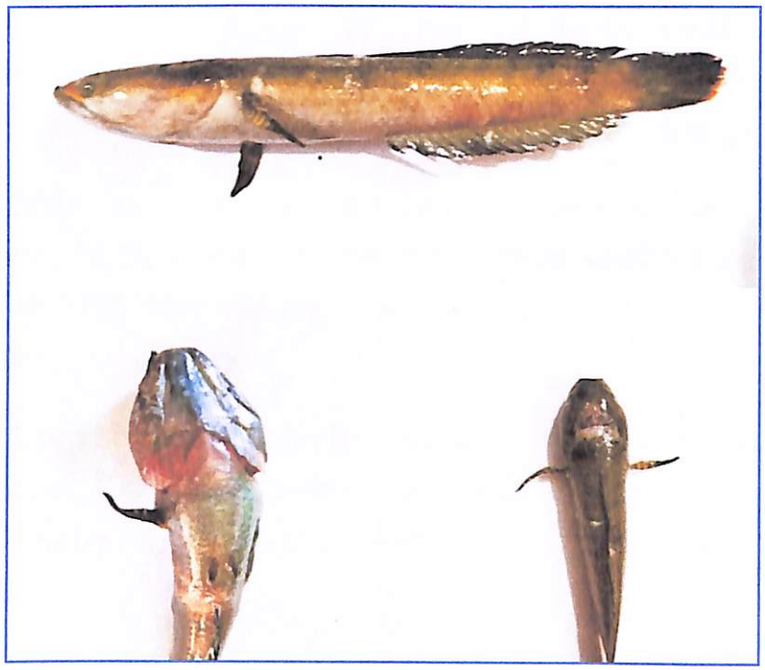
EFFECT OF PESTICIDES ON BEHAVIOURAL AND MORPHOLOGICAL CHANGES IN *Channa gachua.*



A dissertation submitted in partial fulfillment of the requirement for the degree of Master of Science in Zoology

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ASSAM SCIENCE AND TECHNOLOGY UNIVERSITY
(2022)

CERTIFICATE

This is to certify that the dissertation entitled "EFFECT OF PESTICIDES ON BEHAVIOURAL AND MORPHOLOGICAL CHANGES IN *Channa gachua*" submitted in partial fulfillment of the requirement for the degree of Master of Science in Zoology is a compilation of the result of bonafide work carried out by Aparajita Boruah (Reg. no.: 450228220, Roll no: 202820024001), department of ZOOLOGY, Silapathar Science College affiliated by ASSAM SCIENCE AND TECHNOLOGY UNIVERSITY, (ASSAM) under my guidance and supervision .

The dissertation or any its part has not been submitted elsewhere for any other degree of distinction in any other university/institution. All the help and assistance received during the course of work have been duly acknowledged.

I am pleased to forward this dissertation for consideration for the award of the degree of Master in Science in Zoology (Under Silapathar Science College) affiliated by ASSAM SCIENCE AND TECHNOLOGY UNIVERSITY, ASSAM.

Date : 23-07-2022
Place : Silapathar Science College

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CANDIDATE'S DECLARATION

I, Aparajita Boruah, hereby declare that the research work entitled "EFFECT OF PESTICIDES ON BEHAVIOURAL AND MORPHOLOGICAL CHANGES IN *Channa gachua*" in partial fulfillment of the requirement for the degree of Master of Science in Zoology, is being presented in the form of thesis and submitted in the department of Zoology, Silapathar Science College affiliated by ASSAM SCIENCE and TECHNOLOGY UNIVERSITY, (Assam) under the supervision of Dr. Jashodeb Arjun, associate professor.

The matter presented in the project has not been submitted by me for any other degree of this or any other institute.

Aparajita Boruah
Signature of the candidate

This is to certify the above statement made by the candidate is correct to the best of my knowledge.

Date: 23-07-2022

Signature of supervisor

ACKNOWLEDGEMENT

It is always a pleasure to remind the fine people for their sincere guidance I have received to uphold my practical, field and laboratory skills in the accomplishment of my dissertation, as a study like this can never be the outcome of the effort of a single person, rather it bears the imprint of a number of people who directly or indirectly helped me in the partial fulfillment of the project.

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Lastly, I thank my parents, friends for their cooperation and support.

Date : 23-07-2022 Aparajita Boruah

CONTENT

1.INTRODUCTION:

The pollution of rivers and streams with chemical contaminants has become one of the most critical environmental problems. A chemical that is toxic to one animal may also be toxic to other forms of animal life. With the growth of the modern civilization, our life is in danger due to pollution of water both from surface and underground. Water pollution has emerged as one of the most burning tribulations of this century. No other natural resource is more contested than water. Water pollution threatens the survival of life on this planet (Shrivastava et al. 2018).

Nowadays, the use of pesticides has become common around the world not only on farms but in backyard gardens as well. Farmers began to use advance technology to enhance yields by using synthetic fertilizers, pesticides and herbicides. Pesticides include insecticides, fungicides, rodenticides, biocides and herbicides. These can be classified by target organism, chemical structure and physical state. Pesticides are those chemicals, which have been widely used throughout the world to increase crop yield and to kill the insect-pests responsible for transmitting various diseases to humans and animals (Shrivastava *et al.* 2018).

Exposure of insecticides influences some important physiological functions of fish. It greatly affects fish production and human health too through ecological cycling and biological magnification. Accumulated pesticide acts as a prooxidants and enhances prooxiradial generation by reacting with Oxygen. Indiscriminate use of different pesticides in agriculture kills not only the pests but also a wide variety of non target organisms like fish and other aquatic fauna. These pesticides even when applied in

restricted areas are washed and carried away by rains and floods to large water bodies like ponds and rivers and alter the physico-chemical properties of water. Last few years have witnessed about multi fold increase in the use of synthetic pesticides. Due to these synthetic chemicals, environment has failed to keep its healthy characteristics.

Among the various kind of pesticides used in India, the synthetic pyrethroids are being largely used by the farmers due to the low toxicity in mammals compared to other organophosphorus and organochlorine pesticides found in environment (Kannan *et al.* 2014).

In animal kingdom, fishes being the well-defined and highly successful taxonomic group of vertebrates have drawn immense curiosity of the naturalists as well as zoogeographers from the entire environment of the world. In the present global scenario, fish group is formed by 32,700 species which covers more than 50% of the total vertebrate species. With the new discovery of fish species every year, the total fish species raises from 20,000 or 21,723 to 32,700 species. At present of the total species found the 13000 species (39.75%) occupy freshwater habitat. Recently, fish species represent only a fraction of the number and variety that survived in the past. The largest diversity of freshwater fish species found in America followed by Africa and Asia (Bhanjit Bhatt,2009).

India is having 2,500 fish species, where 930 are fresh water with 1,570 marine species and secured ninth ranks regarding mega freshwater biodiversity, with the northeast region recognized as a

global hotspot of freshwater fish species. Fishes are those invariable living components which act as important food resources and good indicators of the ecological health of the aquatic ecosystem they inhabit (Kaushik, 2017).

Assam is truly riverine state drained mainly by two river system, the Brahmaputra and the Barak. In the floodplains of these rivers are found a large number of lakes like natural water bodies and swamps. These water bodies are locally known as beel, while the marshes and swamps are generally known as "jalah", "doloni" etc. (Bhuyan *et al.* 2017).

All snakehead species of *Channa* belong to the family *Channidae*, a lineage of freshwater fishes that is characterized by usually having air-breathing ability given the presence of the supra-bronchial organs. Among various groups of freshwater fishes, "murrels" or "snakehead" is considered as unique group with high commercial value (Haniffa et al., 2003; Benziger et al., 2011; Ali *et al.*, 2013).

Only two genera, *Channa* and *Parachanna are* known under the family Channidae comprising of 41 (38 *Channa* and 3 *Parachanna* species) valid species in the world, of which 16 species have been reported from India (Froese and Pauly, 2019). According to Vishwanath and Geetakumari (2009), North East India has an important diversity for about nine species of *Channa*. Eight species belonging to genus *Channa* (*C. striata, C. punctata, C. gachua, C. marulius, C. stewartii, C. bleheri, C. urantimaculata, and C. barcaand*) are found all over India (Waghmare *et al.* 2017).

The species *Channa gachua* is taken for the behavioral and morphological study. It is least concern as per IUCN red list of threatened species database (IUCN 2014). This fish is distributed throughout Asia (i.e. Bangladesh, India, Nepal, Myanmar, Pakistan, etc.) mostly abundant in ponds, swamps, brackish water (Pethiyagoda, 1991) and also in ditches and beels. It has great commercial importance in Asian region (Rahman, 1989). It is an important food fish and also used in pharmaceutical industries and in household as ornamental species. It is one of the most suitable *Channa* species for aquarium due to its beautiful coloration and small size (Wee 1982; Sampath and Pandian 1984, Williams 2004).

The "dwarf snakehead" or "walking snake" *Channa gachua* inhabits wetlands and riverine ecosystem (Vishwanath and Geeta kumari, 2009).This species can live in poorly oxygenated, turbid and even very foul stagnant water bodies (Kumar et al. 2013). It can be used for the biological control of mosquito in larval stage ultimately helps to control the diseases spread by mosquitos (Phukan and Biswas 2011).

Adaptation to local climatic conditions plays a key role in survival, growth, reproductive success, distribution and evolution of a species (Atkins and Travis, 2018; Petersons, 2014).

COMMON NAME : Sengali

TAXONOMIC CLASSIFICATION:

Phylum– Chordata

Superclass- Actinopterypgii

Class- Teleostei

Order- Perciformes

Suborder- Channoidei

Family- Channidae

Genus- *Channa*

Species- *Channa gachua* (Hamilton, 1822)

MORPHOLOGICAL STUDY:

It is a species of *Channa* having dorsal fin rays 32-37. Body of *Channa gachua* is compressed posteriorly. Head is depressed and flat above. Adult Dwarf snakeheads are typically dark bodied with red or orange margins on their dorsal, anal and caudal fins. 39-48 scales present in the lateral line. Pelvic fin is shorter than half of the length of the pectoral fin rays. There are 15-17 pectoral fin rays, 21-27 anal fin rays and 12 caudal fin rays is present in *Channa gachua*. Dorsal, anal and caudal fin margins are white in color. They are relatively small in size. Because of its pretty coloration and small size, it is an attractive fish and commonly kept in aquaria. Individuals can grow to 28.8 cm. They have sexual reproduction. It is a hardy fish and mouth brooder.

HABITAT:-

They are tolerant of a wide range of water conditions, including both fresh and salt water. They are primarily nocturnal animal, but can adapt to being out during the day. This species can live in poorly oxygenated, turbid and even very foul stagnant water bodies (Kumar et al. 2013).

FOOD AND FEEDING HABITAT:-

The qualitative and quantitative food analysis of fish in their natural habitats helps in understanding the growth, abundance, productivity of water body (Prakash, 2016). They are voracious predators. They will eat almost anything that fits in their mouth. These fish are carnivores. In their native habitat they eat insects, larvae, worms, other invertebrates and even small fish.

2.OBJECTIVES OF STUDY:-

1 To determine the effect of various pesticides (insecticides).

2 To determine the effect of pesticides as water pollutant.

3 To determine the morphological changes in fish *Channa gachua* due to the toxic effect of pesticides.

4 To determine behavioral changes in *Channa gachua* due to the toxic effect of pesticides.

3.REVIEW OF LITERATURE:-

Mahaboob *et al.* (1983) conducted a survey and showed Malathion (organophosphate). BHC (organochlorine) and to some extent carbaryl (carbamate) are much used. Hence, the authors have chosen these three pesticides for the toxicity studies on the freshwater teleost. *Tilapia mossambica*, which is a common species of freshwater ecosystem. The three pesticides selected for the investigation belong to different categories, each having specific properties and a particular mode of action.

Dutta *et al.* (1995) conducted a study to determine whether there are differences in the sensitivity of brain acetyl cholinesterase between juvenile and adult fish exposed to Malathion. Air breathing catfish, *Heteropneustes fossilis* were used. The exposure concentration was 1.2 mg/l (sub lethal), and exposure duration were 24, 48, 72 and 96 hrs. However, in the adults a reduction is seen, only at the 72 hrs exposure duration. T-value and two-tail probability show that only control and 72 hr exposed ones are significantly different. The results show that the juveniles are more susceptible than the adults.

Reddy *et al.* (1995) exposed freshwater fish, *Cyprinus carpio* to sub lethal concentration of cypermethrin (20 micro/l) for 6, 12, 24 and 48 h to determine the protein fractions, amino acids, protease, alanine aminotransferase, aspirate aminotransferase, ammonia, urea and glutamine levels in gill, brain, liver and muscle tissues. Total structural and soluble proteins showed decrement; whereas free amino acids and the activities of protease, aspirate aminotransferase significantly increased at all exposure periods in cypermethrin exposed fish. It was observed that the changes

steadily increased with an increase in the period of exposure and exhibited tissue specificity.

Vishwanath *et al.* (2009), studied on the diagnostic characters and interrelationship of fishes of the genus *Channa.* The study was based on the nine species of *Channa* which is found in northeast India.

Kannan et al. (2014), conducted a study aimed at assessing the effects of a synthetic pyrethroid cypermethrin (10% EC) on *Catla catla* (Indian major carps) on the basis of the results of sub lethal toxicity tests and hematological and biochemical examinations. Experimental carps were exposed to the pesticide cypermethrin in the concentration of 0.0006 ml/lit in a 24h sub lethal toxicity test. It caused significant shifts in hematological and biochemical profile.

Srivastava et al. (2018), studied on pesticides, classification of pesticides, pesticide pollution, pesticides as water pollutants. Also they study on the origins of pesticide entry into water & pesticides management and control method. Water pollution can come from either diffuse or point sources. Waghmare et al. (2017), exposed freshwater *Channa punctata,* as a model for toxicological studies. They used pesticide Applaud for 24, 48, 72 and 96 h exposure to be 459.29, 326.12, 253.30 and 198.84 ppm respectively. They compared toxicity of different pesticides such as buprofezin, fenvalerate, cypermethrin, malathion, etc. on the basis of previous studies.

Islam *et al.* (2019) compared the effect of malathion on some behavioral changes and mortality of three indigenous fish species

Channa punctatus, Heteropneustes fossilis and *Anabas testudineus.* The effects of malathion were observed for different exposure period (24-96 hours) with the parameters of mortality, toxicity and physical behaviors. The LC50 was 5.67 mg/l, 5.19 mg/l, and 5.22 mg/l for *C. punctatus, H. fossilis,* and *A. testudineus* respectively in 96 hours.

Singh et al. (2020), performed a experiment to determine the acute toxicity of cadmium to freshwater fish, *Channa punctatus.* The objective of this study was to grasp the link between mortality and abnormal behavioral and morphological changes of *C. punctatus.* The lowest cadmium chloride concentration at which mortality was observed as 45 mg/l. loss of balance during swimming, observed during this study, might be due to some neurological impairment in the central nervous system.

Sultana et al. (2020), conducted a investigation to assess morphological and behavioural changes and to evaluate the histological changes of liver, stomach and intestine of *Channa punctatus* at different concentration of acid dye. 40%, 60% and 80% mortality were recorded at 0.07, 0.09 and 0.2 ml/L concentration of acid dye. The study revealed toxic effects of acid dye on *C. punctatus.* This study also recommends proper inspection of acid dye toxicity before using it in any industry.

Chaudhary et al. (2021), determined the behavioural alterations and morphological deformities in the freshwater fish *Channa punctatus* due to exposure to different sublethal concentration of carbamate pesticide carbaryl. *C. punctatus* were exposed to concentration ranging between 0.1ppm to 1.31ppm in *in vitro* conditions in the laboratory. 100% mortality was observed due

to exposure to 1.17ppm or more concentration of carbaryl. The response of the fish towards toxicity was dependent on concentration of carbaryl and length of exposure.

Singh (2021), exposed freshwater fish, *Channa gachua* to sub lethal concentration of malathion for 20 days to study histopathological changes. The exposure concentration was 0.8 or 1.6 microg/l Malathion. The exposure of Malathion pesticides can cause serious damages in gills, liver, spleen and trunk kidneys of *Channa gachua*. Mild histological lesions were observed in gills, liver, spleen and trunk kidney of *Channa gachua* exposed to sublethal concentration of Malathion. Serous lesions were observed in the gills of fish exposed to the higher concentration (acute) i.e. 1.6 µg/l. It was observed that the changes increased with an increased in the period of exposure and exhibited tissue specificity.

4.MATERIALS AND METHODS:-

4.1 Period of study:-

The duration of the experiment was from the month of May to June, 2022.

4.2 Collection of sample:-

Channa gachua is used in the present study. The fish samples were collected from local water bodies with the help of traditional fish catching gears. The fish were caught and kept in jar filled with water. The length and weight of the specimens were measured. The length of the specimens were found between 11.1cm to 13.6 cm, width 2.2cm to 2.5 and weight 21gm to 23gm.

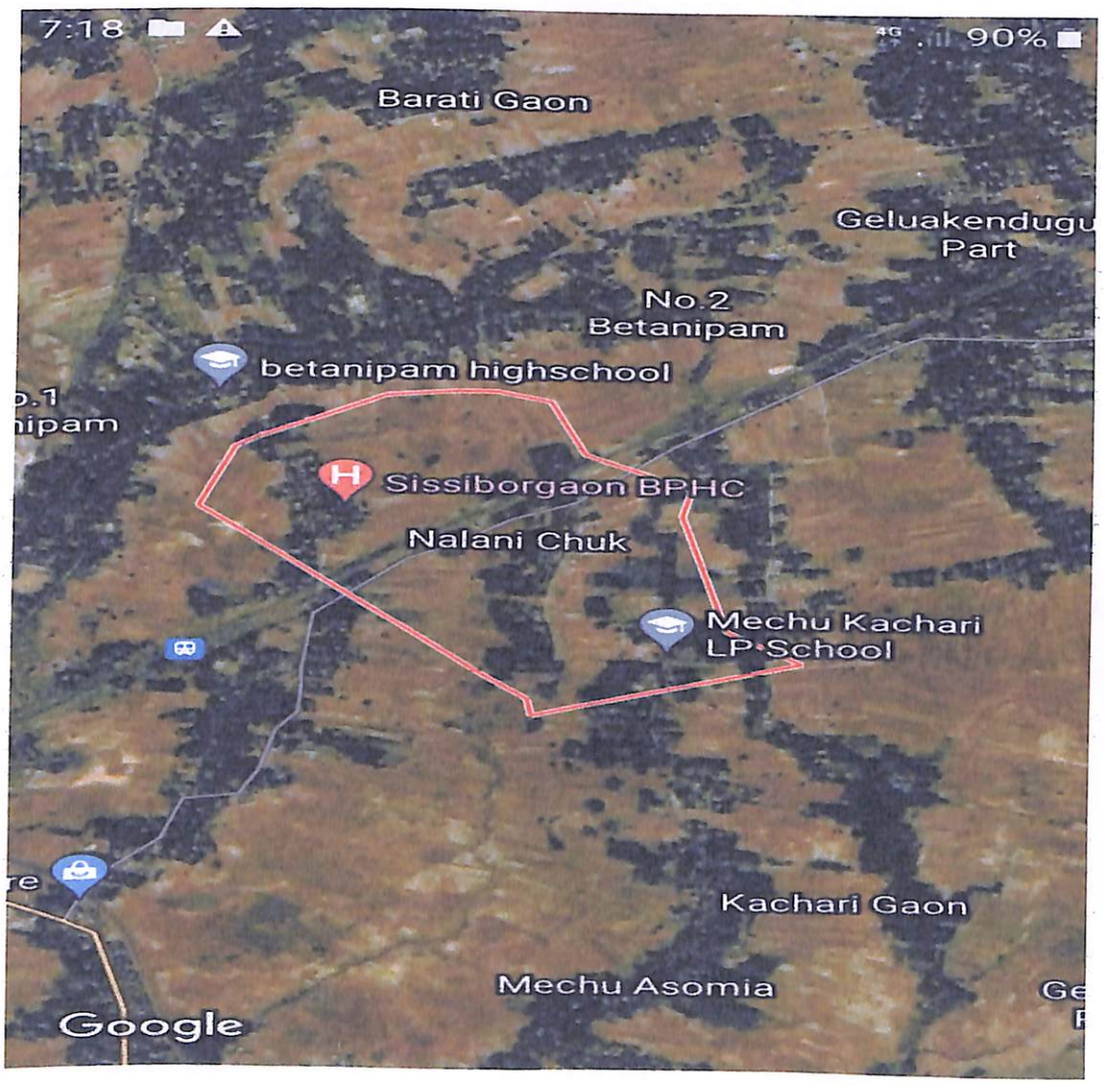
After collection the fishes were immediately released into a large bucket containing tap water with pH 7.2 and maintained with algal food and plankton.

The water medium was changed at 24 hours interval to remove metabolic pollutants. After the acclimation fresh, healthy fishes were used for experiments. Fishes were held in 12 hours light and 12 hours darkness period throughout experiment.

4.3 Study Area:-

Dhemaji district is situated at the north of upper Brahmaputra valley of Assam which is located between the 94° 12' 18" E and 95° 41' 32" E longitudes and 27° 05' 27" N and 27° 57' 16" N latitudes. The district covers an area of 3237 sq. km and is basically plain area lying at an altitude of 104m above the mean sea value.

The samples were collected from local pond of Nalani chuk, which is about 3.5km away from Silapathar town. Silapathar is a beautiful city in the Dhemaji district is located at the northern bank of the Brahmaputra river. It is just 6.7km from the border of Arunachal Pradesh. It is located between 94 83 24 longitudes and 27 53 29 3 latitudes. Silapathar has a humid, subtropical and dry winter climate.



Study site : Nalani chuk , Silapathar , Dhemaj.

4.4 Physiochemical parameters of the water of the site:-

- **Temperature:** The study reveals fluctuation of water temperature between 15.5° C and 26.3° C.
  (Mean= 22.04, σ= 3.41)

- **pH:** Fluctuation of pH is observed between 6.5-7.5.
  (Mean= 7.05, σ= 0.35)

- **Dissolved Oxygen:** Dissolved Oxygen in the studied area seems to be varied from 4.7 to 11.2 mg/l.
  (Mean=7.8, σ=1.82)

- **Free Carbon dioxide:** The value of free CO2 ranges between 4.0-8.0 mg/l.
  (Mean= 5.5, σ= 1.51)

- **Total alkalinity:** The alkalinity of water from the studied area is exclusively represented by the bicarbonate or methyl orange alkalinity. The range of its concentration is recorded in between 26.0-115.0 mg/l.
  (Mean= 60.17, σ=27.37)

- **Total hardness:** The value of total hardness of the studied water bodies is calculated within a range of 34-116 mg/l.
  (Mean= 78.67, σ= 8.65)
- **Calcium:** The range of calcium hardness expressed as calcium carbonate is recorded in between 13.4 to 46.5.
  (Mean= 30.24, σ= 8.64)

- **Magnesium:** The magnesium hardness is ranged between 6.2 to 19.2 mg/l.
  (Mean=12.65, σ=4.73)

All the hydrological parameters were done in Lumding College lab of Zoology Department under the guidance of Dr. J. Arjun.

Experimental water:

The main source of experimental water was tap water. pH paper conducted to test water. The pH of the experimental water was 7.2. Dissolved Oxygen and Carbon dioxide was determined to 6.6 mg/l and 83 mg/l.

4.5 Source of toxicants:-

Malathion 5% dust as Asamal-5 and Cypermethrin 10 Ec as Ustaad were received from an insecticides retailer near daily market of Silapathar, Dhemaji.

4.6 Malathion :-

Malathion ($C_{10}H_{19}O_6PS_2$) is broad spectrum organophosphate insecticide which controls insects by contact action and by ingestion of Thiodan-treated plant material. Malathion is a chemical compound which is used as a racemate. It is a pesticide that is used widely in agriculture, in home, and in public health related pest control programs such as malaria eradication by the govt. of India.

Chemical composition of Asamal-5:-

Malathion technical-5% and

Processed carrier 95%.

4.7 Cypermethrin :-

Cypermethrin is a synthetic pyrethroid pesticide. It is one of the widely used insecticides and very efficient in agriculture and sanitary pest control. After use cypermethrin is released directly into the environment, enters the water body by runoff, and affects the aquatic ecosystem. Cypermethrin is very toxic for fish and aquatic invertebrates LC50 in the range of 0.01-5 µg/l.

It is a widely used to control cotton boll worm and to control the larvae of mosquitoes and milk fishes during pond preparation in urban and agricultural environments. In India, cypermethrin pesticide is widely used to control pests which are attacking cotton, cabbage, brinjal, sugarcane and wheat, etc. (Mohan kannan, et al. 2014).

Chemical composition of Ustaad:-

Cypermethrin 10% EC.

4.8 Mode of treatment: -

Five transparent containers were used for the experiment. Five fishes were kept in each container and each containing 8L of tap water. Fishes were exposed to four different concentrations of two pesticides for chronic toxicity (30 days). Concentrations for chronic toxicity of malathion and cypermethrin were 0.5 mg/l, 1.00 mg/l, 1.5 mg/l and 2.00mg/l and 0.5 ml/l, 1.00 ml/l, 1.5 ml/l and 2.00 ml/l respectively. Control or normal fishes were kept in tap water only. Behavioral and morphological changes were observed and recorded at regular intervals. Maintainece were carried out carefully during the experimental period.

RESULT :

Hydrological parameters of the study site:-

SL. No.	Hydrological parameters	Value
1	Ph	7.05
2	Dissolved oxygen (DO)	7.8 mg/l
3	Dissolved carbon dioxide (DCO2)	5.5 mg/l
4	Alkalinity	60.17 mg/l
5	Hardness	78.67 mg/l
6	Temperature	22.04 ° C

Table 2 : Effect of malathion exposure on Behavioral changes in ***Channa gachua.***

Sl. No.	No. Of Test fishes	Behavioral Changes	Concentration In mg/l	Exposure period Chronic test					
				1day	5days	10days	15 days	20 days	30 days
1.	5	Swimming activity	Control	-	-	-	-	-	-
			0.5 mg/l	+++	+++	+++	+++	+++	++
			1.00 mg/l	+++	+++	+++	++	++	+
			1.5mg/l	+++	++++	+++	++	+	+
			2.00 mg/l	++++	++	++	+	+	+
2.	5	Loss of Equilibrium	Control	-	-	-	-	-	-
			0.5 mg/l	+	++++	+++	+++	++	++
			1.00 mg/l	++	+++	++++	+++	++	++
			1.5mg/l	+++	++++	+++	+++	++	+
			2.00 mg/l	+++	++++	+	+	+	+
3.	5	Jumping	Control	-	-	-	-	-	-
			0.5 mg/l	+++	++++	+++	++	++	++
			1.00 mg/l	+++	++++	+++	++	++	+
			1.5mg/l	+++	++++	+++	++	+	+
			2.00 mg/l	+++	+++	+	+	+	+
4.	5	Opercular movements	Control	-	-	-	-	-	-
			0.5 mg/l	+	+++	++++	+++	++	++
			1.00 mg/l	+++	++++	+++	+++	++	+
			1.5mg/l	+++	++++	+++	++	++	+
			2.00 mg/l	+++	++++	+	+	+	+
5.	5	Restlessness	Control	-	-	-	-	-	-
			0.5 mg/l	++	++	+++	++++	++	++
			1.00 mg/l	++	+++	++++	+++	++	++
			1.5mg/l	++	++++	+++	++	+	+
			2.00 mg/l	+++	++++	++	+	+	+
6.	5	Sluggishness	Control	-	-	-	-	-	-
			0.5 mg/l	+	++	+++	+++	+++	++
			1.00 mg/l	++	+++	++++	+++	+++	++
			1.5mg/l	+++	+++	++++	+++	++	++
			2.00 ml/l	+++	+++	+++	++	+	+

(-) Normal, (+) Nil, (++) Less change , (+++) Moderate & (++++) prominent change

Table 3 : Effect of cypermethrin exposure on Behavioral changes in ***Channa gachua.***

Sl. No.	No. Of Test Fishes	Behavioral Changes	Concentration In ml/l	Exposure period					
				Chronic test					
				1 day	5 days	10 days	15 days	20 days	30 days
1.	5	Swimming activity	Control	-	-	-	-	-	-
			0.5 ml/l	++	+++	++++	+++	+++	++
			1.00 ml/l	++	++++	++++	+++	++	+
			1.5ml/l	+++	++++	++++	+++	++	+
			2.00ml/l	+++	+++	+++	++	+	+
2.	5	Loss of Equilibrium	Control	-	-	-	-	-	-
			0.5 ml/l	+	++	++	+++	++	++
			1.0 ml/l	++	+++	++++	+++	++	+
			1.5ml/l	++	+++	++++	++	++	+
			2.0ml/l	+++	+++	++++	++	+	+
3.	5	Jumping	Control	-	-	-	-	-	-
			0.5ml/l	++	+++	+++	+++	++	++
			1.0ml/l	++	++++	+++	+++	++	+
			1.5ml/l	+++	++++	+++	++	++	+
			2.0 ml/l	+++	++++	+++	++	+	+
4.	5	Opercular movements	Control	-	-	-	-	-	-
			0.5ml/l	++	+++	++++	++	++	++
			1.0ml/l	++	++++	++++	+++	++	++
			1.5ml/l	++	+++	++++	+++	++	++
			2.0ml/l	+++	++++	+++	+	+	+
5.	5	Restlessness	Control	-	-	-	-	-	-
			0.5 ml/l	++	+++	++++	+++	+++	++
			1.0 ml/l	++	+++	++++	+++	+++	++
			1.5 ml/l	+++	+++	++++	+++	++	+
			2.0 ml/l	+++	++++	++	+	+	+
6.	5	Sluggishness	Control	-	-	-	-	-	-
			0.5 ml/l	+	++	+++	+++	++	++
			1.0 ml/l	++	+++	+++	+++	++	++
			1.5 ml/l	++	+++	++++	+++	+++	++
			2.0ml/l	++	+++	++++	++	+	+

(-) Normal, (+) Nil, (++) Less change , (+++) Moderate & (++++) prominent change

Table 4 : Effect of malathion exposure on Morphological changes in ***Channa gachua***

Sl. No.	No. Of Test fishes	Morphological changes	Concentration In mg/l	Exposure period					
				Chronic test					
				1day	5days	10days	15days	20days	30days
1.	5	Patches on the body	Control	-	-	-	-	-	-
			0.5mg/l	-	+	++	++	+++	+++
			1.0 mg/l	+	+	++	++	+++	++++
			1.5mg/l	+	++	++	+++	++++	++++
			2.0 mg/l	++	++	+++	+++	++++	++++
2.	5	Discoloration of skin	Control	-	-	-	-	-	-
			0.5 mg/l	-	-	+	++	++	+++
			1.0 mg/l	-	+	++	++	+++	+++
			1.5mg/l	-	+	++	++	+++	+++
			2.0 mg/l	++	++	++	+++	+++	++++
3.	5	Shedding of scales	Control	-	-	-	-	-	-
			0.5 mg/l	-	+	++	++	+++	+++
			1.0 mg/l	+	+	++	++	+++	++++
			1.5mg/l	+	++	++	+++	+++	++++
			2.0 mg/l	++	++	+++	+++	++++	++++
4.	5	Mucous secretion	Control	-	-	-	-	-	-
			0.5 mg/l	+	++	++	++	+++	++++
			1.0 mg/l	+	++	++	+++	+++	++++
			1.5mg/l	++	++	+++	+++	++++	++++
			2.0 mg/l	++	++	+++	+++	++++	++++
5	5	Bulging eye's	Control	-	-	-	-	-	-
			0.5 mg/l	-	+	+	++	+++	+++
			1.0 mg/l	+	+	++	++	+++	+++
			1.5mg/l	+	++	++	+++	+++	++++
			2.0 mg/l	++	++	+++	+++	++++	++++
6.	5	Clumping of gills	Control	-	-	-	-	-	-
			0.5 mg/l	-	+	+	++	++	+++
			1.0 mg/l	+	++	++	++	+++	+++
			1.5mg/l	+	++	+++	+++	+++	++++
			2.0 mg/l	++	++	+++	+++	++++	++++

(-) Normal, (+) Nil, (++) Less change , (+++) Moderate & (++++) prominent change .

Fig: Patches on Body

Fig:Shedding of scales

Table 5 : Effect of cypermethrin exposure on Morphological changes in ***Channa gachua***

Sl. No.	No. Of Test fishes	Morphological Changes	Concentration In ml/l	Exposure period					
				Chronic test					
				1 days	5 days	10 days	15 days	20 days	30 days
1.	5	Patches on the body	Control	-	-	-	-	-	-
			0.5 ml/l	-	-	++	++	+++	++++
			1.0 ml/l	-	+	++	+++	+++	++++
			1.5 ml/l	+	++	+++	+++	++++	++++
			2.50ml/l	++	++	+++	+++	++++	++++
2.	5	Discoloration of body	Control	-	-	-	-	-	-
			0.5 ml/l	-	+	+	++	++	+++
			1.0 ml/l	-	+	++	++	+++	++++
			1.5 ml/l	+	+	++	+++	+++	++++
			2.0 ml/l	+	+	++	+++	+++	++++
3.	5	Shedding of scales	Control	-	-	-	-	-	-
			0.5 ml/l	-	-	-	++	++	+++
			1.0 ml/l	-	-	++	++	+++	+++
			1.5 ml/l	-	+	++	+++	+++	++++
			2.0 ml/l	-	++	++	+++	+++	++++
4.	5	Mucous secretion	Control	-	-	-	-	-	-
			0.5 ml/l	-	+	++	++	+++	+++
			1.0 ml/l	+	++	++	++	+++	++++
			1.5 ml/l	+	++	++	+++	+++	++++
			2.0 ml/l	++	++	+++	+++	++++	++++
5.	5	Bulging eye's	Control	-	-	-	-	-	-
			0.5 ml/l	-	++	+	++	+++	+++
			1.0 ml/l	+	++	++	+++	+++	+++
			1.5 ml/l	+	++	++	+++	++++	++++
			2.0 ml/l	++	++	++	+++	+++	++++
6.	5	Clumping of gills	Control	-	-	-	-	-	-
			0.5 ml/l	-	-	+	++	++	+++
			1.0 ml/l	-	+	++	++	+++	+++
			1.5 ml/l	+	++	++	+++	+++	++++
			2.0 ml/l	++	++	+++	+++	++++	++++

(-) Normal, (+) Nil, (++) Less change , (+++) Moderate & (++++) prominent change.

Fig: Discoloration of skin

Fig:Patches on body

Table 6 : Mortality of *Channa gachua* in different concentration of Malathion at 30 days exposure period

Concentration of Malathion (mg/l)	% Mortality
control	40%
0.5	80%
1	90%
1.5	100%
2	100%

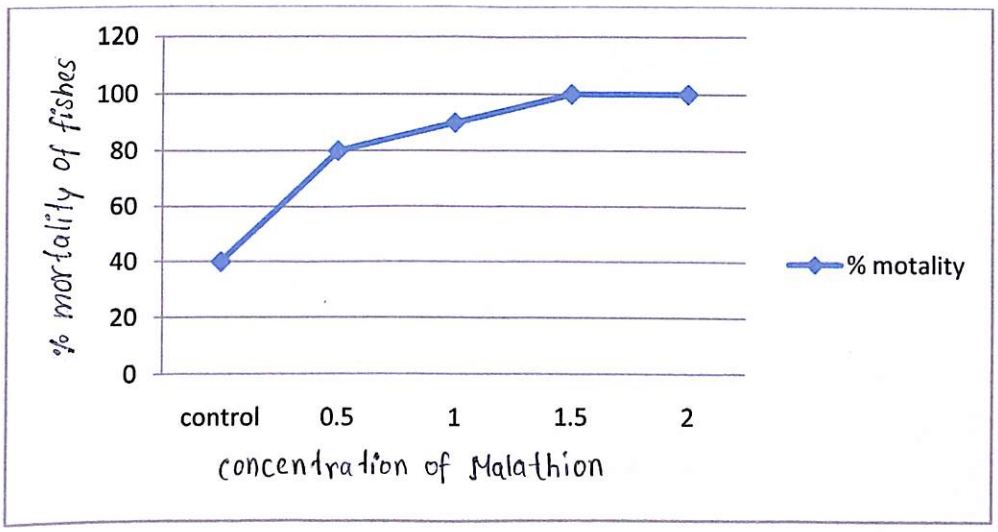


Fig : Line graph showing mortality of *Channa gachua* in different concentration of Malathion exposure

Table 7 : Mortality of *Channa gachua* in different concentration of Cypermethrin at 30 days exposure period.

Concentration of Cypermethrin (ml/l)	% mortality
Control	30
0.5	70
1	80
1.5	90
2	100

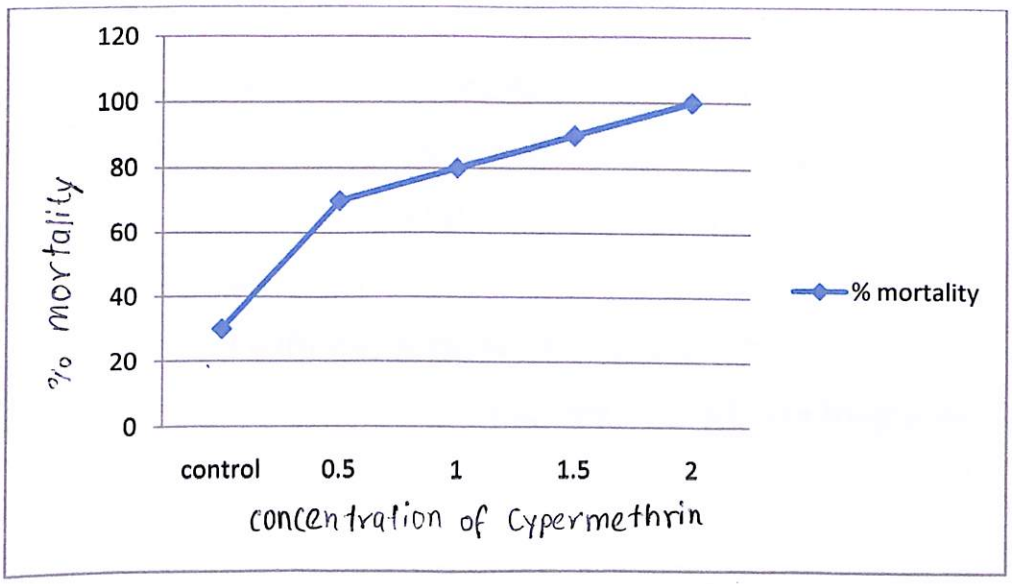


Fig : Line graph showing mortality of *Channa gachua* in different concentration of Cypermethrin exposure

DISCUSSION:-

Fishes are one of the vertebrates group that respond firstly when the environment is contaminated with pollutants (Singh et al. 2020). Fishes are an important indicator of water pollution. Presence of pesticides in surface and subsurface layer of aquatic system brings serious threat to the whole ecosystem. As, fishes are the part of important source of protein, and if human consume pesticide contaminated fishes, it may lead to infection or disease in the human body.

Behavioural changes:-

Behavior allows an organism to adjust to external and internal stimuli with the challenge of surviving in a changing world. Behavior is also the result of adaptations to environmental factors. Fish is extremely sensitive to behavior and any change in behavior of fishes is related with the toxicity. Behavioral changes in any animals are indicates the internal disturbances of the body functions.

In the present studies exposure to malathion and cypermethrin caused alterations in the behavior of *Channa gachua.* In both the control and the test media, the behavior and condition of fishes were observed during the whole experiment.

When fishes were exposed to sub lethal concentration of malathion and cypermethrin, they showed marked changes while in control group such changes were not reported.

Just after introducing the fishes to the test media, they showed the symptoms of swimming disability such as irregular, erratic and

swimming movements, restlessness, loss of equilibrium, hitting against the wall of transparent container and also trying to jump out to avoid the pesticide.

Loss of balance during experimental period was observed during this study. Fast movements, gulping of air and surfacing was also observed.

Behavioral changes were increased with the increase of amount pesticides. Sluggishness observed at the end of exposure period may be due to loss of energy as a result of erratic swimming, jumping and restlessness.

Morphological changes:-

During this study, were recorded the various morphological changes in the body being exposed to different time period and concentration of malathion and cypermethrin. The morphological changes in the fish indicate the toxicity of the pesticides.

The morphological changes included discoloration of skin, mucous secretion, removal of scales and patches on body etc. These changes were first shown by the fishes exposed to malathion.

Sedimentation of malathion on the body also observed during the experiment. These changes seems to appear after 10 days of exposure and were seems to be more prominent at 30 days.

The percentage of all these abnormality was increased with the increase of exposure period.

CONCLUSION:-

Pesticides are used worldwide in agriculture, public health operations and eradicating mosquitoes and other pests. The increased use of the insecticides and pesticides is not only dangerous to the fishes but also other aquatic animals and also to man and other animals through food chain. To develop our fish production, we should check the use of insecticides and pesticides in our country.

In the present study, results showed that when fishes exposed to sublethal concentration of malathion and cypermethrin, they showed a marked changes in their behavioral and morphological changes. These changes show direct response of the animals to the pollutants. Mortality was observed at a time interval. *Channa gachua* has shown differential mortality level at different concentration. The mortality has shown an increased level with an increase in the duration of the exposure period of malathion and cypermethrin.

This type of study helps to understand the effect of pesticides on fishes so as to determine safe environmental concentration where there is no stress and pollution to fishes.

This study also recommends proper inspection of pesticides toxicity before using it.

To save the fish population from different pesticides, we should realize the toxic effects of pesticides and establish reliable waste water treatment from agriculture and various sources.

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