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TOXIC EFFECT OF PYRACLOSTROBIN ON THE FRESHWATER FISH CTENOPHARYNGODON IDELLA

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ABSTRACT:

Pollution the water by harmful chemicals can leads to entry of pollutants into food chain. Water bodies play a major role in assimilating or carrying of industrial and municipal waste water, runoff from agricultural fields, roadways and street which are responsible for water pollution. In the present study influenced the amount of carbamate Pesticide Pyraclostrobin (20%) on freshwater fish *Ctenopharyngodon idella*. The fish were acclimatized to laboratory conditions for 7-10 days and then exposed to different lethal concentrations of pyraclostrobin (24, 48,72,96hrs of 7.2, 6.8, 6.4, 6.0mg/L) for 96hr along with the controls. Water parameters were also before down the experiment. The 96hr LC_{50} *Ctenopharyngodon idella* exposed of pyraclostrobin found in 6.0mg/Lt. The LC_{50} values were calculated in Finney Probit method. lower bond (5.6074) and the upper bond was in (6.3926). The Pyraclostrobin effect on fish showed uncontrolled behaviour like mucus secretion, decreased agility and inability to maintain normal position and erradic swimming were seen in exposure time of the carbamate toxicant. The fish were exposed to lethal and sublethal concentrations for 24hr, 4th and 8th days. After each exposure period, different tissues were taken to estimate the total protein levels.

Introduction :-

Modernization and intensification of agricultural production is generally accompanied by a rapid increase in the use of chemical fertilizers and pesticides. Pesticides are useful tools in agriculture, but the gradual degrading of aquatic ecosystem. In addition to agriculture practices together with pest control programmes the surface runoff and aerial spraying forming the major source for translocation pesticides into aquatic ecosystems (Joseph *et al.,* 2011). The improper management of pesticides in agriculture crops could result in contamination of water bodies (Candida Toni *et al.,* 2011; Capkin and Altinok, 2013). When pesticide reaches the aquatic environment, it may be present there for several days or weeks, depending upon its solubility, producing of mass mortality, morphological, physiological and behavioral changes in the organisms. Studies reveal that fluoride accumulates in bones and flesh of fish (2018). Studies suggest that dietary fishes are one of the major contributors in dental fluorosis in human beings(2016). h increasing concentration of fluoride ion in water, severity of effect of fluorite toxicity on fishes and other aquatic organisms increase. . It has been reported that butachlor is moderately toxic and irritating for R. rutilus caspicus and S. lucioperca (Mohammad & Hedayati,2017). At present increased food production to meet the energy requirements of the ever-increasing population, is a major problem of our country. To meet these requirements man has employed modern techniques both in agricultural and industrial production. But at the same time the population explosion possesses a great threat to human society**.** Ali S, *et al*.,2016**).** Studies reveal that fluoride accumulates in bones2 and flesh of fish3 significant source of fluoride**.** (Chowdhury *C et al*.,2018**).** Studies suggest that dietary fishes are one of the major contributors in dental fluorosis in human beings S, Yousuf A, Nagaraj A, *et al***.,** 2015) **.** With increasing concentration of fluoride ion in water, severity of effect of fluorite toxicity on fishes and other aquatic organisms increase. Tirumala Rao B, *et al*., (2017). Contamination in water is through pesticides and industrial effluents1 . Fluoride toxicity effects aquatic life adversely. Whereas, concentration of chloride and calcium ions decreases the severity of fluoride toxicity in fishes . Our studies on effects of fluoride exposure on Channa punctatus reveals that fluoride induces oxidative stress and causes significant toxic effect on fish liver.(Ghosh S, *et al*.,2018). Labeo rohita showed a decrease in total protein content in liver muscle, gill and kidney upon sublethal exposure to fluoride, Kale MD, Muley DV,2015).

Material and methods: -.

Experimental profile: - Healthy freshwater fish, Ctenopharyngodon idella size [6±7 cm total length (TL) and 6.5±7.5 g body weight] were collected from the fish farm, Kuchipudi, Guntur District of A.P, India. Then the fish was acclimatized to the laboratory conditions with sufficient dechlorinated water for 4 to 15 days at room temperature 28±20C. The fish were fed with meal fish, rice and commercial fish pellets once in two days, at the same time water was renewed every day rich in oxygen (aeration) and feeding was stopped one day prior to the experimentation. The test compound for the study belonged to Carbamate pesticide Pyraclostrobin (20%WG) widely used in agriculture field. Pyraclostrobin is a broad-spectrum pesticide used to control insects, pest, and mites in agricultural field. Concentration of Pyraclostrobin was found in 96hr 6.0 mg/lt as a lethal and 1/10th was taken as a sublethal concentration along with control

group was maintained for each experiment. Where maximum 10 fish were used per each concentration of the test toxicant, 10 fish were also maintained in separate container along with experimental group, served as control.

EFFECTS OF PYRACLOSTROBIN TOXICITY ON FISH HEALTH

The pesticide pollution is one of the major pollutions in aquatic field which effect the aquatic animals. The carbamate Pyraclostrobin (20% WG) pesticide exposed for 96hr the value was found in 6.0 mg/l and the sublethal was taken where as in 1/10th of the lethal. The 96hr LC_{50} values with 95% confidence limits for Pyraclostrobin upper bond (1.4497) and the lower bond (1.3503) was shown in Table.5. Allium sativum, commonly known as garlic, is a member of the family Alliaceae. (2015). History of dietary and medicinal applications for curing various diseases. (2014). The use of garlic in aquaculture has been reported to promote growth, stimulate appetite, provide a tonic to improve the immune system, provide antistress protection and can be a proven prophylactic and therapeutic agent. (2015). The percent mortality of the freshwater fish Labeo rohita in different concentrations was shown in table. the probit mortality of the fish in different concentrations along with log concentration was shown in table , under the toxicity evaluation. Decreased swimming behaviour and increased respiration rate were other effects of pesticide in the present study, found that contaminates such as pesticides disturb normal fish behaviours after exposure. The physiological and biochemical alterations observed in an animal under any physiological stress can be correlated with the structural and functional changes of cellular proteins. Among the non-essential metals, arsenic, mercury, cadmium, lead and silver, poses serious threat. (Vanitha, A., *et al.*, 2017). The investigations on pretilachlor have focused on removal of pretilachlor from the environment and the effect of pretilachlor on soil microorganisms (Saha *et al.*, 2012; Toan *et al.*, 2013). reported that the toxicity of pretilachlor on Gambusia increases with increasing concentration and exposure time. (Ali,S. Mohamd, R.I.,2013). The exposed fish Ctenopharyngodon idella showed irregular, erratic, and darting swimming movements, rapid opercular movements, gulping of air at the surface, hyperexcitability, increased mucus secretion, loss of equilibrium and hitting to the sidewalls of the test tank and finally sinking to the bottom. (Rajeswari *et al.*, 2020).

Discussion: -

Present investigation the inherent capacity to test fish *Ctenopharynogodon idella* effct of Pyraclostrobin methods to evolute the taxicity of the toxicity by the determination of lethal concentration (LC), which represents the amount of chemical require for the death of 50% population amoung the experimental Fish. The value is noted and calculated according to Probit Analysis method and LC_{50} Values were determined. In the present study Pyraclostrobin caused 50 % mortality to *Ctenopharyngidon idella* for 96hrs at 6.0 mg/l. the pesticide effect of fish change behaviour like jurking movement, loss of equilibrium sluggishness, mucus secretion, spiral movement, remove scales and container turned in colour. Opening of mouth gasping. I obesrved similar to other reseachers, Pesticides can produce adverse effects in a biological system, seriously damaging its structure and function of living system finally leads to death of organism. Pesticides are entering into aquatic ecosystem by agriculture runoff from land, impairing the quality of the water and making it unfavourable for aquatic life (Tilak et al., 2009). The evaluation of fish physiological status (Ogundiran, 2007; Adewoye, 2010 and Yekeen and Fawole, 2011). The present investigation was revealed that LC_{50} value in 96hr lethal concentration found in static renewal method. The sublethal

concentration was taken as 1/10th of the lethal concentration. The pesticide effect influences the fish behaviour like jerking movements and mucus section were observed. When pesticide concentration was increasing the mortality of the fish was increased. (Nwani *et al.*, (2013) reported that median lethal concentration of chlorpyrifos based pesticide Termifos to African catfish *clarias gariepinus* were found to be 0.861 mg/Lt. The toxicity of a pesticide could vary from species and this variation is due to differential tolerance of animals to pesticide exposure (Prabhupatkar, 2004; Ram Nayan Singh, 2013). *Labeo rohita* showed a decrease in total protein content in liver muscle, gill and kidney upon sublethal exposure to fluoride ,2015). Our studies on effects of fluoride exposure on *Channa punctatus* reveals that fluoride induces oxidative stress and causes significant toxic effect on fish liver (2018). Often water from fields where fertilizers containing fluoride 2018. induce different types of toxicity in fish such as changes in fish behaviour and biochemical changes in antioxidant defence system. (Neelanjana Choudhurya,. Jayanta Tarafdar *et al.*,2017). Lamda cyhalothrin highly toxic to fishes because it is strongly absorbed by the gills even at very low concentration in water due to its high lipophilicity. (Dhruv Kumar and Mamta Kumari,.2019). Observation of blood parameters allows the most rapid detection of changes in fish after the exposure to xenobiotics. (Ogundiran M.A. and Fawole O.O.2018). when the fish was exposed to mercury, erratic swimming, abnormal posture, dis-balance, sluggishness, imbalance in posture, increase in surface activity, opercular movement, gradual loss of equilibrium and spreading of excess of mucus all over the surface of the body were observed. (N.Vasanthi, *et al* 2019). These huge benefits need to sustained by preventing human activities that have potential of releasing toxic chemicals and environmental stressors into aquatic bodies in order to avoid the deterioration of the freshwater quality. (Patrick Amoatey *et al.*, 2019). Villa *et al.* (2018) studied the behavioural effects of pharmaceuticals on the larvae of Diamesa zernyi, a wild alpine species in Rio Present, Italy. assessed the endocrine disrupting effects of the embryo of zebrafish Danio rerio through metabolomics analysis. (Ortiz-Villanueva *et al.* (2018). The results showed that the illicit drug and its metabolites caused alterations on the protein profile of the fish including the modulation of several proteins of different functional classes. (Parolini *et al.* (2018). Quintaneiro, et al. (2018) studied the effects of linuron and S-metolachlor herbicides on Perez's frog Pelophylax perezi embryo for about 192 hr of both herbicides. Stara *et al.* (2018). conducted a study on the biochemical and histological effects of sub-chronic exposure to atrazine in crayfish Cherax destructor. The study results found decreased in the molluscicide impacts and number of haemocytes following the exposure of the snail to the insecticide. (Ibrahim, Ahmed., 2018). Pollutants like insecticides have the capacity to penetrate into the RBC (Yekeen and Fawole, 2011). The effect of carbamate, fish showed uncontrolled behaviour like mucus secretion, decreased agility and inability to maintain normal position and erratic swimming were seen in exposure time of the carbamate toxicant. (A. Anitha and V. Venkata Rathnamma., 2016). The toxicant accumulated is further increased by biomagnification of the synthetic pesticides from water by aquatic organisms. heavy use of synthetic pesticides results in a lethal effect on various non-target organisms in the aquatic environment and direct or indirect effect to users. (Sathyamoorthi A, *et al.*, ,2020). The high toxicity of synthetic pesticides has been found to aquatic, zooplankton, and mammalian species. Mossa ATH, Swelam ., 2015). According Dey C, Saha et al, Dimethoate and Lambda-cyhalothrin showed to be lethal for *Labeo rohita*. (2014). effects of synthetic pyrethroid insecticides on several toxicological end-points in fish is provided. toxic effects of pyrethroids to biomarkers in different fish species were studied. **(**Ullah S, Li Z, Zuberi A *et al*., 2019). Bantu N, *et al.*, 2018, The variations in the lethal and sub-lethal concentration values depend upon various factors viz. sensitivity to the toxicant, its concentration, and duration of exposure. According to the pyrethroid pesticide λ-cyhalothrin is highly toxic to fishes as it is strongly absorbed by the gills even at very low concentrations in water due to its high lipophilicity **(**Kumar D and Kumari M.,2019. Nirmala K, *et al*., 2016**..** According to Gadhave PD, Brar, 2014. the non-essential metals, arsenic, mercury, cadmium, lead and silver, poses serious threat. (Vanitha, A. *et al.*,2017). The fish maintained in freshwater behaved normal as usual. But when the fish was exposed to mercury, erratic swimming, abnormal posture, dis-balance, sluggishness, imbalance in posture, increase in surface activity, opercular movement, gradual loss of equilibrium and spreading of excess of mucus all over the surface of the body were observed Increased opercular movements were seen in the fish, *O. mossambicus* exposed to mercury, which was in accordance to the report put forth by Amitakiran and Jha in *Clarias batrachus* exposed to herbicide, herboclin. (Amita Krian and A.K. Jha.2009). This toxicity test on the effect of mercury on O. mossambicus offers a rapid method for assessing the heavy metal impact on this fish.

(Vasanthi *et al.,* 2019**)**. The results clearly indicated that exposure of fishes to pretilachlor based herbicide resulted in increased mortality with increasing concentration of the herbicide , (Rakesh Soni, and Sushant Kumar Verma∗(2018). It has been reported that butachlor is moderately toxic and irritating for *R. rutilus caspicus* and *S. lucioperca* **.** Additionally, the toxic impact of pesticides can also be reversed as suggested by Rezakhalatbary et al.2016 where the olive oil can ameliorate the deltamethrin induced nephrotoxicity in albino mouse.The LC50 of *triazophos* for 96 h treatment period in different fish species are as follows: 0.008 mg/L for Peseudorasobora parva, 0.035 mg/L for *Tilapia nilotica*, 0.060 mg/L for *Colossoma* brachypomum1.05 mg /l for *Cirrhinus mrigala.* (T. Karatas, 2016). LC_{50} is the ambient aqueous chemical activity that causes 50% mortality in an exposed population. These calculations are based on two important assumptions. The first assumption is that the exposure time associated with the specified LC_{50} is sufficient to allow almost complete chemical equilibration between the fish and the water. The second assumption is that the specified LC_{50} is the minimum LC_{50} that kills the fish during the associated exposure interval. Fortunately, most reliable LC_{50} satisfy these two assumptions OECD guidelines 220 advocates that this should be the method normally applied to determine LC_{50}. Toxicity are relative property of a chemicals l which discusses to its potential to injurious to health on living organisms. (Anitha smruthi Ch.et al.,2018). The exposure duration surges median lethal concentrations (LC50) decline.(Rakesh Soni, and Sushant Kumar Verma *et al.,*2018)

Toxicity Evaluation:

Table: 4 Observed percent mortality, probit mortality and LC_{50} value of the fish *Ctenopharyngodon idella* exposed to **Pyraclostrobin (20% WG)** for static 96 hrs.

S. NO	DOSE IN PPM	MORTALITY (%)	PROBIT VALUE (Y)	LOG (100* DOSE) =X	X*X	X*Y	Y*Y
1	5.6	10	3.72	2.748188027	7.552537432	10.22325946	13.8384
2	5.8	30	4.48	2.763427994	7.636534278	12.38015741	20.0704
3	6.0	50	5.00	2.77815125	7.718124368	13.89075625	25
4	6.2	70	5.52	2.79239169	7.79745135	15.41400213	30.4704
5	6.4	90	6.28	2.806179974	7.874646046	17.62281024	39.4384
			25	13.88833894	38.57929347	69.53098549	128.8176

X	2.777667788
Y	5
SXX	2.10177
SYY	3.8176
SXY	0.08929079
SLOPE B	1.802287684
VARIANCE B	0.047578945
VARIANCE A	0.467092416
M	2.777667788
Anti Log	5.9933

LC_{50}	6.0 mg/lit
R^2 Value	0.9936

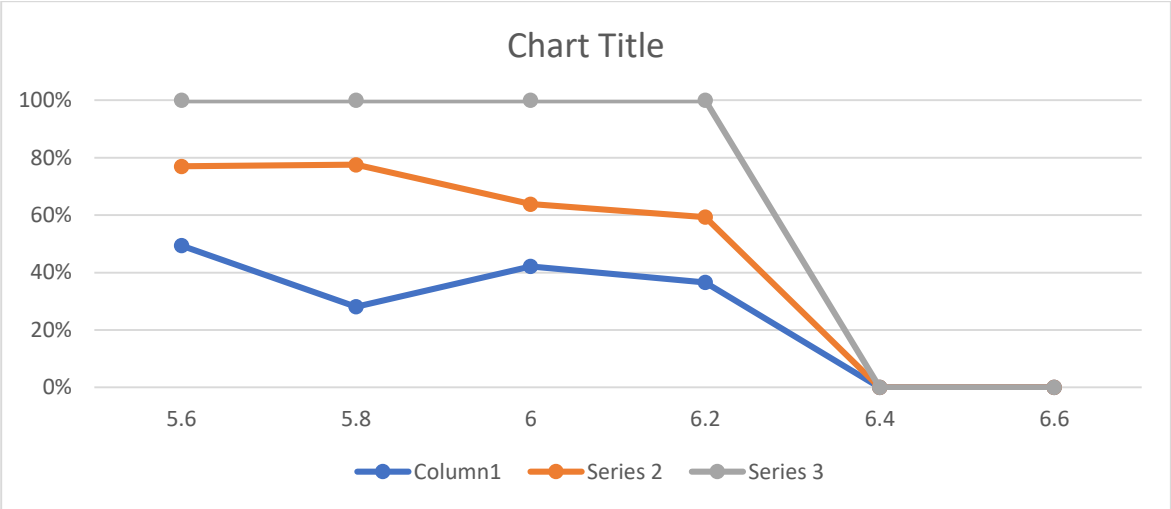

Table: 5 Confidence Levels of 95% for exposed to **Pyraclostrobin (20% WG)** fish *Ctenopharyngodon idella* at different exposed periods.

Hours of exposure(hrs)	Lower and Upper confidence limits
24	6.8074 ± 7.5926
48	6.4074 ± 7.1926
72	6.0074 ±6.7926
96	5.6074 ± 6.392

Table: 6 Regression values for toxicant exposed to **Pyrachlostrobin (20% WG)** fish *Ctenopharyngodon idella* at different exposed periods.

S.NO	Exposure time in hrs.	LC_{50} in % Concentration	No. of fish exposed	% Mortality	Regression equation Y=(y-bx) +bx
1	24	7.2	10	50	Y= 51.00*X - 140.7 R^2=0.9937
2	48	6.8	10	50	Y=48.16*X - 131.4 R^2=0.9937
3	72	6.4	10	50	Y=45.32*X - 122.2 R^2=0.9937
4	96	6.0	10	50	Y=42.48*X - 113.0 R^2=0.9936

Conclusion:

When the effect of pesticide, fish show behavioural changes were observed. The pesticide concentration was increased and fish living survivability were gradually decreased. The LC50 can be used as a relative measure to study the impact of the pyraclostrobin 20%(WG) concentration on test fishes at different intervals. This toxicity test on the effect of pyraclostrobin on Ctenopharyngodon Idella a rapid method for assessing the pyraclostrobin impact on this fish. This type of preliminary investigations can be useful for deriving the safe level of Pyraclostrobin concentration that can be released into the aquatic environments.

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