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Effect of Organophosphate Pesticides Dichlorvos on Excretory Function of the Channel Cat Fish, Heteropneustes Fossilis (Bloch)

Sanjoy Deka[1], Rita Mahanta[2]
[1,2]*Dept. of Zoology, Cotton College, Gauhati University,*
Gopinath Bordoloi Nagar, Guwahati - 781014, Assam, India.

Abstract: After the ban on Organochlorine pesticide during 1970s, organophosphate pesticide was introduced in the agricultural practices owing to its rapid biodegradability and less persistence properties in nature. Present investigation is aimed to study the 'Effect of organophosphate pesticide dichlorvos on excretory function of the channel catfish, Heteropneustes fossilis (Bloch)'. A sub-lethal concentration of 2.5 ppm and 5 ppm dichlorvos were prepared and used in this investigation process. In this study, acclimatized fishes were divided into three groups, a control group (Group 1) and two experimental groups (Group 2). Experimental group 2.1 and 2.2 were treated with sub lethal doses of the pesticides dichlorvos whereas control group was being freed from the treatment of the pesticide. Level of Ammonia and Urea in Serum and GLDH and Arginase activity in liver tissues were estimated at the interval of 10, 20 and 30 days in the fishes under investigation. A dose and duration dependent alteration was observed in the level of serum ammonia, serum urea, serum creatinine, and GLDH activity in the liver tissue among the experimental group of fishes exposed to dichlorvos.
Key words: Ammonia; Urea; Creatinine; GLDH; Dichlorvos.

I. INTRODUCTION

Pesticide as defined by United Nations Environment Programme is any substance or mixture of substances intended for preventing, destroying, repelling or mitigating any pest. Amongst others, organophosphorus pesticides (OPs) are the most commonly used pesticides in the world due to their rapid degradation ability. Unfortunately, OPs harm beyond their target and can contribute severe, long term population effects on terrestrial and aquatic non-target species, predominantly vertebrates[1].

Of all pesticides, the organophosphates have the highest level of toxicity in vertebrates and are all derived from phosphoric acid[2]. The main mechanism by which the organophosphates exert a toxic effect is the inhibition of cholinesterases (ChEs), an important group of enzymes of the nervous system of both vertebrates and invertebrates[2, 3]. Organophosphates are used on a large scale in the agricultural sector to control invertebrate pests associated with fruit and cotton production.

The hazardous pesticide toxicants have been known to accumulate in tissues of fishes and other edible organisms have a chance to reach the predators like birds and man through food chain. Food organisms may be impacted either directly by the toxic effects of pesticides or marked as unsuitable for human consumption due to elevated level of these substances[4]. The harmful effects of pesticides on aquatic organisms are due to the indiscriminate use, careless handling, accidental spillage or discharge of untreated effluents into natural water ways.

Pesticides are also well known for causing more toxic effects in teleost fishes[5]. The magnitude of pesticide pollution was studied in the Indian fishes by various workers[6,7,8,9,10,11,12,13,14,15,16].

Dichlorvos (DDVP) is an insecticide of the organophosphate (OP) group. It has been in use since about 1955 and is used in United Kingdom both professionally and in homes and gardens in a number of areas: in agriculture and horticulture[16]. Even though dichlorvos is reported to be a contact and stomach insecticide for variety of crop pests, it is also toxic to fish and other aquatic organisms[17].

Present investigation is aimed to carry out an empirical study on the effect of organophosphate pesticides dichlorvos on excretory function of the channel cat fish *Heteropneustes fossilis* (Bloch).

II. MATERIALS AND METHODS

A. Specimen

Healthy and sexually mature specimen of *Heteropneustes fossilis* of equal size group (12 ± 3 cm) and average weight (12 to 15 gm) are procured from the local market and the fishes were kept for 15 days in glass aquarium containing 80 litres of fresh water in the

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laboratory at about water temperature 25 ± 3º c for acclimatization. Fishes are starved for 24 hours prior to the experiment and are not fed during the period of experiment[6].

B. Pesticide

The organophosphorus pesticide dichlorvos (76% E.C) was procured from the local market for present investigation purpose.

C. Lc_{50} Calculation

A pilot experiment was done to find out the LC_{50} value of dichlorvos by probit analysis [18] and LC_{50} for 96 hours is found to be 19 ppm for dichlorvos. Sub-lethal concentrations of 2.5 ppm and 5 ppm for dichlorvos were prepared by using standard technique[19].

D. Design of the Experiment

In this experiment, the specimens were kept mainly in two groups namely Control Group (Group I), Experimental Group (Group 2) as follows-

Investigation group	Sub group	Sub-lethal dose
Control Group	Nil	Free from the treatment of dichlorvos and malathion
Experimental Group 2	Group 2.1	Treated with sub-lethal concentration of 2.5 ppm dichlorvos
	Group 2.2	Treated with sub-lethal concentration of 5 ppm dichlorvos

E. Collection of Sample

Blood were collected from the fishes of both the groups and serum was separated by centrifugation technique. Tissue homogenate was prepared from the collected liver tissue and was centrifuged based on the protocol of the individual experiments.

F. Methods of Estimation of Biochemical Parameters

Ammonia in serum was estimated by the method of Anken and Schiphorst[20] at the wavelength of 340 nm in spectrophotometer. Urea in Serum was estimated through Modified Berthelot Method by Fawcett and Scott[21]. GLDH activity in liver tissue was determined spectrophotometrically by adoption of the method of Doherty[22]. Arginase activity in liver tissue was estimated by the method of March et al[23]. and Creatinine was assayed spectrophotometrically using modified Jaffe Method[24].

G. Duration of Treatment

The investigation parameters were studied in control and experimental groups after 10, 20 and 30 days of experimental period.

III. RESULTS

A. Serum Ammonia

Changes in the mean ± SD values of serum ammonia in the *Heteropneustes fossilis* are shown in the table-1 and the percentage deviations of different experimental groups from the mean values of control group are presented in the fig-1. Serum ammonia was observed to be in decreasing trend significantly ($p< 0.01$) at sub-lethal dose of 2.5 ppm and 5 ppm dichlorvos from 10th day onwards to 30th day in comparison to control group of fishes.

Table 1: Presenting the mean ± SD values of serum ammonia (mg/dl) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

Investigation Group of fish n=20		Sub-lethal Dose of Pesticide	Mean ± SD Values		
			10 days	20 days	30 days
Control Group	Group I	Nil	5.29±0.03	5.64±0.02	5.46±0.04
Experimental Group 2	Group 2.1	2.5 ppm dichlorvos	2.71±0.03	2.45±0.03	2.01±0.04
	Group 2.2	5 ppm dichlorvos	1.85±0.03	1.45±0.05	1.44±0.03

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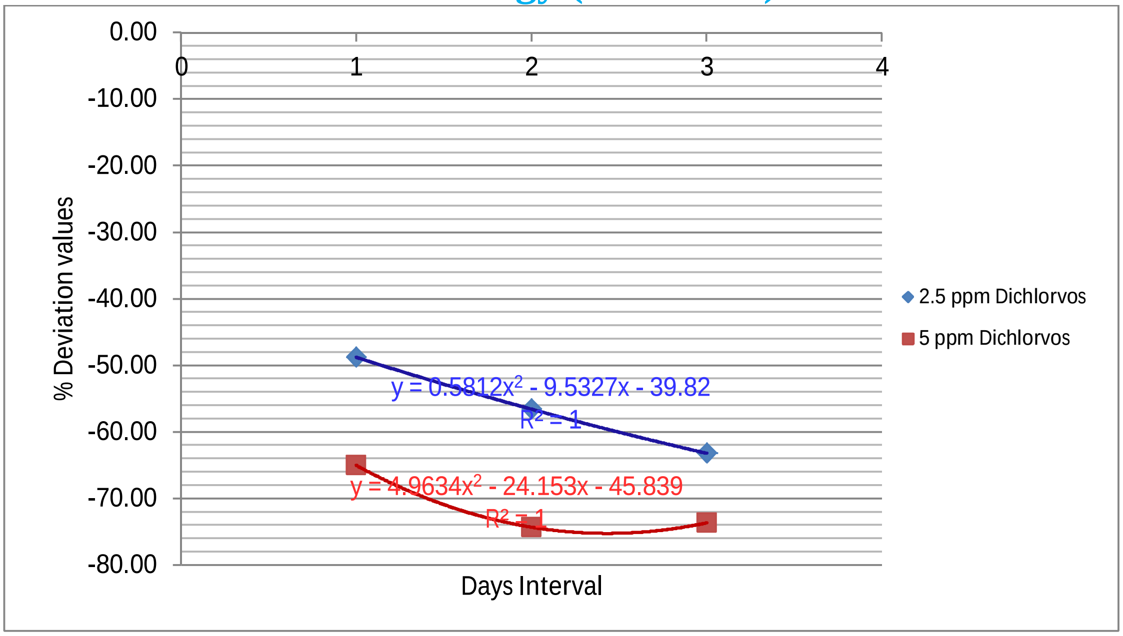

Figure 1: Presenting the % deviation values of serum ammonia (mg/dl) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

B. Serum Urea

Alterations in the mean ± SD values of Serum urea in the *Heteropneustes fossilis* are shown in the table 2 and the percentage deviations of different experimental groups from the mean values of control group are presented in the fig-2. A significant ($p< 0.01$) increase in the level of serum urea was recorded at sub-lethal dose of 2.5 ppm and 5 ppm for dichlorvos throughout the experimental period of 30 days in comparison to the control group of fishes. It was observed that level of urea was higher in the fishes exposed to 5 ppm dichlorvos than 2.5 ppm dichlorvos treated fishes.

Table 2: Presenting the mean ± SD values of Serum urea (mg/dl) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

Investigation Group of fish n=20		Sub-lethal Dose of Pesticide	Mean ± SD Values		
			10 days	20 days	30 days
Control Group	Group I	Nil	23.97±1.09	24±1.08	33.87±2.1
Experimental Group 2	Group 2.1	2.5 ppm dichlorvos	42.56±2.65	42.25±2.7	42.95±2.45
	Group 2.2	5 ppm dichlorvos	44.01±3.08	43.95±3.04	44.21±3.09

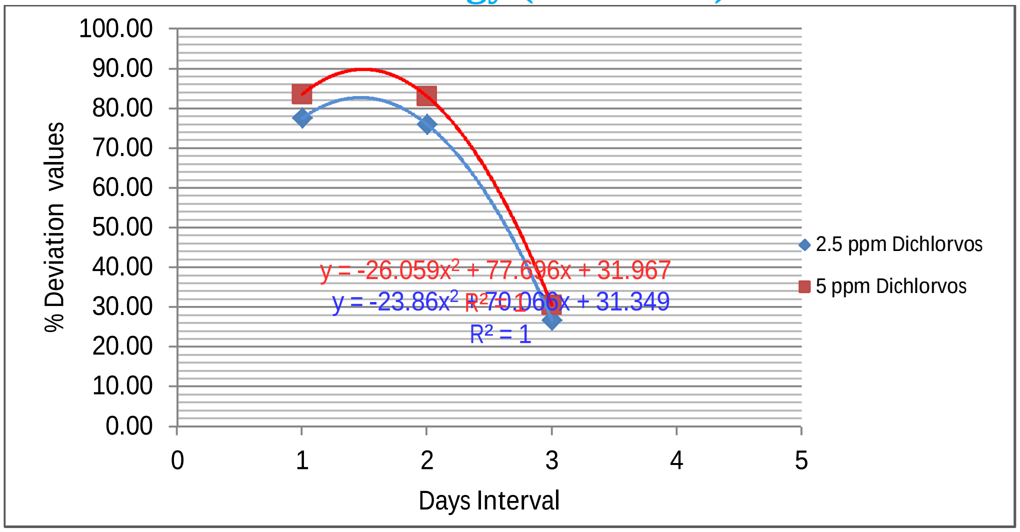

Figure 2: Presenting the % deviation values of Serum urea (mg/dl) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

C. Creatinine

Alterations in the mean ± SD values of creatinine in the *Heteropneustes fossilis* are shown in the table 3 and the percentage deviations of different experimental groups from the mean values of control group are presented in the fig-3. A significant increasing ($p < 0.01$) trend in the mean ± SD values serum creatinine was observed in the group of fishes administered with sub-lethal dose of dichlorvos from 10th day onwards and reached maximum value on 30th day compared to control group of fishes.

Table 3: Presenting the mean ± SD values of serum Creatinine (mg/dl) in control and experimental groups of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

Investigation Group of fish n=20		Sub-lethal Dose of Pesticide	Mean ± SD Values		
			10 days	20 days	30 days
Control Group	Group I	Nil	0.67±0.08	0.69±0.08	0.64±0.08
Experimental Group 2	Group 2.1	2.5 ppm dichlorvos	0.91±0.02	1.55±0.12	1.89±0.06
	Group 2.2	5 ppm dichlorvos	1.05±0.06	1.70±0.12	2.1±0.08

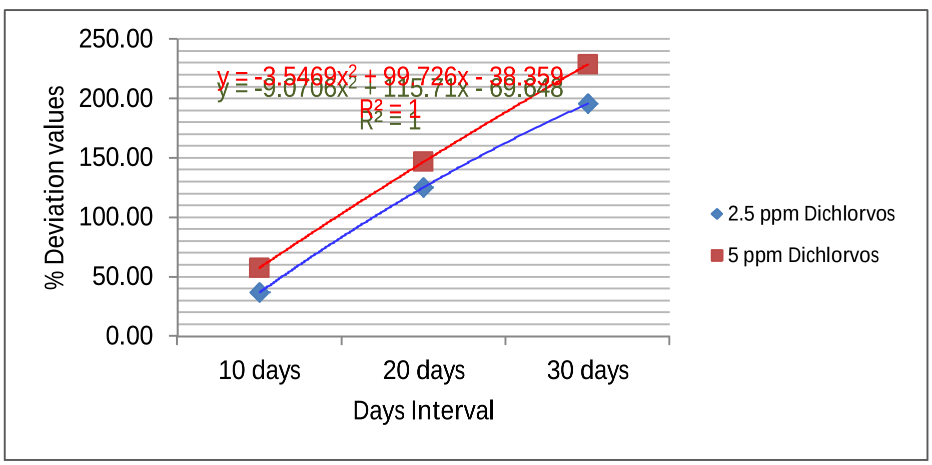

Figure 3: Presenting the % deviation of serum Creatinine (mg/dl) in control and experimental groups of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

D. Glutamate Dehydrogenase (GLDH) Activity

Variations in the mean ± SD values of GLDH activity in the *Heteropneustes fossilis* are shown in the table 4 and the percentage deviations of different experimental groups from the mean values of control group are presented in the fig-4. GLDH activity in liver tissue of the fishes showed a significant increase (p< 0.01) at sub-lethal dose of 2.5 ppm Dichlorvos compared to control group of fishes and was observed to be maximum on 20th day of the treatment. A significant (p< 0.01) increasing trend in the mean ± SD values of GLDH activity was noticed in the fishes of experimental group treated with sub-lethal concentration of 5 ppm dichlorvos from 10th day onwards up to 30th day with maximum level of GLDH activity on 30th day compared to any other investigating group.

Table 4: Presenting the mean ± SD values of GLDH activity (U/mg) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos.

Investigation Group of fish n=20		Sub-lethal Dose of Pesticide	Mean ± SD Values		
			10 days	20 days	30 days
Control Group	Group I	Nil	1.2±0.05	1.23±0.02	1.24±0.02
Experimental Group 2	Group 2.1	2.5 ppm dichlorvos	1.33±0.06	1.56±0.06	1.41±0.05
	Group 2.2	5 ppm dichlorvos	1.31±0.03	1.39±0.04	1.65±0.05

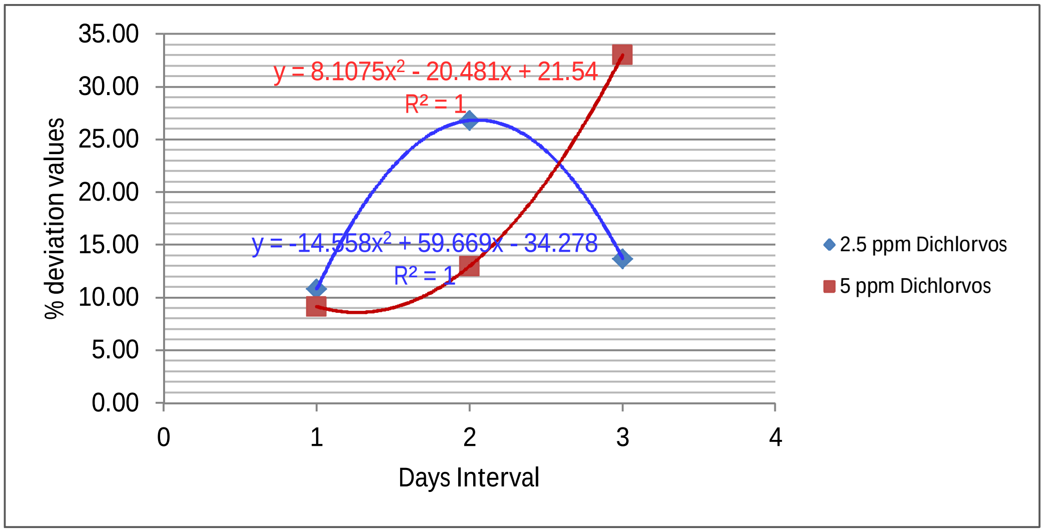

Figure 4: Presenting the % deviation values of GLDH activity (U/mg) in control and experimental group of Heteropneustes fossilis (Bloch) treated with sub-lethal dose of dichlorvos

IV. DISCUSSION

The freshwater fish, *Heteropneustes fossilis* prefer to live in aquatic muddy habitat and are very resilient in nature. Present investigation finds a significant (p< 0.01) decrease in the level of serum ammonia in the fishes exposed to dichlorvos. On the contrary, a significant increase (p< 0.01) in the level of serum urea, serum creatinine and GLDH activity was observed in the *Heteropneustes fossilis.* According to Campbell (1991), fishes, like any other animals cannot store excess amino acids. As a result, excess amount of dietary amino acids after the assimilation of the amounts needed for growth and maintenance of protein turnover are preferably degraded in the liver[25]. Decrease in ammonia in serum and increase in urea and GLDH activity in the present study supports the earlier findings which are indicative of activation of a second mechanism of ammonia detoxification, that is, ureogenesis by the fishes under pesticide-induced stress[26]. This ureogenesis may be due to sufficient production of ammonia in mitochondrial matrix which could not permeate mitochondria, instead, formed carbamoyl phosphate and entered ornithine cycle to liberate urea in liver. GLDH activity also increases with hepatocellular damage[27].

In teleost fish, ammonia excretion accounts for 55 to 80 % of total nitrogen excretion, while urea excretion is about 6 to 8 %[28]. Common cause for increase in blood urea is atypical nitrogen excretion usually due to kidney damage and urinary obstruction[29].

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However, under some circumstances as stress or enhanced ammonia level in the surrounding, fishes are reported to change their nitrogen excretion mechanism by forming urea as the end product for nitrogen excretion. Increased urea in the experimental fish was also reported to be due to the inability of the damaged kidney to filter the urea up to the normal levels[1,30].

Methyl guanidine-acetic acid or commonly called Creatine is an endogenously formed molecule that is stored largely in skeletal muscle, in both free creatine and *phosphocreatine* or *creatine phosphate (*phosphorylated form). Fish muscles have higher creatine contents than mammalian muscles[31]. Creatinine is solely considered a waste product and is then diffuses into the bloodstream from the muscle and enters to the renal parenchyma where it is filtered by the glomerulus and excreted in the urine. Creatinine is found to be high in blood when it is not reabsorbed by the renal tubules. Since its daily production is directly proportional to the creatine content of the body, the concentration of Serum Creatinine is more or less constant than that of any of the common excretory products. The constancy of creatinine formation and excretion makes creatinine a useful index of renal function, primarily glomerular filtration[32]. In this study, Creatinine was found to increase significantly in *H. fossilis* exposed to different sub-lethal dose of dichlorvos, which may indicate glomerular insufficiency and cellular damage and also to some changes in the muscular activity. Increased level of creatinine also reveals the malfunctioning of kidney under stress after pesticide exposure[33].

V. CONCLUSION

A significant ($p< 0.01$) decrease in the level of serum ammonia in *Heteropneustes fossilis* (Bloch) exposed to sub-lethal doses of dichlorvos were observed in the present investigation. On the other hand, a significant increase ($p< 0.01$) in the level of serum urea, serum creatinine and GLDH activity were observed in *Heteropneustes fossilis* (Bloch) administered with sub-lethal doses of dichlorvos.

REFERENCES

[1] Deka S and Mahanta R. Effect of Dichlorvos on hepato-renal functions of Indian catfish Heteropneustes fossilis (Bloch). International Journal of Advanced Research. 2015; 3 (3): 1112-1119.

[2] Ware GW. The Pesticide Book. WH Freeman and Company, USA. 1978; pp 28-52.

[3] Walker CH, Thompson HM. Phylogenetic distribution of cholinesterases and related esterases. In: Mineau P, ed., Cholinesterase-inhibiting insecticides. Their impact on Wildlife and the Environment. Chemicals in Agriculture Vol. 2. Elsevier Press, Amsterdam. 1991; pp 1-16.

[4] David W and Marius. Detoxification of accumulated trace metals by the American Oyster Crassostrea virginica: laboratory Vs environment physiological mechanisms of marine pollutant toxicity. Academic Press.1982: 89-107.

[5] Muthukumaravel K, Rajaraman P, Nathiya N, & Govindarajan. Studies on the Histopathology of Selected Organs of Freshwater Fish Labeo rohita. International Journal of Recent Scientific Research. 2013; 4 (11): 1728- 1735.

[6] Dalela RC, Bhatnagar MC, Tyagi AK and Verma SR. Histological damage of gills in Channa gachua after acute and subacute exposure to endosulfan and Rogor. Mikroskopie (Vienna). 1979; **35**: 301-307.

[7] Dubale MS and Shah P. Toxic effect of Malathion on the Kidney of a Freshwater teleost Channa gachua. Comp. Physiol. Ecol. 1984; 9(3): 238-244.

[8] Shukla, Lata, and Pandey AK. Restitution of thyroid activity in the DDT exposed Sarotherodon massambicus: A histological and histochemical profile. Water, Air, & Soil Pollution. 1980; 27: 225-236.

[9] Rashatwar and Ilyas. Effect of Phosphamidon in a Freshwater Teleost Fish Nemachelius denisonii (Day)- Histopathological and Biochemical Studies. J.Environ.Biol. 1984; 5(1): 1-8.

[10] Sadhu AK, and Mukhopadhyay PK. Comparative Effect of Two Pesticides Malathion and Carbofuran on Testes of Clarias batrachus (Linn.). J.Environ.Biol. 1985; 6(3): 217-222.

[11] Ghosh and Chatterjee. Disruption of pituitary-ovarian axis by carbofuran in catfish, Heteropneustes fossilis (Bloch). Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology**.** 1989; 129(3): 265-273.

[12] Dutta HM, Munshi JSD, Roy PK, Singh NK and Richmonds CR. Variation in toxicity of malathion to air and water-breathing teleosts. Bulletin Environmental Contamination and Toxicology. 1992; 49: 279-284.

[13] Medda C, Bhattacharyya B, Sarkar SK, Ganguly S, Basu TK. Effect of rotenone on activity of some enzymes on freshwater fish. Bull. Environ. Contam.Toxicol. 1995; 16(1): 55-60.

[14] Shukla S, Dass SK, Saksena DN. Effect of Sublethal Exposure of an Organophosphorus Pesticide, Nuvan on liver of catfish, Clarias batrachus. Nature Env Polln Techno. 2005; 4(3): 447–452.

[15] Velmurugan B, Selvanayagam M, Cengiz EI, Unlu E. Histopathological changes in the gill and liver tissues of freshwater fish, Cirrhinus mrigala exposed to dichlorvos. Braz Arch Biol Technol. 2009; 52(5): 1291-1296.

[16] Lakshmanan SA, Rajendran C, Sivasubramaniyan. Impact of dichlorvos on tissue glycogen and protein content in freshwater fingerlings, Oreochromis mossambicus (Peters). Int J Res Environ Sci Technol. 2013; 3(1): 19-25.

[17] Suntio LR. Shiu WY, Glotfelty D, MacKay D, Seiber JN. Critical review of Henry's Law constants for pesticides. Rev Environ Contam Toxicol. 1988; 103: 1-59.

[18] Finney DJ. Probit analysis. Cambridge University Press, London. 1964; 333-360.

[19] American Public Health Association. Standard Methods for the Examination of Water and Wastewater. 16th ed. / Washington, DC: American Public Health Association, 1985.

[20] Anken HC, Schiphorst ME. Kinetic determination of ammonia in plasma. Clin. Chim. Acta. 1974; 56: 151-157. DOI: 10.1016/0009-8981(74)90223-X.

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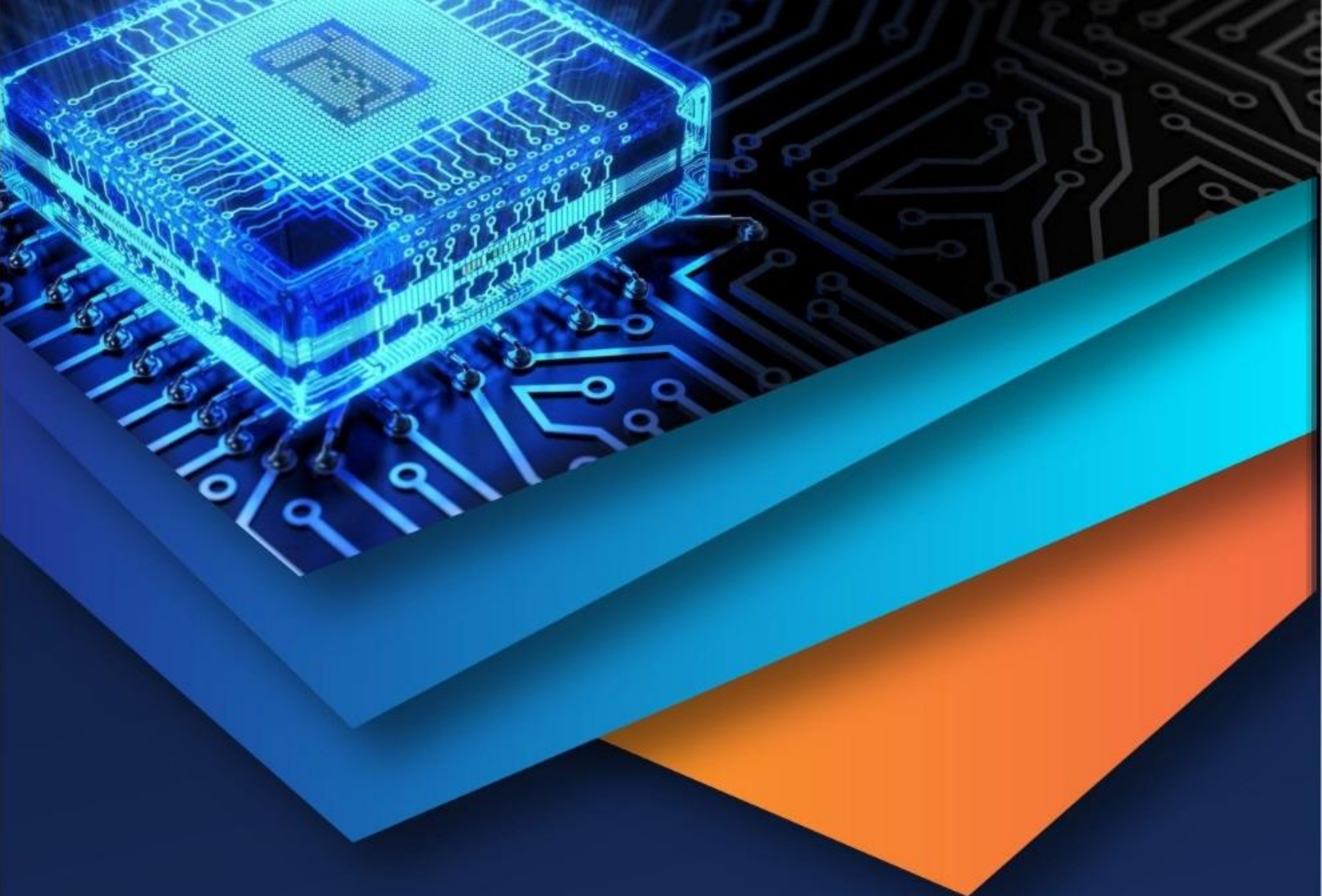
[21] Fawcett JK and Scott JE. A rapid and precise method for the determination of urea. J Clin Path. 1960; 13: 156-159.
[22] Doherty D. In Methods in Enzymol, (Eds. Tabor, H and Tabor, C W). 1970; 17 (Part A): pp. 850.
[23] Marsh WH, Fingerhut B, Miller H. Automated and manual direct methods for the determination of blood urea. Clin. Chem. 1965; 11: 624–627.
[24] Slot C. Plasma creatinine determination. A new and specific Jaffe reaction method. Scand.j.Clin.Lab.Invest.1965; 17: 381-387.
[25] Campbell JW, “Excretory nitrogen metabolism” in Environmental and Metabolic Animal Physiology. Comparative Animal Physiology, 4th Edn., ed. Prosser C. L., editor. (New York: Wiley-Interscience). 1991; 277–324.
[26] Kumar A, Sharma B, Pandey RS. Assessment of stress in effect to pyrethroid insecticides, λ-cyhalothrin and cypermethrin in a freshwater fish, Channa punctatus (Bloch). Cell. Mol. Biol. 2012; 58(1):153-9.
[27] O’Brien PJ, Slaughter MR, Polley SR, Kramer K. Advantages of glutamate dehydrogenase as a blood biomarker of acute hepatic injury in rats. Lab Anim. 2002; 36: 313-321.
[28] Vellas F & Serfaty A. Ammonia and urea in a freshwater teleost, the carp (Cyprinus carpio L). J Physiol Paris. 1974; 68: 591.
[29] Goel S. Role of vitamin B12 against methyl-parathion induced haematological indices in a freshwater teleost, Channa punctatus. Ploo. Res. 1996; 15(4): 339.
[30] Saha N, Datta S, Biswas K, and Kharbuli ZY. Role of ureogenesis in tackling problems of ammonia toxicity during exposure to higher ambient ammonia in the air-breathing walking catfish Clarias batrachus. J. Biosci. 2003; 28(6): 733-742.
[31] Hunter A. The creatine content of the muscles and some other tissues in fishes. J Biol Chem. 1929; 81: 513–523.
[32] Lynch MJ, Raphael SS, Mellor LD, Spase PD and Inwood MJH. Medical Laboratory Technology and Clinical Pathology, 2nd Ed. W.B. Saunders Co. Toronto, London. 1969.
[33] Goel KA and Garg V. 2, 3, 4- Triaminobenzene induced haematological anomalies in fish Channa punctatu. Bull Environ Contam Toxicol. 1980; 25, 136.



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