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Assessement of nuvan toxicity to lipids in snake headed fresh water fish *Channa punctatus*

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Abstract

The healthy functioning of the biosphere in our planet, the life depends entirely on the water flow and steady state phenomenon. Lethal concentration of Nuvan calculated for the fresh water snake headed fish *Channa punctatus* 0.27ml/L after 24, 48, 72 and 96 hours. For the chronic study 1/10th Nuvan concentration (0.027ml/L) provided to observe fish. Blood serum cholesterol (Chol.) and triglycerides (TG) estimated after chronic toxic stress of Nuvan to fish *Channa punctatus.* Fish serum TG revealed significant decrease level while Cholesterol showed significant elevated level after 7, 14, 21, and 28days at different level $p > 0.05$, $p < 0.01$ and $p < 0.001$ in fresh water fish *Channa punctatus.*

***Keywords**: Toxicity, fish, cholesterol, fish serum*

Introduction

Water is not only a vital environment factor to all form of life but it has also a great role to play in socio-economic development of human population, so earth is most intimately linked with water. Aquatic environment is subjected to different types of pollutants which enter water bodies with industrial domestic agriculture wastes water and severely affect the water. Aquatic environment is subjected to different types of pollutants which enter water bodies with industrial domestic agriculture wastes water and severely affect the water. Due to the intensive development of agriculture and growing food demands, there has been a great agriculture and growing food demands, increase in the manufacture and utilization of pesticides like insecticides, herbicides and other organic chemicals.Organophosphates widely used in agricultural field including the major crops such as cotton, rice, corn, wheat, barley, sorghum and soyabean. Their use is also important in top fruit, and vegetables for both foliage and root protection. Selective toxicity data have been exploited in veterinary uses of the organophosphorus as for ectoparasites control on cattle and sheep in the form of ear tags sprays and dips. Organophosphate and their residue which used in farming are continuously discharge into the environment. When the fresh water get contaminated by the various kind of pollutant like Nuvan that create mainly by humans. So the fish live in very intimate contact with their environment and are therefore very susceptible to measure water quality which may be reflected by the fish health. Blood is highly susceptible to internal environment. In most cases in the medium for signals in the animal disturbance a integrated functions can be detected or strongly indicated, with rather simple analysis of blood parameters. Cholesterol is a compound of all the cell membrane as well as membranes of the cellular organelles. It is also a precursor of steroid as cortisol, testosterone and estrogen hormones and bile salts. Cholesterol serves as a marker for both cardiovascular disease and oxidative stress. It also linked with increased carotid plaque and CVD, coronary disease, biliary cirrhosis, liver disease and nephrotic syndrome.While Triglycerols (TGs) are non polar hydrophobic molecules essential insoluble in water TGs provide stored energy and insulation. Immediately after a meal TG appear in

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the blood as major constituent of chylomicrons. The remaining TGs, plus additional triglycerides synthesized within the liver are then re-packaged as VLDL and secreted into the blood from liver. TG level may be associated with a higher risk of heart disease and stroke, cirrhosis, nephrotic syndrome, diabetes obesity and cardiovascular disease (CVD).

If the toxicant disrupt fish blood biochemistry blood serum parameters choosed to present finding to find out effect of toxicant on whole body of experimental animal fish *Channa punctatus* lipids like cholesterol and triglyceroides to heart function.

Material and Methods

Healthy live fresh water snake headed fish *Channa punctatus* weighing 50to70 gms and 12-14 cms in length were collected from local fresh water pond Malpura at Agra district (U.P.) in November month. Fishes were kept in large glass aquarium (75×37.5×37.5 cm) capacity 25 nonchlorinated tap water, which was stored one week before experiment. Aquaria bathed 1% $Kmno_4$ solution to avoid any kind of dermal infection. Fishes were acclimatized seven days prior to experiment at temp. ± 20-25°C with 7.2 pH. During experimentation commercial marketed food or egg yolk was provided to fish twice in a day 10.30 am and at 4.30 pm. Feeding was stopped 24 hours before starting the experiment. Dead fish (if any) removed from aquaria as soon as possible to avoid water fouling, water was changed after 2 or 3 days.

Nuvan "Dichlorvos" (DDVP) purchased from local Chipitola market at Agra which manufactured by Syngenta India Ltd. 14 J Tata road, Mumbai has taken for present study. Experiment divided into two parts, (i). for LC_{50} determination and (ii) for biochemical estimation.

For LC_{50} determination five aquaria were setup, four treated with different concentrations (0.1, 0.2, 0.4 & 0.8 ml) and one control group of healthy fish maintained simultaeneously.In each aquaria, six fishes were taken in 25 tap dechlorinated water. After 24, 48, 72 & 96 hours survival and mortal no. of fishes were observed and calculated mortality percentage and draw mortality percentage graph. With the help of standard table and regression line analysis calculated LC_{50} value (Table I and Fig. 1). The data was analysed statistically by log dose/probit regression line method (Finney, 1971). For chronic study sub lethal concentration of Nuvan 1/10th was applied to fish. At the end of each experimental duration 7, 14, 21 and 28 days, fish sacrificed simply by a little struck on fish head by the help of hand and after the autopsy, blood directly collected from heart chamber with the help of scissor, forceps and sterilized disposable syringe. Blood collected in centrifuge tube, kept it 30 minutes in saliently position then centrifuged for 30 minutes at 3000 rpm and after two hours supernatant carefully separated in glass vials with help of rubber bulb pipette. Fish blood serum cholesterol estimated by Wybenga *et al*. Method (1970) and serum triglycerides (TG) estimated by McGowan Method (1983). Data was analysed statistically by student 't' test, Fischer and Yates (1950).

Table – I : Toxicity evaluation of Nuvan to *Channa punctatus* specifying fiducial limits

Experimental Animal	Compound	Regression equation	LC_{50}	Variance	Fiducial limits
Channa punctatus	Nuvan (DDVP)	$Y = 5.01 + 2.22 (X_m – 2.46)$	0.27 ml/L	0.042	$m_1 (+) = 2.53$ $m_2 (–) = 2.41$

Result and Discussion

When a fresh water fish *Channa punctatus* treated with 1/10th sublethal dose of Nuvan that is 0.027 ml/L to chronic study for the period of 7, 14, 21 and 28 days then cholesterol estimated highly significant elevation in blood serum of *Channa punctatus* (Table II and Fig. 2). Increased in serum cholesterol due to organophosphate toxicity. It may be due to liver dysfunction enhanced the cholesterol production and cholestasis occuring in liver with liberation of cholesterol into blood serum by the liver cell destruction after Nuvan stress it linked with greater risk of Coronary Artery Disease (CAD).Same results came from Perrier *et al.* (1972) to *Cyprinus carpio*. Verma *et al.* (1980) resulted that cholesterol is an important biochemical component in vertebrates because of its relationship to many physiologically active steroid, it

relationship to many physiologically active steroid sex hormones, adrenal cortex hormone and bile salts etc. Tyagi (1984) have also resulted hyper cholesterolaemia in *Channa punctatus* and *Heteropneustes fossilis* respectively under exposed of various dyes.

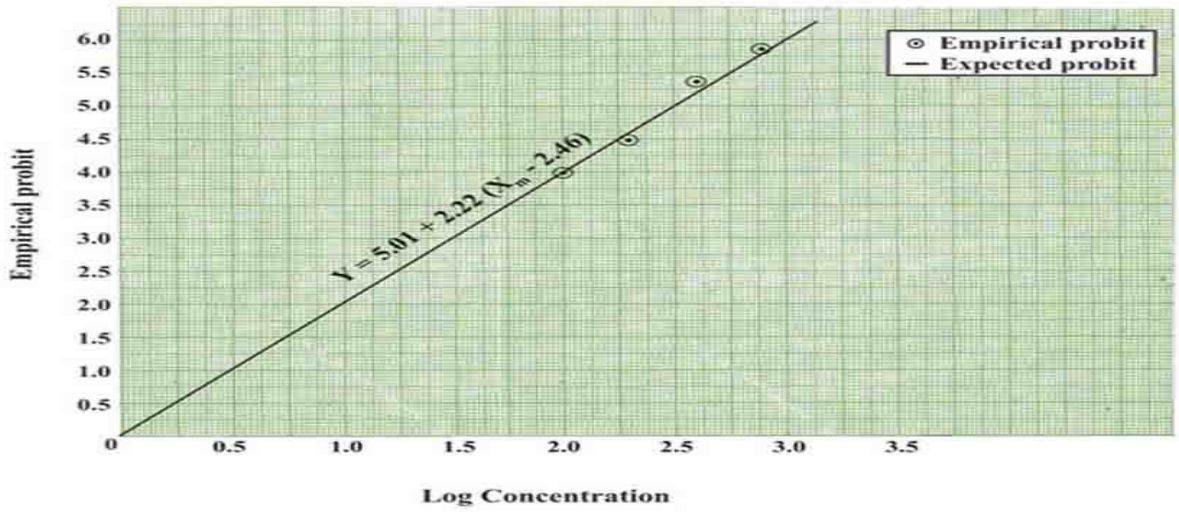
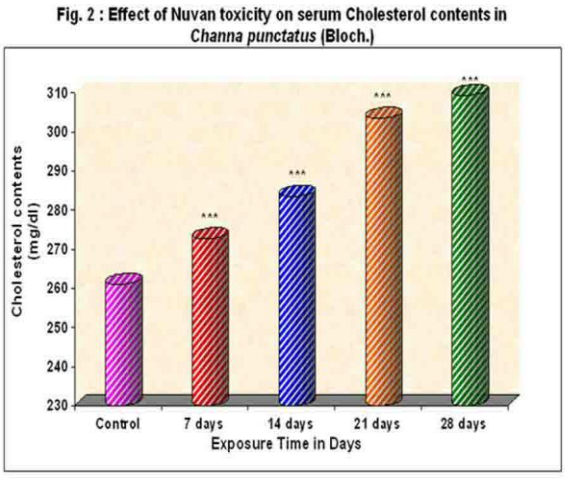


Fig. 1 : LC_{50} determination

Table II: Cholesterol content (mg/dl) in blood serum of *Channa punctatus* after Nuvan toxicity

Cholesterol (mg/dl)	Control	Exposure Time				Result
		7 days	14 days	21 days	28 days	
Range	258 – 264	270 – 276	281 – 286	300 – 307	306 – 312	Increased
Mean	261.0	272.67	283.5	303.5	309.3	
± S.Em.	0.805	0.751***	0.795***	0.982***	1.55***	
t value		10.90	19.91	30.57	21.76	

± S.Em. – Standard error of mean
***** – Very highly significant (p< 0.001)**



Bandopadhyay *et al.* (1983) viewed to elevation in cholesterol content in the fish treated with toxicant is due to necrosis of the liver cells. Hilmy *et al.* (1983) reported sharply elevation in serum cholesterol in *Angilla vulgaris* and *Mugil cephalus* due to DDT and endrin toxicity. Elevation showed, may be due to impaired function as evidence by the transfer of major cations from hepatic tissue to the serum and by elevated serum cholesterol.Awasthi *et al.* (1984) reported hypercholesterolemia to *Channa punctatus* and *Heteropneustes fossilis* due to organophosphate stress. Rao and Rao (1984) observed rise cholesterol level in *O. mossambicus* under methyl parathion. This result indicated relation to induced gluconeogenesis and the diversion of acetyl-Co A to cholesterol synthesis

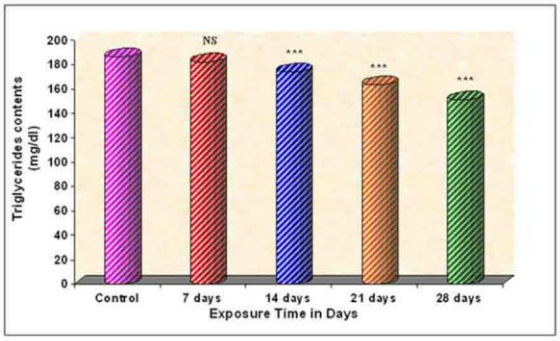
during methyl parathion intoxication. Gluth and Hanke (1985) supported alternation in level of cholesterol in *Cyprinus carpio* due to several pollutants.Radhaiah *et al.* (1987) reported increased level of total lipid content suggested the lipogenesis under pesticidal heptachlor intoxication in *Tilapia mossambica*.Tewari and Reddy (1988) find out hypercholestrolemia in *Heteropneustes fossilis* under starvation stress, it suggested probably the inactivity of liver during saturation cause irregularities in cholesterol metabolism resulting hypercholestrolemia. Reddy and Raw (1989) resulted that cholesterol enhance in *Metapenaeus monoleros* exposed to phosphomidon methyl parathion and lindane. This further stated enhancement due to increased diversion of acetyl-CoA to acetoacetate formation for cholesterol synthesis.Same observation given by Gill *et al.* (1990) to *Puntius conchorius* under phosphomidon stress and Akela *et al.* (1991) to *Clarias batrachus* under eldrin stress. Sen *et al.* (1992) also reported enhance blood cholesterol may be due to structural damage of the liver cell to *Channa punctatus*. Singh and Srivastava (1995) estimated hypercholestermia in blood of *Heteropneustes fossilis* due to formothion and propoxur stress.Goel and Agarwal (1996) depicted increased in blood cholesterol level significant in *Channa punctatus* under methyl parathion toxicity. Elevation in the blood cholesterol level may be due to the hypermetabolic state of fish or to impaired liver function. Singh and Singh supported (1997) our finding under DAP stress to *Channa punctatus*. Sharma, B. (1999) also observed very little change in serum cholesterol in *Clarias batrachus* under carbaryl toxicity. Geetha *et al.* (1999) suggested increased plasma cholesterol in *Catla-catta* due to methomyl exposure may as result of damage of liver cell. Srivastava *et al.* (2000) observed significant elevation in total cholesterol level of blood all concentration of malachite green in respect to all intervals in *Heteropneustes fossilis*.Begum and Raghawan (2001) resulted carbofuran toxicity to lipid metabolism in physiologically important tissue in food fish *Clarias batrachus*, carbofuran intoxication has elevated total lipid in all tissue investigated. Elevation suggested initiation of lipogenesis including sterols tissue steroidogensis was activated that resulting formation of corticosteriods since stress condition elevate the corticosteroids in the blood of animal.Yadav and Akela (2002) suggested increase level of cholesterol in *Channa punctatus* due to heat stress, it might be due to cholesterol metabolism is primary function of liver so the rise in level if gonad cholesterol enhanced production of it in the liver of inhibits excretion to bile duct. Adbelmeguid *et al.* (2002) explained elevation in lipid contents are frequently association with in lipid contents are frequently associated with increased bio concentration of lipophilic toxicants, which is usually correlated enhanced toxicity of the *Cyprinus carpio* and *T. zilli* under water pollution. Malhotra and Sharma (2003) resulted slight increase in blood cholesterol of *C. striatus*. Increased in cholesterol content of the blood suggested that this pesticide either enhanced the cholesterol production or inhibited excretion through the bile duct. It may also be due to necrosis of liver cells. Maruthanagayan and Sharmila (2004) reported that cholesterol level of the blood was found to be increased *C. carpio* on sublethal monocrotophos treatment.Okechukwu and Auta (2007) resulted increase level in serum cholesterol level under λ-cyhalothrin, may cause obstruction in the liver within intra and extra hepatic. However in chronic condition such as cirrhosis that's involving cells, destruction in liver cells because liver is the key organ to synthesis and excretion of cholesterol. Logaswamy *et al.* (2008) supported significant increase in cholesterol indicated lipid profile in blood thyperlipidanemia may be due to abnormal lipid metabolism which is probably the result of hepatic dysfunction and chronic hypoxic condition. Min and Kang (2008) resulted higher level of cholesterol in *Nile tilapia* and *O. niloticus* under water borne benomyl.When a fresh water fish exposed 1/10th sublethal dose of Nuvan (0.027ml/l) from the period at 7, 14, 21 and 28 days. TG level showed reduces significantly in blood serum of *Channa punctatus* (Table III and Fig. 3).Decrement in triglycerides in stress fish might be occur liver cirrhosis, affecting synthesis of TG due to reduce glucose availability in treated fish is essential for TG synthesis because it form alpha glycerophosphate which is the specific precursor of glycerol with fatty acids and toxicant may block the secretion into the serum of *Channa punctatus* under toxic stress. Same result in our favour with pesticide stress on fish.

Table III:Triglyciredes contents(mg/dl) in blood serum of *Channa punctatus* after Nuvan toxicity

Triglycerides (mg/dl)	Control	Exposure Time				Result
		7 days	14 days	21 days	28 days	
Range	184 – 189	177 – 188	169 – 178	160 – 166	147 – 155	Decreased
Mean	186.5	182.5	174.0	163.3	151.0	
± S.Em.	0.636	1.26^{NS}	0.968***	1.21***	0.913***	
t value		2.235	9.12	13.41	27.51	

± S.Em. – **Standard error of mean**
NS – **Non significant (p > 0.05)**
******* – **Very highly significant (p< 0.001)**

Fig. 3 : Effect of Nuvan toxicity on serum Triglycerides contents in *Channa punctatus* (Bloch.)



Lombardi (1966) described four several mechanisms that can effect for accumulation of TG. The rate of synthesis of hepatic triglyceride is normal, but the liver cell is unable to secrete the triglyceride into the plasma serum. This secretion is normal but rate of synthesis is increased. There is both an increase in the rate of synthesis and a block in the secretion of the synthesized TG and the TG synthesis takes place in a compartment of the cell other than endoplasmic reticulum and the pool is not accessible to the normal secretary pathway. It appears that a combination of liver necrosis, effecting the synthesis of TG and blockage of the secretion into the serum was responsible for the inhibition observed prolonged exposure period in fish *Channa gariepinus* after λ-cyhalothrin.

Folmar (1993) resulted decrease level of TG in *Lagodon rhomboides* blood after exposure of three chemical. Hussien *et al.* (1996) depicted decrease TG level in *Chrysichthyes auratus* under atrazine toxic stress, decrement if TG concentration could be due to decreased glucose availability in exposed fish glucose in essential for TG synthesis, because it form alpha glycerophosphate which is the specific precursor of glycerol with which fatty acids. Das and Bhattacharya (2002) resulted decrease TG value in *Channa punctatus* in the toxic administration.

Okechukwu and Auta (2007) reported inhibition in TG level may be due to rate of synthesis and rate of release of TG by the parenchymal cells into the systemic circulation under toxic stress in *Clarias gariepinus* λ-cyhalothrin. Kori *et al.* (2007) depicted decrease in plasma TG level allow to pressure that the lipolysis procedings during exposure period was the major source of energy. In exposure condition TG are known to be lipolytically broken down to glycerides and fatty acid and the muscle stop using glucose and restrict their ketone utilization to necessary energy being supplied via oxidation of fatty acid. Min and Kang (2008) did not find significant result in TG level of *Nile tilapia* and *O. niloticus* under benomyl stress. Velisek *et al.* (2008) reported reduce level of TG in rainbow trout under metribuzin stress.

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