Effect of thiamethoxam alters serum biochemical parameters in *Channa punctatus*(Bloch)

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On treatment with sublethal concentration of Thiamethoxam the serum biochemical parameters of fish, *Channa punctuatus* could be altered. The levels of glucose, lactate, amino acid nitrogen (AAN), creatine, urea, bilirubin, phospholipids, triglycerides and nonesterified fatty acids (NEFA) increased with decrease in the level of protein, non-protein nitrogen (NPN), pyruvate and albumin during the toxic exposure periods at different time intervals in the serum of the fish. There was increase in the level of glucose due to glycogenic activity and decrease in protein level due to enhanced proteolytic activity under stress condition. Low levels of pyruvate and high level of lactic acid indicated that fish can shift from aerobic to anaerobic condition. The elevated levels of creatine, bilirubin, urea and decrease in the albumin context under stress condition revealed renal failure and acute hepatic necrosis. The levels of serum phospholipids, triglycerides, non esterified fatty acids (NEFA) were increased due to elevation which might be due to increased esterification reactions under stress condition.

Key words : Thiamethoxam, Biochemical parameters, Blood, Channa punctatus

INTRODUCTION

The pesticides in aquatic ecosystems affect non target organisms such as fishes and prawns. Pesticide hazard on fish mortality, growth and tissue damage has been amply reported by Jackson (1976). These toxic chemicals change the quality of water that affect the fish and other aquatic organisms (Dhasarathan *et al.*, 2000). Among all insecticides the organochloride (OC) are widely used to control pests because of their rapid effectiveness and easy biodegradation (Mahboob and Siddiqui, 2002). According to other researches OC cause a number of subsidiary problems like effecting growth and the reproductive and immune systems by causing morphological, pathological and physiological changes and by altering biochemical constituents of fish and other animals (Singh *et al.*, 2004; Seth and Saxena, 2003).

A number of recent clinical studies revealed that most of the OC and other toxic chemicals could alter the immune system (Barcarolli and martinez, 2004; Thangavel *et al.*, 2004; Chen *et al.*, 2004). Blood being the medium of intercellular transport comes indirect contact with various organs and tissues of the body. The physiological state of an animal at particular time is reflected in its blood. Moreover, pesticides rapidly bind to blood proteins and induce the immune system.

In the present study, the toxic effect of thiamethoxam (1-(2-chloro-1, 3-thiazol-5-ylmethyl)-5-methyl-1, 3, 5-oxadiazinan-4-ylidene N-nitroamine) on kidney and liver

function and on serum biochemical parameters of *Channa punctatus* was investigated. The effect of a sublethal concentration of thiamethoxam in short term experiments is studied to understand the nature of the toxicity exerted by this pesticide on the vital activities of this species.

MATERIALS AND METHODS

Channa punctatus a fresh water edible fish, weighing average of 82-120 g and 25.5 ± 1.21 cm in length, were procured from a local market, Warangal (AP). The collected fish were kept in a cement tank ($6x_3x_3$ feet) atleast for one month for acclimatization under continuous water flow. The average temperature of water was $22\pm1^{\circ}$ C. The fish were fed *ad libitum* with groundnut cake along with the commercial pellets (1-1.5% body weight). They were starved one day before experiment (Butlerworth, 1972). Without discrimination of sexes, both the sexes of fish were used for the experiment. The physiological parameters of water are given in Table 1. The LC 50 of commercial grade thiamethoxam (114.8 ppm) was determined for 48 hours by the method of Bayne *et al.*(1977).

Batches of six (6) fish were exposed to 24,48,72 and 96 hours for sublethal concentration (38.26 ppm) along with control fish in separate tanks consisting of six liters of water, at the room temperature. After the stipulated time intervals, the fish were removed and the blood was collected in the tubes by caudal puncture. For

Table 1 : Physico-chemical parameters of tap water						
Sr. No.	Parameters	Values				
1.	Temperature	$22 - 24^{\circ}C$				
2.	pH Hydrogen ion concentration	7.2 - 7.3				
3.	Electrical conductivity (milli ohmes/cm)	0.52				
4.	Calcium (mg/litre)	5.0				
5.	Sodium (mg/litre)	2.1				
6.	Bicarbonates (HCO ₃) (mg/litre)	142				
7.	Total alkalinity (mg/litre as Ca ₂ CO ₃)	69				
8.	Sulphate (mg/litre)	7.1				
9.	Nitrates (mg/litre)	3.4				
10.	Iodine (mg/litre)	0.01				
11.	Chlorides (mg/litre)	37.0				
12.	Dissolved oxygen (mg/litre)	9.2				
13.	Biological oxygen demand (BoD)(mg/litre)	1.6				
14.	Chemical oxygen demand (CoD) (mg/litre)	0.008				
15.	Free carbondioxide (mg/litre)	10.0				
16.	Floride (F ⁻)(mg/litre)	0.03				

further investigations of toxic effects the following methods have been adopted.

Glucose content was determined by the method of Folin and Wu (1920) and pyruvate by the method of Fridmann and Haugen (1942). The levels of lactic acid were estimated by the method of Barker and Summerson (1941). The protein content was determined by Doomas et al.(1981). The non protein nitrogen was estimated as described by Gupta and Bhargava (1985) and amino acid nitrogen (AAN) according to Goodwin (1970). Creatine were determined as described by Gupta and Bhargav (1985). The levels of bilirubin were determined by the method of Gupta and Bhargava (1985) and albumin according to Doomas et al.(1981). The levels of urea were determined by the method of Columbo and Favpreaon (1965) and phospholipids by the method of Connerty et al.(1961). The triglycerides content was determined by Foster and Dunn (1973). The activities of non esterified fatty acids (NEFA) were estimated by the method of Trout et al.(1960).

RESULTS AND DISCUSSION

It is observed from Table 2 that thiamethoxam significantly influenced serum biochemical parameters of *Channa punctatus*. The data revealed that increase in glucose, lactate, amino acid nitrogen (AAN), creatine, urea, bilirubin, phospholipids, triglycerides and non-esterified fatty acids (NEFA) with a decrease in the levels of protein, non-protein nitrogen (NPN), pyruvate and albumin was due to pesticide influence. Increase in blood

glucose level was due to breakdown of stored glycogen which also indicated that the fish could yield excessive energy by the utilization of reserved glycogen. Similar findings were also observed by Abdul Naveed et al. (2005), Tugiyono and Gagnon (2002), Anitha Kumari and Sree Ramkumar (2006) and Chakravarthy et al.(2006). The level of protein decreased due to increased proteolytic activity which might be increased in amino acid pool during the pesticide exposure period (Kaur and Kaur, 2006). Increased lactic acid levels are more in thiamethoxam treated fish under stress condition due to more mobilization of pyruvic acid into lactic acid (Thangavel et al., 2004). The levels of pyruvic acid was found to be decreased in thiamethoxam treated fish may undergo to physiological stress under the impact of insecticide. According to Das and Mukharjee (2003) that the pesticide could cause enhance the muscle movement causing decrease in the levels of pyruvic acid. The non-protein nitrogen (NPN) content is decreased in C.punctatus due to the pathological conditions under toxicity of pesticides. (Abdul Naveed et al., 2004).

The amono acid nitrogen (AAN) content was increased due to increase proteolytic activity and also renal failure under toxicity. Harold varley (2005) reported that the liver deamination of amino acids in only impained when liver damage is severe. Creatine was increased during prolonged exposure of thiamethoxam in *C.punctatus*. Creatine is synthesized in liver, from there it enters into the circulation to be taken up by the muscle in which it is converted into creatinine phosphate (David and Michael, 2005; Tyagi and Srivastava, 2005).

The levels of urea may be increased in the serum due to renal failure or kidney necrosis. According to Kumari et al. (2006) that the increase in serum urea levels is due to conversion of toxic ammonia in to non toxic urea and glutamine. The enhanced levels of bilirubin indicated that these insecticides could cause a severe damage to liver. Jain (2006) reported that the PCB metabolites cause necrosis of liver cells. Damaged liver cells are enable to conjugate bilirubin and therefore, the levels of serum bilirubin are enhanced. The decrease in albumin indicatd fall in osmotic pressure leading to enhanced fluid retension tissue spaces causing edema in animals. According to Sathyanarayana (2005) the major role of albumin in the animals is to maintain the osmatic pressure. The levels of phospholipids are increased during prolonged exposure periods in Channa punctatus. Singh and Singh (2006) reported the effect of endosulfan exposure for various phospholipids on hepatic lipogenesis and mobilization to test via plasma during the different phases of its reproductive cycle in a tropical telecast

Parameters	Control	Thiamethoxam treated			
Tarameters		24 Hrs	48 Hrs	72 Hrs	96 Hrs
Glucose	56.43 <u>+</u> 1.39	59.03* <u>+</u> 1.39	64.82* <u>+</u> 4.81	67.33 <u>+</u> 1.19	70.84 <u>+</u> 1.86
(mg/100ml)		PC=4.60	PC=14.86	PC=19.31	PC=25.53
Pyruvate	15.36 <u>+</u> 1.09	14.16* <u>+</u> 0.86	12.32 <u>+</u> 0.63	10.89 <u>+</u> 0.53	9.23 <u>+</u> 0.45
(micrograms/100ml)		PC=-7.81	PC=-19.79	PC = -29.10	PC=-39.90
Lactate	5.03 <u>+</u> 0.38	5.89 <u>+</u> 0.67	6.71 <u>+</u> 0.75	8.36 <u>+</u> 0.82	9.42 <u>+</u> 0.36
(micrograms/100ml)		PC=17.09	PC= 33.39	PC= 66.20	PC= 87.27
Proteins	308.29 <u>+</u> 6.82	286.69* <u>+</u> 1.39	264.36* <u>+</u> 2.66	238.46 <u>+</u> 1.86	231.34 <u>+</u> 2.36
(mg/100ml)		PC = -7.006	PC=-14.24	PC=-22.65	PC=-24.96
Non-protein-nitrogen	16.34 <u>+</u> 0.89	15.01* <u>+</u> 0.78	13.01 <u>+</u> 0.68	12.31 <u>+</u> 0.51	10.16 <u>+</u> 0.85
(mg/100ml)		PC=-8.13	PC=-20.37	PC=-24.66	PC=-37.82
Amino acid nitrogen	0.78 ± 0.02	0.81* <u>+</u> 0.06	0.93 <u>+</u> 0.062	1.07 <u>+</u> 0.03	1.58 <u>+</u> 0.06
(mg/100ml)		PC= 3.84	PC=19.23	PC=37.17	PC=102.56
Creatinine	0.24 <u>+</u> 0.01	$0.26* \pm 0.012$	0.29 <u>+</u> 0.017	0.34 <u>+</u> 0.013	0.36 <u>+</u> 0.013
(mg/100ml)		PC=8.33	PC= 20.83	PC=41.66	PC=50
Urea	15.21 <u>+</u> 1.63	16.65* <u>+</u> 2.11	17.63* <u>+</u> 1.23	18.91 <u>+</u> 1.36	20.08 <u>+</u> 0.92
(micromoles/litre)		PC=9.46	PC=15.91	PC=24.32	PC=32.01
Bilirubin	0.12 <u>+</u> 0.001	0.135 <u>+</u> 0.03	0.168 <u>+</u> 0.015	0.174 <u>+</u> 0.051	0.183 <u>+</u> 0.081
(mg/100ml)		PC=12.5	PC=40	PC=45	PC=52.5
Albumin	1.98 <u>+</u> 0.01	1.86* <u>+</u> 0.14	1.71* <u>+</u> 0.12	1.66 <u>+</u> 0.19	1.39 <u>+</u> 0.21
(micrograms/100ml)		PC = -6.06	PC=-13.63	PC=-16.16	PC=-29.79
Phospholipids	2.1 <u>+</u> 0.01	2.36* <u>+</u> 0.72	2.50 <u>+</u> 0.38	2.96 <u>+</u> 0.32	3.0 <u>+</u> 0.17
(mg/100ml)		PC=12.38	PC=19.04	PC=40.95	PC=43.33
Triglycerides	1.84 <u>+</u> 0.06	2.41 <u>+</u> 0.15	2.68 <u>+</u> 0.11	2.79 <u>+</u> 0.32	2.95 <u>+</u> 0.25
(mg/100ml)		PC=30.99	PC=72.28	PC=51.63	PC=60.33
Non-esterified fatty acids	86.20 <u>+</u> 3.71	90.21* <u>+</u> 4.72	47.86* <u>+</u> 5.82	109.65 <u>+</u> 4.82	115.31 <u>+</u> 3.82
(micromoles/liter)		PC=4.65	PC=13.52	PC=27.20	PC=33.82

Each value is mean \pm S.D of (6) individuals. PC denotes percentage change over control. Means were compared with Mannu-Whitney u-test at p<0.05 for statistical significance. *Non significant.

H.fossilis.

Increase in the triglyceride content was consideration of elevation which might be due to increased esterification reactions under stress condition (Balasubramaniyam *et al.*, 2003). The raised values in non-esterified fatty acids (NEFA) may be due to starvation or due to emotional stress condition. The levels of NEFA may be enhanced due to hormonal imbalance under toxic stress. Shankar and Kulkarni (2005) observed the enhanced levels of NEFA indicate that the fish can utilize the NEFA for producing additional energy to support the metabolic activities during toxic condition.

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