# Biochemical changes in the blood of *Oreochromis mossambicus* exposed to Textile Dyeing effluent

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# Abstract

Toxic effect of textile dyeing effluent in the fish, *Oreochromis mossambicus* was investigated in the present study. Textile dyeing effluent discharged from dyeing units at Telungupalayam, Coimbatore, Tamil Nadu were collected and adult fish of size 16  $\pm$  2 g were exposed to a sublethal concentrations (1/10<sup>th</sup> and 1/20<sup>th</sup> LC50 value) of the effluent for a period of 10, 20 and 30 days. The treated fish were compared with the control group for the biochemical alterations in the blood. The study revealed marked changes in the blood of the effluent treated fish.

Key Words: Oreochromis mossambicus, biochemical, blood, textile dyeing effluent.

# INTRODUCTION

The discharge of untreated and partially treated waste water from various industries like chemicals , pesticides , fertilizer , pulp and paper mill , sugar and distillery , tannery and textile processing etc., have polluted the aquatic bodies drastically. The discharge of textile wastewaters into aquatic habitats is of great concern since the discharges are mostly made untreated or partially treated due to poor enforcement of existing laws in the developing world including India(Sharma *et al* 2007). The toxicity bio assay is a biological tool used to evaluate the risk of pollutants and it gives a global answer for chemical agents dissolved in industrial and domestic effluents (Parthipan, 2014), as they effect the activity of biologically active molecules in aquatic living organism. The present study aims at evaluating the effect of textile effluents on the concentration subsequent changes occurring in the blood of the fresh water fish, *Oreochromis mossambicus*.

# **MATERIALS AND METHODS**

*Tilapia mossambica* is considered as the test organism in the present study. Mature male and female *Tilapia mossambica* were collected from Tamil Nadu Fisheries Development Corporation Limited, Aliyar Fish Farm, Aliyar, Tamil Nadu, India. The  $LC_{50}$  value of the textile dyeing effluent to *Tilapia mossambica* was 12.06 %. Sublethal concentration of the test was made at  $1/10^{\text{th}}$  and  $1/20^{\text{th}}$  of  $LC_{50}$  value. After the end of each exposure period four healthy male and four healthy female were sacrificed immediately after taking a drop of blood from caudal vein for blood preparation. The blood samples for biochemical analysis was centrifuged for 10 minutes at 4,000 rpm supernatant serum was decanted and stored at  $-2^{0}C$ .

# **Estimation of Blood sugar**

Blood sugar was analysed using Glucose test kit (AGAPPE Diagnostics, India) using GOD – PAP methodology (Trinder, 1969).

# **Estimation of Serum Urea**

Urea was analysed in the sample serum with Urea test kit (AGAPPE Diagnostics, India) based on Urease – Glutamate Dehydrogenase (GLDH) methodology (Kassirer, 1971).

#### **Estimation of Serum Creatinine**

Serum creatinine was analysed using creatinine test kit (AGAPPE Diagnostics, India) based on method described by Artiss *et al.*, (1984

#### **Estimation of Bilirubin**

Bilirubin was analysed in the sample serum with Bilirubin test kit (AGAPPE Diagnostics, India) based on modified TAB method (Water and Gerard, 1980).

#### Estimation of Sodium

Sodium was analysed in the sample serum with Sodium test kit(AGAPPE Diagnostics, India) based on Uranyl Acetate method (Trinder, 1951).

#### **Estimation of Potassium**

Potassium was analysed in the sample serum with potassium test kit (AGAPPE Diagnostics, India) based on Na - Tetraphenylborate Turbidometric) method (Henry, 1974).

#### Results

Sugar and serum cholesterol in the blood of fishes exposed to the effluent had significant **decrease** (P < 0.05) over control. Whereas serum urea, serum creatinine, serum bilirubin, sodium and potassium in blood of fish exposed to textile dyeing effluent showed significant **increase** (P < 0.05) over control for both the concentrations. The mean values of the results obtained are presented in table 1–7.

#### **Blood** sugar

The percentage of decrease noticed in fish groups exposed to  $1/20^{\rm th}$  of  $LC_{50}$  concentration was -6.54 to -12.09%, whereas for  $1/10^{\rm th}$   $LC_{50}$  concentration it was -10.28 to 21.98% over fish under control.

#### Serum cholesterol

The percentage decrease over control in fish groups exposed to  $1/20^{\rm th}$   $LC_{50}$  concentrations were -22.02 to -30.67% and for fish groups exposed to  $1/10^{\rm th}$  of  $LC_{50}$  was -23.21 to -48%.

#### Serum urea

The percentage increase over fish under control was 5.45 to 31.57% for fish group exposed to  $1/20^{\text{th}}$  LC<sub>50</sub> concentration. For  $1/10^{\text{th}}$  LC<sub>50</sub> concentration, this percentage increase over fish under control were 21.81 to 56.14%.

#### Serum creatinine

The percentage increase of serum creatinine in  $1/20^{\rm th}$  LC<sub>50</sub> concentration exposed fish was 4.5 to 9.35% over fish under control. This percentage difference in  $1/10^{\rm th}$  LC<sub>50</sub> concentration exposed groups were 9.09 to 15.8%.

# Serum bilirubin

The serum bilirubin content in the blood of *T.mossambica* exposed to  $1/20^{\text{th}}$  of LC<sub>50</sub> concentration showed 9.09 to 10% increase over fish under control. This percentage increase in fish exposed to  $1/10^{\text{th}}$  of LC<sub>50</sub> was 20 to 27.7%.



#### Sodium and Potassium

The percentage increase of sodium content in fish groups exposed to  $1/20^{th}$  LC<sub>50</sub> concentration was 7.61 to 13.27% and for fish groups exposed to  $1/10^{th}$  of LC<sub>50</sub> was 21.74 to 32.65% when compared to fish under control. The percentage increase of potassium content in fish groups exposed to  $1/20^{th}$  LC<sub>50</sub> concentration was 12.5 to 33.12% and for fish groups exposed to  $1/10^{th}$  of LC<sub>50</sub> was 18.75 to 40.62%.

#### Statistical analysis

All the parameters showed significant difference between durations and concentrations at 5% levels (Table – 8).

#### DISCUSSION

#### Blood sugar

Glucose is one of the most sensitive indices of the stress state of an organism. In the present study, the level of blood glucose decreased over fish under control, and as the days of effluent exposure increased the decrease was prominent. The results show that the percentage decrease was high in higher concentration of the effluent. Greater reduction of glucose was noticed in 30 days exposure and lowest in 5 days exposure of the effluent. The decrease in glucose content is most probably could be related to increased glucose utilization required for the induction of liver enzymes and microsomal protiens (Bhattacharya *et al.*, 1987).

#### Serum cholesterol

Cholesterol is an important component of cell membranes and functions as a precursor for the synthesis of sexual hormones (Munoz-Cueto *et al.*, 1996). Cholesterol levels can indicate disorders of lipid and lipoprotein metabolism and especially related to liver disease (Smolowitz *et al.*, 1998). Serum cholesterol content in fish exposed to different effluent concentrations showed a decreasing pattern as the exposure period increased in all the concentrations they were exposed. The decrease in cholesterol level in the present investigation may be due to the lack of or low availability of acetyl CoA for cholesterol synthesis in the absence of glucose utilization and dependence of body on alternate energy sources.

#### Serum urea

Urea is present in all fish and liver is the primary organ of production and it is excreted out through gills (Walsh *et al.*, 2003). Therefore an elevated blood urea is probably not indicative of renal disease as it might be in humans, but is more likely associated with gill or liver disease. In the present study, fish exposed to textile dyeing effluent at different concentrations showed an increase in serum urea with duration of exposure and increased concentration of the effluent. Thus from the results obtained, it is revealed that the effluent has greater impact on the rise in serum urea content. However, increasing urea content in the blood is likely an indicator of failing of osmoregulatory capability of the gill.

#### Serum creatinine

Creatinine is a nitrogenous waste product that is eliminated by the kidneys, then excretion is suppressed in renal insufficiency. In the present investigation, the creatinine level was increased in the blood of *Tilapia mossambica* after exposure to textile dyeing effluent. The serum creatinine content increased significantly in the effluent exposed fish as the experimental periods increased. A marked increase was noticed as the exposure of duration increased. Increased level of serum creatinine after exposure to the effluent indicates the functional defect of kidney. Similar observations were made on Tilapia on exposure to carbofuran pesticide (Soufy *et al.*, 2007).

#### Serum bilirubin

Bilirubin is the main bile pigment that is formed from the breakdown of haeme in red blood cells (Gupta and Shubhra, 2006). The bilirubin content in the serum of fish exposed to the effluent was found to increase with increase in concentration of the effluent and also exposure time. At 20 and 30 days exposure even though there was increased bilirubin content when compared to fish under control, the increase was not that much pronounced as noticed in 5 and 10 days exposure. The reason for this may be that after 20 days exposure, the bile production might have hindered due to damage of liver cells. From the results, it is clear that as the concentration of the effluent increases, there was marked increase in bilirubin content in effluent exposed fish groups. The increase may be attributed to the damage of hepatocytes, obstruction of bile duct, enlargement of gall bladder or a resultant hemolysis. Similar result was noticed by Jayantha *et al.*, (1984) in *O.mossambicus* treated with phosphamidon.

#### Serum sodium and potassium

In the present study, on exposure to textile dyeing effluent, sodium and potassium content in the blood sera showed increase with increased concentration of the effluent and duration of exposure. This increased sodium and potassium concentrations may be attributed to kidney impairment where the kidney is the normal pathway for Na and K. Such an increase in sodium and potassium content in the serum of fish exposed to textile dye stuffs were reported by Atamanalp *et al.*, (2002) and Atamanalp (2007).

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# Table 1. Changes in the Blood Sugar of T. mossambica exposed to textile dyeing effluent

		Concentrati	on of Effluent		
		$1/10^{\text{th}}$ of LC <sub>50</sub>		$1/20^{\text{th}}$ of LC <sub>50</sub>	
Duration	Control	Expt.value	%difference	Expt.value	%difference
(Days)	± SD	(gm%)	over	(gm%)	over
	(gm%)	± SD	control	± SD	control
5	107±2	96±7	-10.28%	100±4	-6.54%
10	101±4	89±3	-11.88%	93±6	-7.92%
20	95±5	80±1	-15.79%	86±8	-9.47%
30	91±2	71±5	-21.98%	80±3	-12.09%

Table 2. Changes in the Cholesterol in the blood of T. mossambicaexposed to textile dyeing effluent

		Concentration of Effluent				
		$1/10^{\text{th}}$ of LC <sub>50</sub>		$1/20^{\text{th}} \text{ of } LC_{50}$		
Duration	Control	Expt.value	%difference	Expt.value	%difference	
(Days)	± SD	(mg/dl	over control	(mg/dl	over control	
	(mg/dl)	± SD		± SD		
5	168±4	129±5	-23.21%	131±7	-22.02%	
10	160±6	120±4	-25.00%	123±6	-17.5%	
20	154±3	95±7	-38.31%	112±8	-27.27%	
30	150±2	78±10	-48.00%	104±5	-30.67%	

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		Concentration of Effluent				
		$1/10^{\text{th}}$ of LC <sub>50</sub>		$1/20^{\text{th}} \text{ of } LC_{50}$		
Duration	Control	Expt.value	%difference	Expt.value	%difference	
(Days)	± SD	(mg/dl)	over	(mg/dl)	over	
	(mg/dl)	± SD	control	± SD	control	
5	16.5±0.27	20.1±0.13	21.81%	17.4±0.52	5.45%	
10	16.9±0.14	22.8±0.22	34.91%	18.2±0.3	7.69%	
20	17.5±0.41	26.2±0.9	49.71%	20.7±0.9	18.28%	
30	17.1±0.35	26.7±0.5	56.14%	22.5±0.6	31.57%	

# Table 3. Changes in the Serum Urea in the blood of T. mossambica

# Table 4. Changes in the Serum Creatinine in the blood of T. mossambicaexposed to textile dyeing effluent

	pncentration of Effluent				
		$10^{\text{th}} \text{ of } \text{LC}_{50}$		20 <sup>th</sup> of LC <sub>50</sub>	
Duration	Control	Expt.value	%difference	Expt.value	%difference
(Days)	± SD	(mg/dl)	over control	(mg/dl)	over
	(mg/dl)	± SD		± SD	control
5					
	1±0.0129	20±0.017	9.09%	15±0.011	5%
10	09±0.0132	23±0.014	12.84%	17±0.018	33%
20	07±0.0105	24±0.016	16.16%	18±0.015	09%
30	08±0.0155	25±0.019	15.8%	18±0.013	35%

# Table 5. Changes in the Serum Bilirubin in the blood of T. mossambicaexposed to textile dyeing effluent

		Concentrati	Concentration of Effluent			
		$1/10^{\text{th}}$ of LC	$1/10^{\text{th}} \text{ of } LC_{50}$		$20^{\text{th}}$ of LC <sub>50</sub>	
Duration	Control	Expt.value	difference	Expt.value	%difference	
(Days)	± SD	(mg/dl)	over	(mg/dl)	over control	
	(mg/dl)	± SD	control	± SD		
5	22±0.02	0.28±0.03	27.27%	24±0.01	9.09%	
10	25±0.01	32±0.02	28.00%	28±0.03	12.00%	
20						
	29±0.04	35±0.01	20.00%	30±0.04	3.44%	
30						
	30±0.04	38±0.02	26.66%	33±0.02	10.00%	

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		Concentration of Effluent			
		$1/10^{\text{th}} \text{ of LC}_{50}$		$20^{\text{th}}$ of LC <sub>50</sub>	
Duration	Control	Expt.value	%difference	Expt.value	%difference
(Days)	± SD	(mmol/L)	over	(mmol/L)	over
	(mmol/L)	± SD	control	± SD	control
5	92±2	112±4	21.74%	99±3	7.608%
10	96±1	118±3	22.916%	103±2	7.29%
20	95±2	125±6	31.50%	107±5	12.63%
30	98±4	130±5	32.65%	111±3	13.27%

Table 6. Changes in the Serum Sodium in the blood of T. mossambicaexposed to textile dyeing effluent

Table 7. Changes in the Serum	Potassium in the blo	ood of T. mossambica
exposed to	textile dyeing effluer	nt

		Concentration of Effluent			
		1/10 <sup>th</sup> of LC <sub>50</sub>		$1/20^{\mathrm{th}}$ of LC <sub>50</sub>	
Duration	Control	Expt.value	%differen	Expt.value	%difference
(Days)	± SD	( mmol/L)	ce over control	( mmol/L)	over control
	mmol/L)	± SD		± SD	
5	1.6±0.3	1.9±0.2	18.75%	1.8±0.2	12.5%
10	1.7±0.8	2.1±0.1	23.53%	2.0±0.1	17.64%
20	1.6±0.4	2.2±0.3	37.5%	2.1±0.2	31.25%
30	1.6±0.1	2.25±0.5	40.62%	2.13±0.3	33.12%