Toxicity of Composite Tannery Effluent and Extent of Amelioration by Traditional Method of Effluent Stabilization

Anulipi Aich[1], B. Chattopadhyay[1], S. Dutta[2] and S. K. Mukhopadhyay[3]

[1]Government College of Engg. and Leather Technology, LB, III, Salt Lake, Calcutta-700 098, India
[2]Department of Chemical Engineering, Jadavpur University, Calcutta-700 032, West Bengal, India
[3]Durgapur Government College, Durgapur-713 214, Burdwan, West Bengal, India
e-mail : aanulipi@yahoo.co.in

ABSTRACT

Throughout the year composite industrial and municipal wastewater contaminate the East Calcutta Wetlands a special conservation site (Ramsar Site No.1208), at the eastern fringe of Kolkata city, West Bengal, India. The aim of the present study was to characterize the raw tannery effluent and its amelioration through stabilization when it is productively used in pisciculture. Physico-chemical features of raw composite tannery effluent at the tannery discharge site, unmixed tannery wastewater collected from wastewater-carrying canal, 1 km away from the vicinity of Dhapa tannery agglomerate and the stabilized tannery wastewater from the selected fishpond were compared to study the extent of amelioration by natural processes. Acute toxicity tests were also performed with a fish model (Guppy; *Poecilia reticulata*) to study the impact of physico-chemical characters of tannery effluent on fish biosystem. Natural stabilization of the composite tannery effluents using traditional method was observed to be very efficient in ameliorating critical pollutants like TDS (74.18% reduction), TSS (59.62% reduction), total hardness (22.65% reduction), PO_4^{3-} (increment by 64.37% allowing nutrient support of live protoplasm building), BOD_5 (70.25% reduction), COD (56.53% reduction), and the heavy metals like Cr (98.33% reduction and Pb (34.29% reduction). Interesting to note that the stabilized tannery wastewater-fed fishpond exhibited on an average 70 times better mixing condition than the 96hr-LC_{50} mixing ratio of 1:7 for composite tannery effluent and freshwater respectively as the 96hr-LC_{50} value of the composite tannery effluent was 137.5 ml L^{-1}. The result of the present study amply justifies the reasons for sustainability of this traditional resource recovery system practiced in ECW for the last ten decades or so.

INTRODUCTION

For nearly hundred years East Calcutta Wetland (ECW) ecosystem (22^0 25'N to 22^0 40'N and 88^0 20'E to 88^0 35'E; elevation of ~2m; area 12,500 ha), a Ramsar site (No. 1208) of India at the eastern side of Kolkata city (Previously Calcutta) has been receiving contaminated composite wastewater which is about 600 million liters a day, and more than 2500 metric tons of solid wastes, discharged by 538 tanneries and nearly 5500 other small-scale establishments such as battery, electroplating, rubber, pottery and pigment manufacturing units. Tannery wastewater and solid wastes from the east Kolkata tannery agglomerates at Dhapa, Topsia and Tangra were primarily responsible for contaminating this wetland area. However, very recently slow shifting of the tannery units are being made into a specially allotted area, the Calcutta Leather Complex (CLC) at Karaidanga, Kolkata, which is about 15 km away from the previously established area, eastwards towards the Kultigong River, the final effluent discharge site. In the tanning industry, processing of every 1,000 kg wet salted hides and skins yield around 150 kg finished leather, while 150 kg split, 700 kg solid wastes in different forms and 30m3 of effluent containing 400 kg dissolved and suspended solids (Puntener, 1995). Tannery wastewater is mainly characterized by high salinity, high organic loading and harmful heavy metals, especially chromium (Tunay et al. 1999; Song et al. 2000). East Calcutta wetland ecosystem plays a major role in amelioration of the composite effluents. However, the composite wastewater is observed to play an important role in ECW ecosystem as the discharges are being productively utilized in the aquaculture after stabilization and also being used to irrigate adjoining low land agricultural effort. It is a unique agro-ecosystem that has sustained the world's largest practice of integrated resource recovery for more than 50 years. The aim of the present study was to characterize the raw tannery effluent and its amelioration through stabilization when it is productively used in pisciculture. Physico-chemical features of raw composite tannery effluent at the tannery discharge site, unmixed tannery wastewater collected from wastewater-carrying canal, 1 km away from the vicinity of Dhapa tannery agglomerate and the stabilized tannery wastewater from the selected fishpond were compared to study the extent of amelioration by natural processes. Acute toxicity tests were also performed with a fish model (Guppy; *Poecilia reticulata*) for the evaluation of quality of wastewater and to study the impact of physico-chemical characters of tannery effluent on fish biosystem. The 96-h LC_{50} tests were conducted to measure the susceptibility and survival potential of organisms exposed to tannery effluent contaminated with harmful chemicals and especially heavy metals.

MATERIAL AND METHODS

The areas under investigation are located in ECW (lat.22°33′ - 22°40′N; long. 88°25′ -88°35′E), a special conservation site (Ramsar Site No.1208), at the eastern fringe of Kolkata city, West Bengal, India. The raw tannery effluent was collected directly from the effluent discharge site of the tanneries. The first sampling station (site-I) was 1 km away from the vicinity of Dhapa tannery agglomerate, from where unmixed and untreated tannery wastewater was collected. The second sampling station (site-II) was a tannery effluent stabilized shallow fishpond (mean area 46.82 ha and mean depth 1.83 m) situated near the site site-I. Since physico-chemical composition of tannery discharges varies widely at different hours of the day due to different operational schedule at the production site, samples were collected at different hours of the day to get a good average result.

Analysis of Physico-chemical Parameters

Collected samples of water were analyzed potentiometrically by Mettler checkmate 90 Toledo (pH, Conductivity and TDS) and titrimetrically on the spot using E. Merck, Germany, field testing Aquamerck reagent kits ($NO_3$1-, $PO_4$3-, Cl-1, total hardness, carbonate hardness, acidity and alkalinity). Total suspended solid (TSS) was analyzed gravimetrically after filtration; Biological oxygen demand (BOD_5) and Chemical oxygen demand (COD) were analyzed following standard methods (APHA 1995).

Analysis of Metals

For metal analysis, samples were acidified with 1N HNO_3 to pH 2.50 and were brought to the laboratory in 500 ml glass stopper dark bottles. For the extraction of different metals Conc. HNO_3 (Analytical reagent) and $HCLO_4$ digestion of the samples was done (APHA 1995). All metals were detected by Atomic Absorption Spectrophotometer (Perkin-Elmer AAnalyst-100) using element-specific hollow cathode lamps in default condition in flame absorption mode.

Toxicity Test

Healthy adult Guppy fish (*Poecilia reticulata*) were brought to the laboratory from market for experiments. Fish were reared in 50 litre capacity aquaria for 1 week to acclimatize them with the laboratory condition. After 7 days, four individuals (2 males and 2 females) in each set was exposed to five (10%, 20%, 30%, 50% and 100%) different concentrations of raw tannery effluent of site I in five rectangular jars of 1litre capacity and were exposed for 96 hours. A control was also set for refinement of the test results. The whole experimentation was replicated for six times. Experimental water was not renewed and fishes were not fed during experiment. Mortality was recorded at the end of exposure by lack of mobility of the organisms when the test vessels were shaken. The 96-h median lethal concentrations were calculated using Karber's method (Ghosh, 1984).

Statistical Analysis

All the statistical analyses were made using computer software package *Statistica* for windows, version 5.1A, Statsoft Inc., 1996.

RESULT AND DISCUSSION

DO, BOD and COD are two most important parameters to measure the organic pollution of a water body and to measure the O_2 status of wastewater. No DO was recorded in site-I and in site-II after stabilization of the effluent, DO was recorded as 6.01 mg L-1. BOD and COD of the raw composite tannery effluent were 1800 mg L-1 and 9886 mg O_2 L-1 respectively, though the BOD value reduced to 499.17 mg L-1 after traversing 1km from the source point on the wastewater carrying canal at site-I, COD value increased to 10903.74 mg $O_2 L^{-1}$. Both BOD and COD of unmixed tannery effluent at site-I reduced over 70.25%and 56.53% at the site-II where the effluent is stabilized for pisciculture, the result is in good conformity with the findings of Chattopadhyay *et al.* (2000). Although BOD_5 values reduced considerably in the course of flowing down the wastewater carrying canal up to site-I or in the stabilization pond, COD values did not reduce appreciably. Since, in tanneries a huge amount of Na_2S is used in the liming process as a depilation agent, spent lime liquor discharged contains a lot of Na_2S. This along with the other inorganic oxidizable matters is responsible for the increment of COD values. Treatment of wastewater with catalytic redox system or recycling in the tannery process will be a useful measure for the efficient COD reduction by nature otherwise it seems to be inefficient when compared to the others measures of oxygen status. According to Ghosh *et al.* (1991), wastewater stabilization ponds of ECW that are being used for pisciculture after stabilization of the wastewater, have been recognized as one of the most efficient and cost effective means of wastewater treatment, where land resources permit and weather conditions are favorable. When chemical parameters are considered, NO_3^{-1} concentration showed a relatively high value (375 mg L^{-1}) in the raw composite tannery effluent, and the value dropped radically to (39.58 mg L^{-1}) as the wastewater flowed down to the waste water carrying canal. In site-II, NO_3^{-1} concentration was found to be 43.58 mg L^{-1}. PO_4^{3-}, on the other hand, though showed a considerable concentration (62.5 mg L^{-1}) in composite effluent, a drastic reduction over 99% occurred in site-I, which was appeared again in site-II (0.87 mg L^{-1}) only after natural stabilization. 64.37% increment of PO_4^{3}- concentration occurred in site-II, allowing the nutrient support of live protoplasm building. Bunce (1995) mentioned that the incorporation of nutrients into biomass is promoted by providing nutrients in the correct proportions i.e. a ratio of C: N: P of about 100:15:1. As carbon is never a limiting factor in wastewater, a suitable N:P ratio (15:1) would be important for uptake of nutrient elements to biomass (Bunce, 1995; Talling and Lamoallae, 1998). However, when ecological importance point is considered, our present study justifies the remark, put forwarded by Chu (1942) and Cairns et al. (1972) that the optimum concentrations of N and P in natural environments is perhaps exception more than rule. The TSS, TDS, Conductivity, total hardness, chlorides and alkalinity values were high in composite tannery effluent. Over 59.62% reduction of total suspended solids between site-I and site-II indicates efficient removal of suspended matters in the process of stabilization in site-II. TSS was positively correlated with Cl- ($r = 0.66$) and COD ($r = 0.53$) and negatively correlated with Alkalinity ($r = -0.51$) at $P<0.05$ (Table 6). TDS decreased to 5618.08 mg L^{-1} at site-I from the raw composite effluent where the concentration was 24.10 g L^{-1}. Over 74% reduction of TDS occurred in site-II from the site-I. The quantity (1450.63 mg l^{-1}) of TDS at site-II is well below the CPCB's limit of 2.1 g L^{-1} (CPCB, 1997). TDS concentration showed a positive correlation with Conductivity ($r = 0.98$) and BOD ($r = 0.43$) at $P<0.05$. Chloride content of the tannery effluent generally remains very high as the tanning processes require the input of wet salted hides and skins, which contain NaCl of about 25-30% of the green weight of the

Table 1: Physico-chemical parameters of raw composite tannery effluent and wastewaters measured at the study sites-I and II. Concentrations are given as (mean ± SD). N=24

Parameters	Composite tannery effluent	Site-I	Site-II
Air Temperature (^0C)	-	32.29±2.7	32.40±2.7
Water Temperature (^0C)	-	31.84±2.97	31.26±4.04
pH	8.63±2.71	7.81±0.63	7.91±0.69
Dissolved Oxygen (mg L^{-1})	Not detected	Not detected	6.01±1.59
Total Dissolved Solid (mg L^{-1})	24100±9.33	5618.08±1464.79	1450.63±685.77
Conductivity (ms)	47.10±20.95	11.80±2.6	2.87±1.39
Total suspended solid (mg L^{-1})	2130±0.91	1343.27±298.29	542.38±378.08
Chloride (mg L^{-1})	14900.00±7143.62	3759.38±1184.83	987.71±695.19
Phosphate (mg L^{-1})	62.50±28.39	0.31±0.11	0.87±0.66
Nitrate (mg L^{-1})	375.00±145.68	39.58±23.31	43.58±29.01
Carbonate Hardness (mg L^{-1})	961.20±509.97	760.31±506.23	449.11±396.51
Total hardness (mg L^{-1})	2136.00±1376.4	974.58±646.45	753.81±325.36
Acidity (mmol L^{-1})	0.78±0.98	1.04±0.69	1.00±0.38
Alkalinity (mmol L^{-1})	6.32±5.12	31.47±21.79	26.72±25.57
Biological Oxygen Demand (mg L^{-1})	1800±565.69	499.17±128.09	148.50±76.81
Chemical Oxygen Demand (mg O_2 L^{-1})	9888±480	10903.74±4175.1	4739.91±4407.61

raw material. Furthermore, a huge amount of NaCl is also consumed by the pre-tanning operations in leather industry. The Cl- content was excessively high (14900 mg L-1) in the composite effluent, whereas it dropped down drastically in site-I (3759.38 mg L-1) and then at site-II (987.71 mg L-1) after stabilization.

Raw composite tanning wastewater contained a significant amount of total chromium (232.33 mg L-1) as Cr was used for the production of high performance leather as an alternative of vegetable tanning nearly 100 years back. Vlimmeren, 1972 reported that a typical tannery discharge may contain 75-200 mg L-1 of total chromium. However he reported that in practice soluble chromium in tannery effluent was always well below 10mg L-1 and this concentration has hardly any effect on the biological system. In the present study, total Cr in the raw effluent dropped to 0.6 mg L-1 after 1 km away from the source point at site-I. Raw composite tannery effluent fed fish tank at site-II contained 0.01 mg L-1 Cr, which is quite negligible and much below the CPCB's standard of 2.0 mg L-1 for inland surface water and public sewer and even below the USEPA (1997) guideline of maximum concentration level for total chromium (with no designation of valence state) in potable water of 0.1mg L-1 (Petura et al. 1999). Five other hazardous metals, viz, Pb, Zn, Fe, Mn and Cu were also studied in the wastewater (table2). Mostly the inorganic pigments, which are used extensively in leather finishing, may be the source of these metals. The Pb concentration in the site-I was found to be 0.70 mg L-1 and in site-II after stabilization it was noted as 0.46 mg L-1, which is considerably higher than the permissible limit of 0.05 mg L-1 (Sittig, 1985). The Fe and Mn concentrations too were much higher than the safe limits of 0.30 mg L-1 and 0.05 mg L-1 respectively (Sittig, 1985). While copper contents were far below the maximum permissible limit set by Indian council of medical Research for drinking water or EPA safe limit of 1.0 mg L-1 (Sittig, 1985).

The present data depicted at Table-3 shows that fishes exposed in 30%, 50% and 100% tannery effluent for 96 hours, were found to have a highly adverse effect on them with 100% mortality in all the cases. Whereas, in other diluted concentrations (10% and 20%), the mortality was about 50% and 75%. Calculated LC_{50} values from six replicates ranges from 10%-17.5% (table 3). 96-h LC_{50} value (13.75%) calculated by the mortality of fishes in each respective concentration after total six replicates also satisfies the obtained result (table 4). In the present acute toxicity experiment, the findings are comparable with what was obtained by Muley et al. (2007), where they reported 96-h LC_{50} concentration of 20% tannery effluent in a fresh water fish *Labeo rohita*. High mortality in higher concentrations may be due to the metal salts present in the effluent along with the other toxic components. It is also seen from the present study that the tannery effluent contains a high quantity of TDS, TSS, COD and BOD which contribute to the organic pollution of a water body. So in higher concentrations, the organic pollutants present there are gradually depleting the oxygen content from the medium, increasing its toxicity (Ravibabu *et al.*, 2007). A correlation matrix was formed at 95% level of significance between parameters and LC_{50} value, where the LC_{50} value showed no significant

Table 2: Elements profile of raw composite tannery effluent and wastewaters measured at the study sites-I and II. Concentrations are given as (mean ± SD). N=24

Elements	Raw tannery effluent	Site-I	Site-II
Chromium (mg L^{-1})	232.33±67.55	0.60±0.25	0.01±0.03
Lead (mg L^{-1})	0.08±0.03	0.70±0.23	0.46±0.12
Copper (mg L^{-1})	Not detected	0.56±0.24	0.66±0.34
Zinc (mg L^{-1})	0.15±0.03	0.26±0.06	0.11±0.06
Manganese (mg L^{-1})	2.09±0.93	1.25±0.32	2.36±1.65
Iron (mg L^{-1})	14.29±6.71	4.14±0.32	2.16±0.76

Table 3: Determination of LC_{50} value of Tannery Effluent by Karber's method using Guppy fish (*Poecilia reticulata*) (Abbreviations: R= Replicate)

Replicate	Group	Conc. In ml/l	No. of animal (n)	Conc. Diff, (a)	Dead	Mean mortality (b)	Product	Calculated LC_{50} value
R_1	1	100	4	0	0	0	0	150ml/l
	2	200	4	100	2	2	200	
	3	300	4	100	4	4	400	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	
R_2	1	100	4	0	0	0	0	125ml/l
	2	200	4	100	3	3	300	
	3	300	4	100	4	4	400	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	
R_3	1	100	4	0	0	0	0	100ml/l
	2	200	4	100	4	4	400	
	3	300	4	100	4	4	400	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	
R_4	1	100	4	0	0	0	0	150ml/l
	2	200	4	100	2	2	200	
	3	300	4	100	4	4	400	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	
R_5	1	100	4	0	0	0	0	175ml/l
	2	200	4	100	2	2	200	
	3	300	4	100	3	3	300	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	
R_6	1	100	4	0	1	1	0	125ml/l
	2	200	4	100	3	3	300	
	3	300	4	100	4	4	400	
	4	500	4	200	4	4	800	
	5	1000	4	500	4	4	2000	

Table 4: Calculated LC_{50} value of tannery Effluent after six replications

Conc. In ml/l	Dead						Conc. Difference (a)	Mean mortality (b)	Product (a*b)	Calculated LC 50 value
	R_1	R_2	R_3	R_4	R_5	R_6				
100	0	0	0	0	0	1	0	0.17	0	137.5ml/l
200	2	3	4	2	2	3	100	2.67	267	
300	4	4	4	4	3	4	100	3.83	383	
500	4	4	4	4	4	4	200	4	800	
1000	4	4	4	4	4	4	500	4	2000	

Table 5: Co-relation matrix of mean values of the elements showing interdependence between them and also with LC_{50} value (Marked correlations are significant at $p < 0.05$) N=24

Cr	Cr						
Pb	0.24	Pb					
Cu	-0.26	0.17	Cu				
Zn	0.58*	0.08	-0.13	Zn			
Mn	-0.13	-0.27	-0.24	-0.02	Mn		
Fe	0.43*	0.17	-0.59*	0.19	0.30	Fe	
LC_{50}	-0.66*	-0.21	0.21	-0.20	0.25	0.01	LC_{50}

correlation with those of the physico-chemical parameters (Table 6). This result is in good agreement with the observations of Yatrabi and Nejmeddine (2000) and Cooman et al (2003), who concluded that it is too difficult to correlate the LC_{50} values with those of the physico-chemical parameters of the effluent in which they were exposed, as the tested effluents are very complex matrices. But only a negative correlation ($r = -0.66$) was found between LC_{50} and Cr at $p<0.05$ (Table 5). This means that LC_{50} value of tannery effluent decreases with an increment in its chromium content, which in turn makes it more toxic. It is evident that higher LC_{50} value is less toxic because greater concentrations are required to produce 50% mortality in organisms. The present findings can be explained by the work of Vutukuru at al. (2007) where they reported adverse pathological alteration in a fresh water fish, *Labeo rohita*, in terms of increased serum amino transferase activity due to an elevated level of chromium in water at 96-h LC_{50} concentration.

Table 6: Co-relation matrix of mean values of the parameters showing interdependence between them and also with LC_{50} value (Marked correlations are significant at $p < .05000$) N=24 (Abbreviations: AT-Air temperature; WT-Water temperature; DO-Dissolved oxygen; COND-Conductivity; ACID-Acidity; ALK-Alkalinity; TH-Total Hardness; CHL-Chloride)

AT	AT													
WT	0.87*	WT												
PH	-0.24	-0.42*	PH											
TDS	-0.33	-0.39	0.14	TDS										
COND	-0.31	-0.40	0.18	0.98*	COND									
TSS	0.36	0.22	0.21	0.16	0.24	TSS								
ACID	-0.27	-0.18	0.41*	-0.05	-0.08	-0.02	ACID							
ALK	-0.31	-0.28	0.39	0.03	0.00	-0.51*	0.44*	ALK						
TH	-0.01	0.19	0.12	0.07	0.07	0.20	0.37	0.02	TH					
CHL	0.27	0.11	0.46*	-0.27	-0.16	0.66*	0.24	0.02	0.03	CHL				
NO_3^{1-}	-0.01	0.05	-0.21	-0.13	-0.09	0.03	-0.08	-0.24	-0.30	-0.13	NO3			
BOD	-0.43*	-0.37	0.11	0.43*	0.46*	0.30	-0.29	-0.18	0.04	0.11	-0.26	BOD		
COD	-0.09	-0.23	0.69*	0.11	0.17	0.53*	0.34	-0.18	0.18	0.52*	-0.23	0.26	COD	
LC_{50}	0.04	0.10	-0.01	0.00	0.02	-0.19	-0.24	0.20	-0.22	0.12	-0.19	0.04	-0.15	LC_{50}

CONCLUSIONS

This study shows the amelioration of exceedingly high concentration of critical toxic pollutants like TSS, TDS, BOD, total hardness in raw composite tannery effluent after natural stabilization in a wastewater fed productive fish pond. It is evident from our study that an ideal wastewater fed fish pond should have an average 70 times mixing condition as the 96hr-LC_{50} (137.5 ml L-1) mixing ratio was 1:7 for composite tannery effluent and freshwater respectively. Our present findings signifies the fact that those wastewater fed fish ponds in East Calcutta Wetland ecosystem satisfies the truth and has been proved to be highly productive. Though certain control measures like removal of salt crystals before soaking, recycling of chrome-liquor, minimum use of Pb based pigments in leather finishing, treatment of wastewater with catalytic redox for reduction of COD etc. should be taken. Over the past decades, however, more than 30% of this wetland has been reclaimed under the pressure of rapid urbanization. Care has to be taken to stop further encroachment on this efficient, cost effective natural stabilization area. The result of the present study amply justifies the reasons for sustainability of this traditional resource recovery system practiced in ECW for the last ten decades or so.

ACKNOWLEDGEMENTS

Authors would like to express their earnest gratitude to the Center Director, UGC-DAE consortium for scientific research, Kolkata Center, Salt Lake City, Kolkata-700098 for his Co-operation and inspiration, including a research fellowship. Authors also express gratitude to the AICTE, Director of Technical Education and the Director of Public Instruction, Govt. of West Bengal, India for co-operation and necessary support.

REFERENCES

1. APHA (American Public Health Association). 1995. Standard methods of the examination of water and wastewater, 19th edition. APHA, Washington D.C. 2. Bunce, N.J. 1995. Aspects of the environmental chemistry of the elements. In: Encyclopaedia of Environmental Biology, Academic Press Inc., N.Y. I, pp. 105-138.
3. Cairns J.Jr. G.R. Lanza and B.C. Parker. 1972. Pollution related structural and functional changes in aquatic communities with emphasis on freshwater algae and protozon, Proc. Acad. Nat. Sci. Philadelphia. 124(5), pp.79-127.
4. Chattopadhyay, B., R. Gupta and S.K. Mukhopadhyay,. 2000. Physico-chemical and biological characterization of effluent of Calcutta tanneries. B.B. jana, R.D. Banerjee, B. Guterstam and J. Heeb (Eds). University of Kalyani, India and international Ecological Engineering Soceity, Switzerland. Waste Recycling and resource Management in the Developing world. Pp.361-370.
5. Chu S.P. 1942. The influence of the mineral composition of the medium on the growth of planktonic algae. I. methods and culture media. J. Ecol., 31 pp. 109-148.
6. Cooman, K., M. Gajrado, J. Nieto, C. Bornhardt, G. Vidal, 2003 Tannery waste

water characterization and toxicity effects on Daphnia Spp, Environmental toxicol. 18, pp. 45-51.

7. CPCB. 1997. Standards for liquid effluents, gaseous emissions, automobile exhaust. Noise and ambient air quality. Central Pollution Control Board, Parivesh Bhawan. Delhi.
8. Ghosh D. 1991. Sustainable development or real state take-over? A report, WWF. Calcutta, pp 24.
9. Ghosh, M.N. 1984. Fundamentals of experimental Pharmacology (2nd edition), Scientific book agency, Calcutta.