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Study of Seasonal Variations in Oxygen Consumption of Estuarine Clam, *Meretrix Meretrix* (Linnaeus, 1758) after Acute Exposure of Cadmium Chloride

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ABSTRACT

The estuarine clam, *Meretrix meretrix* was exposed to predetermined seasonal sublethal and lethal concentrations of $CdCl_2$, 2½ H_2O for 96 hrs. Experiments were conducted during summer, monsoon and winter by keeping control group of clams. Estuarine water parameters like temperature, pH, salinity, rainfall and dissolved oxygen were recorded. In the present study, it is found that, it has significant influence on rate of oxygen consumption and toxicity of cadmium chloride. During summer, clams from LC_0 and LC_{50} group were treated with 1.1ppm and 1.8 ppm respectively. During monsoon LC_0 and LC_{50} group were treated with 1.6 ppm and 2.0 ppm respectively. During winter clams from LC_0 and LC_{50} group were exposed to 1.4 ppm and 2.1 ppm cadmium chloride respectively. During summer, as compared to control group, there were 3.83, 17.04, 16.77 and 10.63% increase in oxygen uptake at the end of 24, 36, 48, and 60 hrs. There were 0.35, 4.97 and 21.75% decrease at the end of 48, 72, 84 and 96 hrs. Moreover, similar trend of oxygen consumption was observed in LC_0 and LC_{50} group of clams in winter and monsoon season. During monsoon and winter clams from control group showed similar trend of oxygen uptake with less significant fluctuations. Clams from control group and LC_0 and LC_{50} group showed less oxygen consumption during monsoon than summer and winter.

KEYWORDS: *Meretrix meretrix, Cadmium chloride, Oxygen consumption, estuarine water parameters*

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INTRODUCTION

Rapid increase in population and industrialization along the bank of Bhatye estuary, Ratnagiri has started threatening the quality of estuarine water and life of iota. One of the recent trend is construction of tourist resorts along the bank of estuary and backwaters. Lot of waste, sewage .untreated domestic and industrial effluents is continuously dumping in estuarine water. This waste is impregnated with biological and chemical load. Bhatye estuary is a cradle bed for many marine and estuarine animals like clams. Estuarine clam, *M. meretrix* is abundantly found in Bhatye estuary. During lean period of open sea fishery, it provides protein rich food and livelihood to local population. Many scientists have worked on bioaccumulation capacity of heavy metals (Greig et al., 1975[1]; Fretter, J. M., 1976[2]; Hung, T. C., 1982[3]; Krishnakumar, 1989[4]; Joseph and Shrivastava, 1993[5]); *M.meretrix* has great capacity of cadmium accumulation and it showed variance of cadmium accumulation in soft tissues (Kumbhar, Sanjay, 2001)[6] ;. Due to its bioaccumulation capacity these metals are returning back to the human body. Clams are good bioindicators of heavy metal pollution. Many researchers have stressed the need of indicator organisms which could be employed to monitor environmental contamination by heavy metals. (Preston, et al. 1972[7]; Haug, et al. 1974[8]; Goldberg.et al., 1975[9]; Preston, et al., 1977[10]) believed that, bivalves are perhaps the best indicator over a wide variety of environmental conditions. Measurement of oxygen consumption is a very important tool to access the toxicity stress on aquatic organisms, since it is also an index of energy expenditure to fulfill the demands of environmental and biological alterations. Utilization of oxygen is, therefore, a direct measurement of degree of activity, food conversion and heat production (Bishop, 1950)[11] .The process of respiration is sensitive to whole array of environmental and Biological alterations.

Material and Method:

Water samples were collected by dipping plastic jar below 30cm to avoid floating water. It is emptied in previously cleaned and deionised cans. The water samples were filtered and 0.45cm filter paper. The water samples were used for detection of temperature, pH and salinity. Rainfall data was obtained from Government Meteorology Department. Clams of median size (4.0 to 4.8 and 3.8 to 4.8cm in length) were collected and brought to the laboratory. Clams were acclimatised for 96hrs. Acclimatised clams were used to conduct bioassay tests and further experiments. The toxicity tests were repeated three times and LC_0 and $LC5_0$ values were determined for each season. Clams were exposed to predetermined concentrations of cadmium chloride in different season. Oxygen consumption experiments were performed in specially designed glass jars of one litre fitted with rubber tubes. The marked clams were kept for one hour. Dissolved Oxygen was determined by Winklers method .The rate of oxygen consumption of LC_0 and LC_{50} groups along with control after every 12 hrs time interval was determined. The experiments were repeated for three times for confirmation of results. Such experiments were conducted during summer, monsoon and winter of the year 2017.

Results and discussion

Table1. The water parameters of Bhatye Estuary, Ratnagiri (2017–18)

Season	Surface Water Temperature.	Rainfall (mm)	Salinity (mg/l)	pH	Dissolved Oxygen (ml/l)
Summer (April-May)	Avgas- 26.9	-	35.2	8.1	3.5
Monsoon (July- August)	26.1	34.39	5.2	7.29	5.0
Winter (Nov- Dec)	25.5	5.2	30.36	8.5	4.3

In the present study, it was found that during summer and winter, temperature was higher as compared to monsoon. Influx of freshwater through rivers and intrution of sea water through the lower reaches have profound influence on the distribution of temperature in backwater systems (Pillai, et al., 1975)[12]. It was observed that, temperature, oxygen saturation, salinity are interrelated parameters. Temperature was directly proportional to salinity and inversely proportional to dissolved oxygen, it affects oxygen concentration in water. At Ratnagiri, the rainfall was maximum in the month of July (34.39) but later on it decreased. There was scanty rainfall (0.006) in winter and no rainfall in summer. Maximum average salinity was recorded in summer (35.2). It was 30.36mg/l in winter. Minimum average salinity was recorded in monsoon season (5.2). The bioaccumulation and toxicity of metals increases with increase in temperature. The absorption and release of metals can also depend on temperature. This was established for mercury, methyl mercury and phenyl acetate in rainbow trout, *Salmo guardeneri*. (Mc Leod, 1973)[13].

Salinity is key trigger of other environmental factors. It depends on certain factors as local precipitation, water influx, mixing of fresh water with sea water and evaporation. Hence, salinity is more labile parameter than any other estuarine water parameter. In the present study, it varied according to season. It was maximum (35.2) in summer and minimum (5.2) in monsoon. Salinity is key trigger of other environmental characteristics. It depends on certain factors as, local precipitation, water influxes, mixing of seawater with fresh water and evaporation. In the present investigation seasonal fluctuations were quiet well marked. Low salinity was observed during monsoon, due to heavy rainfall in the basin of Kajali river, which causes dilution. Similar observations were also made by other workers (Ramamurti, 1953[14]; Nixon, 2001[15]; Loankar, 1971[16])

The values of oxygen saturation varied seasonally in the estuarine waters. In general, during monsoon, saturation was better than those of summer and winter. It can be attributed to turbulence, freshwater influx and temperature. Similar observations are noted by Imley, A. J., 1968[17]; Keller, A. E., 1991[18]; Lee. H., 1991[19].

The average pH was 8.1, 7.29 and 8.E’E.5 in summer, monsoon and winter respectively. The average dissolved oxygen was 3.5, 5.0 and 4.3 in summer, monsoon and winter respectively During monsoon estuarine water shoed maximum saturation of oxygen (5.0). It was minimum (3.5) in summer variations in pH did not show significant seasonal pattern. However, the pH values were low during monsoon due to heavy influx of freshwater. Similar observations were noted by Chandran and Ramamurti (1984)[20] in velar estuary .The hydronomics of an estuary in general is very complex because of the interdependable variables like tide, river discharge, and density difference in the water masses in the estuary, salinity and temperature. In the present investigation it is found that these interrelated estuarine water parameters were highly liable and had profound influence on oxygen consumption of. *M. meretrix.*

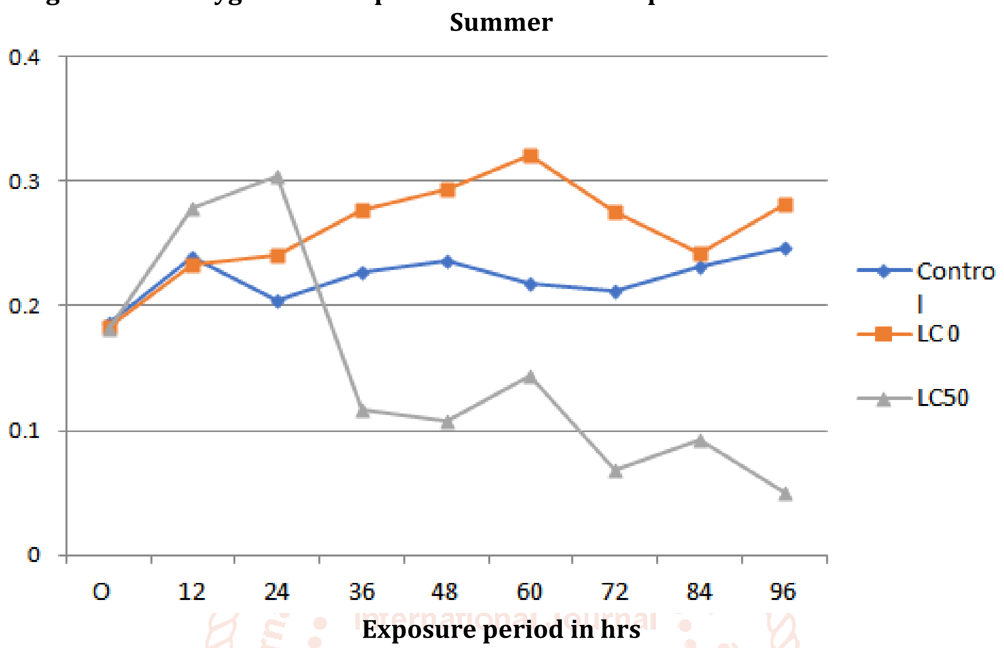
Table 2: Rate of oxygen consumption (ml/l/hr/gm wet wt) in *M.meretrix* exposed to different concentrations of cadmium chloride after acute exposure.

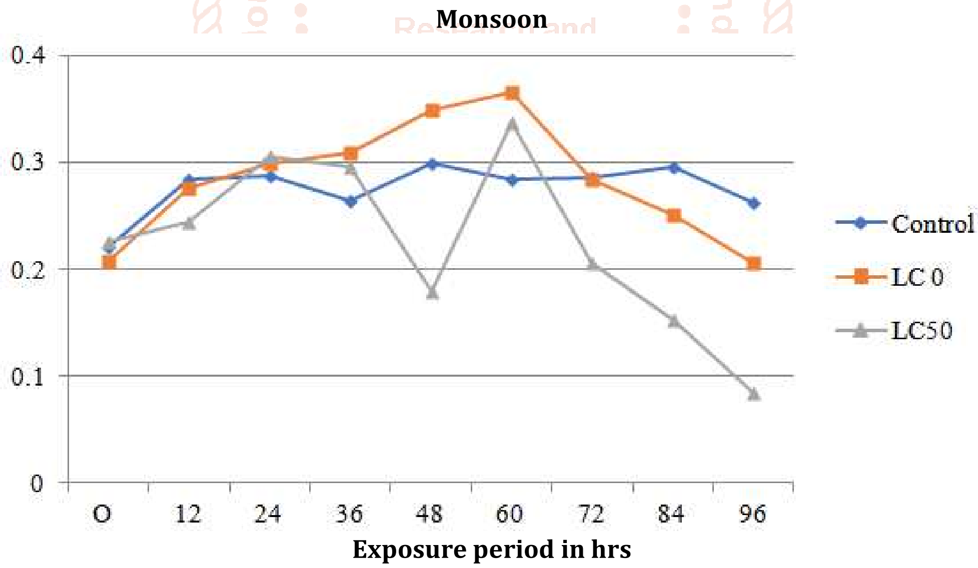
	Group	Exposure Time								
		0 Hrs	12 Hrs	24 Hrs	36 Hrs	48 Hrs	60 Hrs	72 Hrs	84 Hrs	96 Hrs
Summer	Control	0.220 ±0.013	0.283 ±0.061	0.287 ±0.004	0.264 ±0.038	o.298 ±0.01	0.283 ±0.012	0.285 ±0.026	0.295 ±0.004	0.262 ±0.012
	LC0	0.208 ±0.007	0.275 ±0.013 (-2.82)	0.298 ±0.005 (3.83)	0.309 ±0.063 (17.04)	0.348 ±0.013 (16.77)	0.365 ±0.017 (10.63)	0.284 ±0.032 (-0.35)	0.251 ±0.014 (-4.97)	0.205 ±0.023 (-21.75)
	LC50	0.225 ±0.051	0.244 ±0.057 (-13.78)	0.305 ±0.068 (6.27)	0.295 ±0.025 (11.74)	0.179 ±0.004 (-39.93)	0.337 ±0.06 (20.37)	0.205 ±0.02 (39.02)	0.153 ±0.012 (-18.13)	0.084 ±0.005 (-25.89)
Monsoon	Control	0.186 ±0.021	0.24 ±0.015	0.205 ±0.016	0.227 ±0.071	0.237 ±0.005	0.218 ±0.02	0.212 ±0.008	0.231 ±0.078	0.247 ±0.022
	LC0	0.183 ±0.055	0.233 ±0.065 (-2.91)	0.241 ±0.01 (17.56)	0.277 ±0.049 (22.02)	0.294 ±0.036 (24.05)**	0.321 ±0.004 (47.24) **	0.276 ±0.01 (30.18) **	0.243 ±0.069 (5.19)	0.281 ±0.064 (-11.74)
	LC50	0.182 ±0.007	0.279 ±0.025 (-4.12)	0.305 ±0.009 (0.99)	0.117 ±0.088 (-58.65)	0.108 ±0.04 (-62.75)**	0.144 ±0.076 (-45.24)	0.068 ±0.01 (-74.81)***	0.093 ±0.021 (-64.78)	0.051 ±0.014 (-71.71)***

Winter	Control	0.197 ±0.073	0.258 ±0.07	0.243 ±0.017	0.229 ±0.024	0.250 ±0.032	0.230 ±0.019	0.263 ±0.013	0.260 ±0.045	0.237 ±0.015
	LC0	0.193 ±0.018	0.239 ±0.098 (-6.37)	0.283 ± 0.049 (16.46)	0.322 ±0.006 (40.61)**	0.366 ±0.021 (46.40)**	0.385 ±0.055 (67.69)*	0.378 ±0.019 (43.72)**	0.341 ±0.064 (31.15)	0.317 ±0.095 (33.75)
	LC50	0.184 ±0.049	0.261 ±0.005 (1.16)	0.305 ±0.065 (25.51)	0.213 ±0.016 (-6.98)	0.141 ±0.045 (-43.60)**	0.205 ±0.018 (-10.86)	0.243 ±0.052 (-7.60)	0.210 ±0.071 (-7.31)	0.136 ±0.009 (-42.61)***

Values in parenthesis are percent change, ± = S.D. of five animals (*=p < 0.05, * *= p < 0.01, *** = P < 0.001).

Fig.1. Rate of oxygen consumption of *M. meretrix* exposed to cadmium chloride





Oxygen consumption is measure of animals overall energy demand under particular conditions, at which the measurements are made. The process of respiration is sensitive to whole array of environmental as well as biological variables. Oxygen consumption is useful measure to assess the sub lethal effects of xenobiotics, as energy processes serves as an indicators of overall physiological state (Sigmund, 1979)[21] .During summer .In control group, the rate of oxygen consumption fluctuated between 0.220 and 0.298 ml/l/g/h from 0 to 96 h. In LC_0 (1.1 ppm) group, the rate of oxygen consumption fluctuated between 0.208 to 0.365 ml/g/h from 0 to 96 h. From 0 to 60 h, there was considerable increase from 0.208 to 0.365 ml/l/g/h. From 72 h to 96 h, the rate of oxygen consumption decreased from 0.284 to 0.205 ml/l/g/h. As compared to control, there was 3.83, 17.04, 16.77 ($p<0.001$), and 10.63% increase in oxygen uptake at the end of 72, 84, and 96 h respectively. In LC_{50} (1.8 ppm) group, there was considerable increase from 0.225 to 0.305 ml/g/l/h, at the end of 24h. At 36 and 48 h oxygen consumption was decreased up to 0.295 and 0.179 ml/l/g/h respectively. From 60 to 96 h, this rate was considerably decreased from 0.337 to 0.084 ml/l/g/h. In LC_{50} group, compared to control, there is a significant ($p<0.001$) decrease of 39.93, 39.02, 48.13 and 25.89% at the end of 48, 72, 84, 96 h, while 6.27, 11.74, 20.37% ($p<0.001$) increase in oxygen uptake at the end of 24, 36, 60 h respectively

During monsoon, in control group, the rate of oxygen consumption fluctuated between 0.186 to 0.247 ml/l/g/h. In LC_0 (1.3 ppm) group, rate of oxygen consumption fluctuated between 0.183 to 0.321 ml/l/g/h, from 0 to 60 h. From 72 to 84 h, it was decreased from 0.276 to 0.243 ml/l/g/h, again, there was considerable increase at 96 h (0.281 ml/l/g/h). As compared to control group, there was 17.56, 22.02, 24.05 ($p<0.01$), 47.24 ($p<0.01$), 30.18 ($p<0.01$) and 5.19% increase in oxygen uptake at the end of 24, 36, 48, 60, 72 and 84h respectively. There was 2.91 and 11.74% no significant decrease in oxygen uptake at the end of 12 and 96 h respectively. In LC_{50} (2.0 ppm) group, rate of oxygen consumption fluctuated between 0.051 and 0.305 from 0 to 96 h. From 0 to 24 h, rate of oxygen consumption increased from 0.182 to 0.305 ml/l/g/h. From 24 h onwards, it was decreased up to 0.108 ml/l/g/h at the end of 48 h. From 24 h onwards, it was decreased up to 0.108 ml/l/g/h at the end of 48 h. From 60 h, it was sharply decreased from 0.144 to 0.051 at the end of 96 h. In LC_{50} group, compared to control, there was 4.12, 58.65, 63.75 ($P<0.01$), 45.24, 74.81 ($p<0.001$), 64.78 and 71.71% ($p<0.001$) decrease at the end of 12, 36, 48, 60, 72, 84 and 96 h respectively. There was no significant increase of 0.99% at the end of 24 h.

During winter, control group of clam showed fluctuations in rate of oxygen consumption between 0.197 to 0.073 ml/l/g/h. In LC_0 (1.5 ppm) group, rate of oxygen consumption fluctuated between 0.193 to 0.385 ml/l/g/h from 0 to 96 h. There was considerable increase from 0.193 to 0.385 ml/l/g/h at the end of 60h. From 72 to 96 h, there was considerable decrease from 0.378 to 0.317 ml/l/g/h. In LC_{50} group, compared to control, clams consumed 16.46, 40.61 ($p<0.01$), 46.40 ($p<0.01$), 67.69 ($P<0.05$), 43.72 ($p<0.01$), 31.15 and 33.75% more oxygen at the end of 24, 36, 48, 60, 72, 84 and 96 h respectively. There was 6.37% no significant decrease at the end of 12 h. In LC_{50} (2.1ppm) group, rate of oxygen consumption fluctuated between 0.136 to 0.305 ml/l/g/h. There was considerable increase from 0.164 to 0.305 ml/l/g/h from 0 to 24 h. At 36 h, clams consumed 0.213, 0.141 and 0.205 ml/l/g/h at the end of 36, 48, and 60 h respectively. From 72 h onward, rate of oxygen consumption decreased from 0.243 to 0.136 ml/l/g/h at the end of 96 h. As compared to control, there was 6.98, 43.60 ($p<0.01$), 10.86, 7.60, 7.31 and 42.61% ($p<0.001$) decrease .In the present study it is found that clams from control group consumed more oxygen in summer and less in monsoon .The fluctuations in the estuarine environment where clams are found are therefore bound to influence the oxygen consumption correlated with estuarine environment. In the present study, it was found that clams from control group showed fluctuations in oxygen consumption, but fluctuations were more during monsoon. The salinity and temperature data (Table 1) shows that, during monsoon, when river enters the estuary, salinity decreases due to freshwater influx. It has been observed that, the rate of water filtration and metabolic activity decreases during monsoon because for the majority of period, values of clams remained closed in low salinity .In winter, oxygen consumption was less than in summer. It may be due to presence of optimum range of water parameters and physiological status of clams at that time. During summer, clams exhibited higher degree of oxygen consumption. It may be due to depleted oxygen content in water and cadmium induced stress. (Isono. Raisuke et al., 1998)[22] report that, in Japanese little neck clam *ruditapes philippinarium,* the oxygen rate decreased markedly above 40^0 C. Clams suffer from thermal stress over 25^0 C and has significant mortality at around 34^0 C within a few days and no heat resistance over 40^0 C. (Hare et al., 1998)[23] observed enhanced oxygen consumption rate in flat oyster *Ostrea edulis* during summer (Young and Shim- Ieong- Her, 1998)[24] observed increased rate of oxygen consumption during summer in the Antartic clam, *Laternula ellipitica* .Similar observations were made by (Ranade, 1970)[25] in case of clams from Ratnagiri waters.

As compared to summer and winter, in monsoon, control group of clams showed decreased oxygen uptake. Clams closed their shell valves for most of the time to avoid unfavourable estuarine water parameters. Similar observations were noted by (Ranade, 1973)[26] .As compared to control group of clams, LC_{50} group showed decreased oxygen uptake in 3 different seasons. The observed decrease is attributed to variance in the volume of water ventilated through the gills, caused by the intermittent closure and opening of shell valves. Here the main factor which caused oxygen decreased uptake was due to coagulation of mucus on gills due to cadmium exposure. Coagulation of mucus causes reduction in effective transport of oxygen to internal tissues. Coagulation film anoxia adversely affects the absorption of oxygen from the ambient medium. (Brown and Nevel, 1972)[27] have suggested that, reduced oxygen uptake maybe due to reduced energy requirement caused by suppression of ciliary activity in the present study; considerable mucus secretion was found in the clams from LC_0 and LC_{50} group. These fluctuations in oxygen uptake in *M.meretrix* are due to cadmium chloride stress and estuarine water parameters.

Conclusion

The present study gives a detailed account of impact of $CdCl_2$ stress on oxygen consumption of clams from control, LC_0 and LC_{50} group. It reveals interaction between seasonal estuarine water parameters n consumption. Experimental results showed that, clams from control group consume more oxygen in summer and less in monsoon. It is attributed to seasonal estuarine water parameters like temperature, pH, oxygen, saturation and salinity. Clams from LC_0 and LC_{50} group showed marked initial increase and later significant decrease at the end of 96 h. This trend was similar in all three different seasons. It is clear that these fluctuations were due to $CdCl_2$ stress and not due to seasonal estuarine water parameters.

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