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Heavy Metal Toxicity to Fiddler Crabs, *Uca annulipes* Latreille and *Uca triangularis* (Milne Edwards): Respiration on Exposure to Copper, Mercury, Cadmium, and Zinc

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Respiration of animals is considered to be an indicator of their metabolic index (Wolvekamp and Waterman 1960) and is used to evaluate the effect of stress (Krishna Rao 1982) or toxic substances in the environment (Thurberg *et al.* 1977). In an earlier investigation (Uma Devi 1987), an attempt was made to study the toxic effects of four heavy metals viz., Cu, Cd, Hg and Zn on fiddler crabs *Uca annulipes* and *Uca triangularis* obtained from polluted (Viskhapatnam Harbor) and unpolluted (Bhimilipatnam) environs. The present paper deals with respiratory metabolism of fiddler crabs *Uca annulipes* and *Uca triangularis* of Visakhapatnam Harbor by exposing them to different test concentrations of heavy metals i.e. Cu, Cd, Hg and Zn as these are the chief pollutants in their environment. A comparison has also been made with the respiratory metabolism of their counterparts collected from an unpolluted environment i.e. Bhimilipatnam.

MATERIALS AND METHODS

Experimental animals of *U. annulipes* and *U.triangularis* of almost the same size (carapace width: 24-29 mm) were obtained from Visakhapatnam Harbor (Lat 17°41'N and Long 83°20' E) and Bhimilipatnam (Lat 17°54' N and Long 83°28' E). The former is a polluted environment receiving the let-off from the nearby industries, whereas the latter is an unpolluted environment without any industrial pollution (Uma Devi 1987). They were maintained in the laboratory for 48 hr before use. The temperature ($29^{o} \pm 1^{o}$C) and salinity (20 $^{o}/_{oo}$) were monitored throughout the experiment. All the experiments were conducted only in the same period of the day, as these fiddler crabs show a day and night rhythm (Prasada Rao *et al.* 1980; Prasada Rao and Krishna Rao 1982).

Respiratory measurements were made by adopting the method of Ganapati and Prasada Rao (1960) using Winkler's technique (Welsh and Smith 1953). The sea water (20 $^{o}/_{oo}$) was filtered with Whatman 42 filter paper and the test concentrations were prepared by dissolving the different quantities of salts as described earlier (Uma Devi 1987). These test concentrations represent the LC_0, LC_{25}, LC_{50}, LC_{75} for 96 hr and 48 hr of *U. annulipes* and *U. triangularis* (Uma Devi 1987). They are 6.42, 10.17, 12.82, 16.17 ppm

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of Cu; 0.31, 1.33, 2.75, 5.69 ppm of Hg; 29.18, 40.76, 48.21, 57.07 ppm of Cd; 55.69, 69.04, 76.95, 85.74 ppm of Zn for 96 hr and 9.02, 13.23, 16.06, 19.47 ppm of Cu; 0.72, 2.73, 5.35, 10.46 ppm of Hg; 32.86, 44.94, 52.64, 61.63 ppm of Cd; 66.4, 76.05, 81.43, 87.2 ppm of Zn for 48 hr of *U. annulipes* of Visakhapatnam Harbor. For *U. annulipes* of Bhimilipatnam, the test concentrations are 0.50, 3.53, 9.42, 25.18 ppm of Cu; 0.014, 0.038, 0.064, 0.016 ppm of Hg; 3.33, 9.43, 15.91, 26.86 ppm of Cd; 13.74, 24.06, 31.93, 42.35 ppm of Zn for 96 hr and 0.87, 5.51, 21.59, 84.5 ppm of Cu; 0.037, 0.072, 0.1, 0.141 ppm of Hg; 4.3, 13.58, 24.22, 43.21 ppm of Cd; 12.93, 27.69, 40.64, 59.67 ppm of Zn for 48 hr. 9.17, 12.61, 14.81, 17.39 ppm of Cu; 0.59, 1.48, 2.33, 3.69 ppm of Hg; 31.12, 38.73, 43.23, 48.26 ppm of Cd; 24.42, 47.5, 66.42, 92.87 ppm of Zn for 96 hr and 9.6, 14.39, 17.64, 21.63 ppm of Cu; 0.88, 3.13, 5.91, 11.14 ppm of Hg; 26.38, 43.39, 55.75, 71.60 ppm of Cd; 55.76, 74.35, 85.96, 99.38 ppm of Zn for 48 hr represent the test concentrations of *U. triangularis* of Visakhapatnam Harbor. The test concentrations of *U. triangularis* of Bhimilipatnam are 1.5, 4.67, 8.28,14.68 ppm of Cu; 0.025, 0.05, 0.072, 0.103 ppm of Hg; 0.028, 2.54, 7.66, 23.13 ppm of Cd; 19.4, 30.89, 39.05, 49.38 ppm of Zn for 96 hr and 3.41, 8.42, 13.27, 20.92 ppm of Cu; 0.032, 0.088, 0.147, 0.243 ppm of Hg; 1.46, 7.15, 15.91, 35.44 ppm of Cd; 20.65, 35.25, 46.13, 60.39 ppm of Zn for 48 hr. Oxygen consumption was measured initially without any toxic substance (control) and then these estimations were made at the respective test concentrations separately. The animals were subjected to each concentration for one hour and the acute effect of these heavy metals was studied. Hourly samples were taken for oxygen estimation and the results were presented as ml O_2/hr. At the end of the experiment, the crabs were put in boiled water and then dried in an oven at 90^oC for 48 hr. The dry weights of the crabs were taken and they ranged from 1.3177 to 1.4881 g. 't' test was carried out to compare the control values of respiration with those of exposed values.

RESULTS AND DISCUSSION

The effect of different test concentrations of Cu, Cd,Hg and Zn on oxygen consumption is presented in table 1 for *U. annulipes* and table 2 for *U.triangularis*. It is clear from the results that all these heavy metals are found to inhibit the rates of respiration, but the degree of inhibition varies from metal to metal. From the percentage decrease values, it is evident that the effect of heavy metals is more on *U. annulipes* and *U. triangularis* of Bhimilipatnam than their counterparts of Visakhapatnam Harbor (Tables 1 & 2). Similar results were obtained when their mortalities were studied in both the species from two habitats exposed to the above four heavy metals (Uma Devi 1987). The effect of heavy metals on oxygen consumption in both species of crabs was in the order of Hg>Cd>Cu>Zn. The decrease in the respiratory rates of both species was observed to be very high when exposed to different concentrations for 48 hr than for 96 hr (Tables 1 & 2). However, the effect of LC_0 and LC_{25} of 96 hr was not significant ($p>0.01$) (Tables 1 & 2) in both the species for all four metals investigated. Similar inhibition in gill respiration was reported in green crabs *Carcinus maenas* (Thurberg *et al* 1973) When subjected to cadmium. The respiration of *Uca pugilator* was also found to show inhibition when exposed to mercury (Vernberg and Vernberg 1972). While studying heavy metal pollution in shore crabs, *Carcinus maenas*, Depledge (1984) noticed a disruption in the circulatory and respiratory activity following exposure to mercury and copper. De Coursey and Vernberg (1972) reported a decreased metabolic rate in *Uca* larvae on

Table 1. Effect of Cu, Hg, Cd and Zn on the respiration of *Uca annulipes* from Visakhapatnam Harbor and Bhimilipatnam. Values are presented as mean ± standard deviation and percentage decrease over control. n = 5. * $P < 0.01$

Heavy metal	Visakhapatnam Harbor Test Conc. 96 hr (ppm)	Oxy. Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy. Cons. ml O_2/hr	Bhimilipatnam Test Conc. 96 hr (ppm)	Oxy. Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy. Cons. ml O_2/hr
Cu								
Cont.	0	0.3598 ± 0.0111	0	0.3686 ± 0.0089	0	0.3357 ± 0.0101	0	0.3428 ± 0.0084
LC_0	6.42	0.3359 ± 0.0089 6.64	9.02	0.2376 ± 0.0064* 35.54	0.50	0.2562 ± 0.0096* 23.68	0.87	0.1697 ± 0.0092* 50.50
LC_{25}	10.17	0.3009 ± 0.0193 16.37	13.23	0.2258 ± 0.0118* 38.74	3.53	0.2057 ± 0.0106* 38.73	5.51	0.1203 ± 0.0113* 64.91
LC_{50}	12.82	0.2160 ± 0.0101* 39.97	16.06	0.1890 ± 0.0106* 48.73	9.42	0.1801 ± 0.0135* 46.35	21.59	0.0945 ± 0.0204* 72.43
LC_{75}	16.17	0.2023 ± 0.0076* 43.77	19.47	0.1021 ± 0.0125* 72.30	25.18	0.1592 ± 0.0263* 52.58	84.50	0.0706 ± 0.0252* 79.40
Hg								
Cont.	0	0.3755 ± 0.0091	0	0.4146 ± 0.0112	0	0.4042 ± 0.0167	0	0.3894 ± 0.0067
LC_0	0.31	0.3486 ± 0.0072* 7.16	0.72	0.2884 ± 0.0101* 30.44	0.014	0.3073 ± 0.0099* 23.97	0.037	0.2353 ± 0.0109* 39.57
LC_{25}	1.33	0.3368 ± 0.0107 10.31	2.73	0.2545 ± 0.0099* 38.62	0.038	0.2852 ± 0.0101* 29.44	0.072	0.1884 ± 0.0209* 51.62
LC_{50}	2.75	0.2007 ± 0.0059* 46.55	5.35	0.1823 ± 0.0073* 56.03	0.064	0.1739 ± 0.0209* 33.00	0.100	0.1113 ± 0.0112* 71.42
LC_{75}	5.69	0.1780 ± 0.0313* 52.60	10.46	0.0787 ± 0.0209* 81.02	0.106	0.1502 ± 0.0211* 62.84	0.141	0.0589 ± 0.0105* 84.87

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Table 1 contd.....

Heavy metal	Visakhapatnam Harbor Test Conc. 96 hr (ppm)	Oxy. Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy. Cons. ml O_2/hr	Bhimilipatnam Test Conc. 96 hr (ppm)	Oxy. Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy. Cons. ml O_2/hr
Cd								
Cont.	0	0.3508 ± 0.0076	0	0.3545 ± 0.0098	0	0.3646 ± 0.0086	0	0.3951 ± 0.0101
LC_0	29.18	0.3246 ± 0.0104 7.47	32.86	0.2886 ± 0.0104* 18.59	3.33	0.3115 ± 0.0113* 14.56	4.30	0.3048 ± 0.0122* 22.85
LC_{25}	40.76	0.3137 ± 0.0112 10.58	44.94	0.2218 ± 0.0141* 37.43	9.43	0.2507 ± 0.0089* 31.24	13.58	0.2110 ± 0.0134* 46.60
LC_{50}	48.21	0.2396 ± 0.0205* 31.70	52.64	0.1616 ± 0.0167* 54.41	15.91	0.1999 ± 0.0106* 45.17	24.22	0.1701 ± 0.0202* 56.72
LC_{75}	57.07	0.1813 ± 0.0210* 48.32	61.63	0.0840 ± 0.0235* 76.30	26.86	0.1502 ± 0.0216* 58.30	43.21	0.0689 ± 0.0111* 82.56
Zn								
Cont.	0	0.3864 ± 0.0100	0	0.4020 ± 0.0112	0	0.3746 ± 0.0126	0	0.4083 ± 0.0101
LC_0	55.69	0.3646 ± 0.0099 5.64	66.40	0.3246 ± 0.0096* 19.25	13.74	0.3427 ± 0.0112 8.52	12.93	0.3048 ± 0.0069* 25.35
LC_{25}	69.04	0.3443 ± 0.0115 10.90	76.05	0.3115 ± 0.0108* 22.51	24.06	0.2764 ± 0.0176* 26.21	27.69	0.2507 ± 0.0202* 38.60
LC_{50}	76.95	0.2964 ± 0.0068* 23.29	81.43	0.2396 ± 0.0087* 40.40	31.93	0.2543 ± 0.0128* 32.11	40.64	0.1865 ± 0.0165* 54.32
LC_{75}	85.74	0.2329 ± 0.0103* 39.73	87.20	0.1865 ± 0.0209* 53.61	42.35	0.1889 ± 0.0142* 49.57	59.67	0.1616 ± 0.0139* 60.42

Table 2. Effect of Cu, Hg, Cd and Zn on the respiration of *Uca triangularis* from Visakhapatnam Harbor and Bhimilipatnam. Values are presented as mean ± standard deviation and percentage decrease over control. n = 5 * $P < 0.01$

Heavy metal	Visakhapatnam Harbor Test Conc. 96 hr (ppm)	Oxy.Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy.Cons. ml O_2/hr	Bhimilipatnam Test Conc. 96 hr (ppm)	Oxy.Cons. ml O_2/hr	Test Conc. 48 hr (ppm)	Oxy.Cons. ml O_2/hr
Cu								
Cont.	0	0.3792 ± 0.0174	0	0.3942 ± 0.0203	0	0.4119 ± 0.0168	0	0.3799 ± 0.0136
LC_0	9.17	0.3598 ± 0.0129 5.12	9.60	0.3465 ± 0.0078 12.10	1.50	0.3765 ± 0.0156 8.60	3.41	0.2964 ± 0.0069* 21.98
LC_{25}	12.61	0.3232 ± 0.0135 14.77	14.39	0.3147 ± 0.0168* 20.17	4.67	0.3246 ± 0.0126* 21.19	8.42	0.2192 ± 0.0163* 42.30
LC_{50}	14.81	0.2376 ± 0.0187* 37.34	17.64	0.2331 ± 0.0209* 40.87	8.28	0.2329 ± 0.0173* 43.46	13.27	0.1865 ± 0.0148* 50.91
LC_{75}	17.39	0.2020 ± 0.0206* 46.73	21.63	0.1498 ± 0.0172* 62.00	14.68	0.2026 ± 0.0213* 50.81	20.92	0.1285 ± 0.0186* 66.18
Hg								
Cont.	0	0.3578 ± 0.0126	0	0.3884 ± 0.0111	0	0.3646 ± 0.0137	0	0.3575 ± 0.0152
LC_0	0.59	0.3141 ± 0.0208 16.42	0.88	0.2856 ± 0.0101* 26.47	0.025	0.2675 ± 0.0201* 26.63	0.032	0.2283 ± 0.0146* 35.86
LC_{25}	1.48	0.2785 ± 0.0172* 25.89	3.13	0.2110 ± 0.0074* 45.67	0.050	0.1999 ± 0.0118* 45.17	0.088	0.1852 ± 0.0133* 48.20
LC_{50}	2.33	0.2218 ± 0.0099* 40.98	5.91	0.1502 ± 0.0138* 61.33	0.072	0.1613 ± 0.0121* 55.76	0.147	0.1283 ± 0.0088* 64.11
LC_{75}	3.69	0.1744 ± 0.0116* 53.59	11.14	0.0932 ± 0.0157* 76.00	0.103	0.1113 ± 0.0072* 69.47	0.243	0.0404 ± 0.0071* 88.70

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Table 2 contd.....

Heavy metal	Visakhapatnam Harbor				Bhimilipatnam			
	Test Conc. 96 hr(ppm)	Oxy.Cons. ml O_2/hr	Test Conc. 48 hr(ppm)	Oxy.Cons. ml O_2/hr	Test Conc. 96 hr(ppm)	Oxy.Cons. ml O_2/hr	Test Conc. 48 hr(ppm)	Oxy.Cons. ml O_2/hr
Cd								
Cont.	0	0.3632 ± 0.0168	0	0.3755 ± 0.0141	0	0.4042 ± 0.0132	0	0.3864 ± 0.0181
LC_0	31.12	0.3323 ± 0.0174 8.51	26.38	0.3115 ± 0.0102* 17.04	0.03	0.3545 ± 0.0110* 12.30	1.46	0.2964 ± 0.0207* 23.29
LC_{25}	38.73	0.3225 ± 0.0146 11.21	43.39	0.2026 ± 0.0111* 46.05	2.54	0.2886 ± 0.0166* 28.60	7.15	0.1806 ± 0.0123* 53.26
LC_{50}	43.23	0.2218 ± 0.0101* 38.93	55.75	0.1502 ± 0.0127* 60.00	7.66	0.1865 ± 0.0121 53.86	15.91	0.1116 ± 0.0102* 71.12
LC_{75}	48.26	0.1865 ± 0.0172* 48.65	71.60	0.1324 ± 0.0104* 64.74	23.13	0.1749 ± 0.0133 56.73	35.44	0.0604 ± 0.0061* 84.37
Zn								
Cont.	0	0.3894 ± 0.0173	0	0.3686 ± 0.0151	0	0.3646 ± 0.0144	0	0.4083 ± 0.0123
LC_0	24.42	0.3755 ± 0.0201 3.57	55.76	0.3353 ± 0.0148 9.03	19.40	0.3108 ± 0.0109* 14.76	20.65	0.3584 ± 0.0104* 12.22
LC_{25}	47.50	0.3545 ± 0.0145 8.96	74.35	0.3017 ± 0.0124* 18.15	30.89	0.3017 ± 0.0201* 17.25	35.25	0.3115 ± 0.0172* 23.71
LC_{50}	66.42	0.3246 ± 0.0173* 16.64	85.96	0.2675 ± 0.0208* 27.43	39.05	0.2886 ± 0.0126* 20.85	46.13	0.2192 ± 0.0102* 46.31
LC_{75}	92.87	0.2507 ± 0.0161* 35.62	99.38	0.2102 ± 0.0098* 42.97	49.38	0.1502 ± 0.0101* 58.80	60.39	0.1324 ± 0.0111* 67.57

exposure to concentrations of mercury. The adult fiddler crab, *Uca pugilator* was also found to show suppressed rates of respiration when exposed to mercury and cadmium (Vernberg *et al* 1974). Decreased rates of respiration were noticed in *Caridina rajadhari* on exposure to the metals lead, zinc, copper and mercury and the toxicity was in the order of Hg>Cu>Zn>Pb (Chinnayya 1971). Prabhakara Rao *et al* (1986) observed a decrease in the oxygen consumption of barnacles *Balanus amphitrite amphitrite* and *Balanus tintinnabulum tintinnabulum* subjected to copper. According to Deplege, (1984) the differences in the overall responses to heavy metals reflect the different degree of lipid solubility and toxicity of organo-metal complexes formed by them. In contrast to the present observation, Thurberg *et al* (1977) reported an increase in the respiratory rates in *Homarus americanus* exposed to cadmium. They also observed no difference between the respiration of the controls and mercury -exposed *Homarus americans*. They were of the opinion that these differences were either due to species specificity or the concentration of the metals used.

The relatively lower rates of percentage decrease in the respiration and the insignificant decrease in the respiratory rates observed at LC_0 and LC_{25} of 96 hr in both species of crabs of Visakhapatnam Harbor indicate their resistance to these heavy metals when compared to those of Bhimilipatnam. This can be explained by the fact that both species of crabs of Visakhapatnam Harbor experience an exposure to heavy metals in the environment. Therefore, they might have developed resistance to these heavy metals. An earlier investigation on accumulation studies (Uma Devi and Prabhakara Rao 1988) also revealed the same fact that *U. annulipes* and *U. triangularis* of Visakhapatnam Harbor are resistant whereas those of Bhimilipatnam are sensitive. *U. triangularis* of Bhimilipatnam (Uma Devi and Prabhakara Rao 1988) was reported to be sensitive to heavy metals in particular, and in the present investigation this can be further confirmed by the highest decrease in the rates of its respiration (Table 2) following exposure to heavy metals. However, the decreased respiratory rates on exposure to heavy metals result in anaerobic metabolism leading to mortality of the animals.

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