

Cadmium (Cd) High Levels of Concentration Deplete Biochemical Metabolism in (*Portunus sanguinolentus* (L)) and it Contaminate Human Food Chain at Visakhapatnam Coast, India

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DOI: <https://doi.org/10.52403/ijhsr.20240319>

ABSTRACT

Marine aquatic life is one of the primary food sources for human beings and a drinking water source for the Gulf, Mediterranean, and other world countries. In this view, the author has undertaken studies on the distribution of seasonal dissolved heavy metal (Cd) in the coastal waters of Visakhapatnam over one year (February 2003 to January 2004) and to assess the hydrological and biological factors of carbohydrates and lipids in marine Crab (*Portunus sanguinolentus* (L)) affecting their concentration levels. The result has been followed respectively in this research study. The Cadmium (Cd) and lipid levels were noticed in the 11 selected experimental sites (PLW5 to PLW1 – MPS – PRW1 to PRW5) in the present study for all the samples collection. The author has noticed carbohydrates and lipid levels in the same survey.

On the other hand, high depletion was found in postmonsoon at MPS at 5.208 mg/g and 30.128 mg/g at MPS over the all-experimental sites. The present study evaluated Cadmium (Cd), carbohydrates, and lipids in marine water and The Visakhapatnam coast in different seasons like pre-monsoon, monsoon, and postmonsoon. The Cadmium founded to be high at 0.0460 µg/ml and 0.1383 µg/g, respectively, in water and crab tissue at MPS in all three seasons among all sites, and another hand, the author was noticed carbohydrates and found lipids' high reduction percentage (RPC) in postmonsoon at MPS 50.21% and 16.42% at PRW3 in monsoon compare with overall sites in three seasons, due to toxic Cd could be accumulated in marine Crab through the food chain as well osmosis process of water and body flues of Crab. This bioaccumulation process could be a path for a human being through the food chain. Cadmium accumulates its lead toxic effect with systemic effect on human body organs especially cause kidney damage (renal tubular damage). Human and animal studies indicate skeletal damage (osteoporosis).

Keywords: Cadmium, Crab, Carbohydrates, Lipids, Water

1. INTRODUCTION

Visakhapatnam coast is a natural beauty, and it has a wide range of marine resources. Since the 1940s, it has been depleting the natural quality of organ profiles as well ecosystem. Because it has been exposed to heavy urbanization and industrial pollution joining into the coast. In this research

article, the author has observed the seasonal effect of cadmium levels in the water, Crab, and other total carbohydrates and lipid levels in *Portunus sanguinolentus* (marine Crab). Because of this, the author has undertaken studies on the distribution of dissolved heavy metal (Cd) in the coastal waters of Visakhapatnam over one year

(February 2003 to January 2004) and assessed the hydrological and biological factors affecting their concentration levels. The concentration of heavy metal Cadmium (Cd) has chemical and physical properties that make it attractive for scientific, agricultural, and industrial uses. In aquatic environments, especially in rivers and oceans, heavy metal pollution occurs due to anthropogenic activities. Heavy metals from agricultural lands, urban activities, and industrial effluents mix rapidly with the river, estuary, or ocean water and hence deteriorate the water quality.

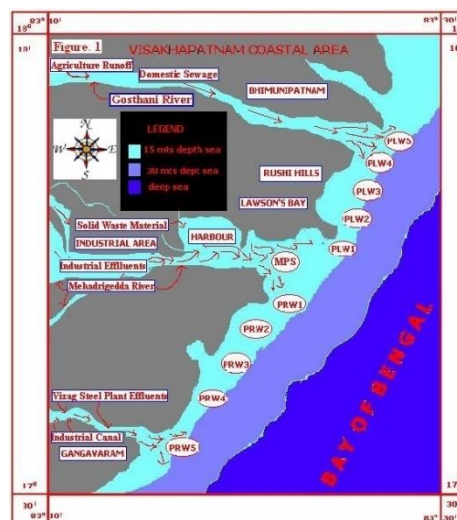
Someswara Rao and Venkata Rao (1989), were reported heavy metals were released either in a combined form with ions like chlorides, sulphates, etc. or in free ionic form (Cd^{+2}). The detailed analysis of the effluents of different industries of the Visakhapatnam coast. Many others also reported the concentration of these heavy metals in harbour water and their accumulation in phytoplankton, zooplankton, etc. (Satyanarayana and Prabhakar Murthy, 1990; Subramanyam and Anantalakshmi Kumari, 1990).

2. MATERIALS AND METHODS

The main objective of the present work is to evaluate the distribution of heavy metal pollution in the Marine Biosphere Reserve, Visakhapatnam coastal area. The collected sample site has located between Bheemunipatnam and Gangavaram (Lat $17^{\circ} 30'$ to $18^{\circ} 10'$ N and Long $83^{\circ} 10'$ to $83^{\circ} 30'$ E) adjoining the industrial area and harbour township of Visakhapatnam on the East coast of India.

In the present study, the author investigated Cadmium (Cd) concentration in Marine crab (*Portunus sanguinolentus*), water, and other biological parameters likely total carbohydrates and lipids followed from February 2003 to January 2004 in selected sites likely 11 experimental sites were selected at Visakhapatnam coast. The present study selected four regions (Gangavaram, Harbour, Rushi Hills, and

Bhimuniptnam) from the Visakhapatnam coast (Fig. 1).



The four selected areas were divided into eleven sample collection sites. The main sample collection points are located in between the industrial and the harbour area of the Visakhapatnam coast, named as Main Pollution Source (MPS). Six sample collection points are located on the left side of the MPS which is named Pollution Left Wing (PLW1 to PLW5), and the remaining five are located on the right side of the MPS, which is named Pollution Right Wing (PRW1 to PRW5). These sites approximate distance between each sample collection site is about 3 km to the argental line followed by the seashore.

2.1 Samples Collection and Preparation for Analysis

The samples were collected from the selected areas seasonally (premonsoon, monsoon, and postmonsoon periods) from February 2003 to January 2004 (one year). There were brought to the laboratory and the heavy metals concentration (Cadmium) in the marine water, and aquatic organisms (Crabs) were estimated biochemical parameters likely carbohydrates and lipids in the dry muscle tissues of marine crabs (*Portunus sanguinolentus*) were also estimated.

For statistical assessment, six replications of each sample (living and nonliving) were

collected from the selected sites in different seasons for the study.

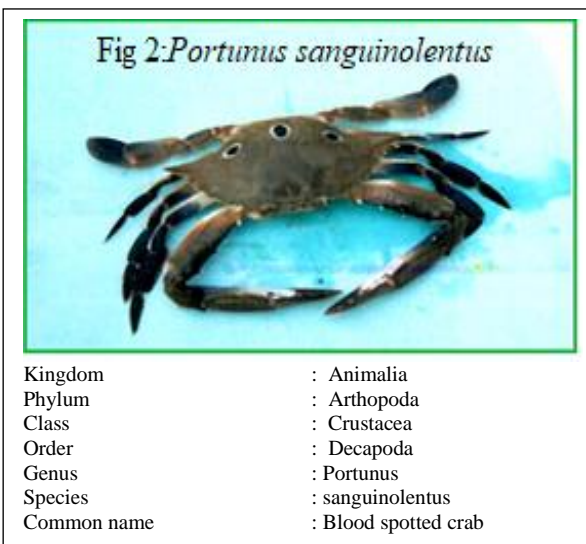
2.1.1. Collection of Samples

At each sampling site, water and crabs are collected from six different spots. Hence, for each site, there are six replications. An average of six replications have been taken in presenting the data.

2.1.2. Test animal - Crab

The collected crab samples were washed with fresh water to remove adherent sand particles and other unwanted materials. The morphological description, along with the site of collection of each sample recorded individually. The cleaned materials were packed individually in Zip log polythene bags and placed in thick-walled thermocol boxes (1½ x 1¼ x 1¼ feet with three inches of wall thickness containing a 1:1 ratio of ice and salt mixture). The sample boxes were labelled and were brought to the laboratory and stored at -20°C to prevent putrefaction until the completion of work. The mean of six samples was taken for all parameters. That is the concentration of water and tissue of marine Crabs.

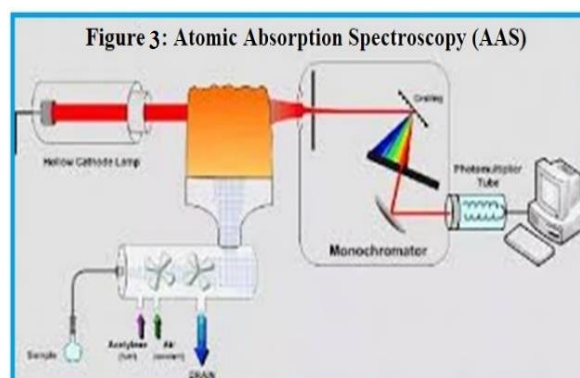
2.1.3. Physical preparation of marine Crab
Crabs, *Portunus sanguinolentus* (Fig 2) were collected at seasonal intervals during the study period from all the selected sites. The muscle samples were collected from the Crab by removing the carapace and cutting the required quantity of muscle mass that only dried at 60°C for 48 hrs then; fibre muscle was taken for quantitative heavy metal estimation and pulverized for biochemical assessment. The muscle and powder are preserved in a desiccator. Individual powder samples were weighed for quantitative assessment of heavy metals, carbohydrates, and lipids.



2.2 Analytical Methods

2.2.1. Heavy metals in marine water

The solvent extraction procedure was adopted for pre-concentration of trace metals from marine waters as a preliminary step prior to their determination by Atomic Absorption Spectroscopy (AAS) (Fig 3). Ammonium pyrrolidine dithiocarbamate (APDC) is used for the chelation of heavy metals in water (pH 2-3) and Methyl Iso Butyl Ketone (MIBK) for extraction of metal chelates from the aqueous medium followed by their determination by AAS (Robert R. Brooks et al., 1967). The Metal chelates are not stable for a long time in MIBK, and hence the metal-APDC complexes were back-extracted from MIBK into an aqueous medium using repeated extractions with 4N HNO₃ (Jan and Young, 1978).



2.2.2. Extraction of metals from marine water

The aliquot (800 ml) of filtered water samples was placed in a Teflon beaker of 1 L capacity, and the pH was adjusted to 2-3 by adding dilute NaOH (6N) solutions drop-wise. Then 10 ml of 2% (w/v) APDC solution was added and thoroughly mixed. The APDC solution was purified by repeated extraction with MIBK prior to use. The contents were transferred to a separating funnel (1 L) treated with 15 ml of fresh distilled MIBK (pre-equilibrated with de-ionized water containing 10 ml. 2% APDC solution) and thoroughly shaken for 5 minutes to attain equilibrium. The two phases were then allowed to separate, and the lower aqueous layer was transferred into another 1-litre-separating funnel it was again treated with 5 ml of 2% APDC and 10 ml of MIBK and thoroughly equilibrated for 5 min. After the second equilibration, the solution containing only major elements that do not form complexes with APDC and thus get extracted into MIBK was used as "TRACE METALS FREE" water for preparation of standard in AAS determination. Organic layers containing metal complexes obtained in the two successive extractions were combined, transferred into a 60 ml separating funnel, and treated with 15 ml of 4N HNO₃ in order to back-extract trace metals into the aqueous medium. The contents were thoroughly shaken for 5 minutes, and the aqueous (4N HNO₃) phase was quantitatively transferred into a 25 ml screw-capped polyethylene bottle. The organic phase left in the first extraction was again transferred with 10 ml of 4N HNO₃, thoroughly shaken for 5 minutes, the two phases were allowed to separate and the aqueous phase was again transferred into the polythene bottle containing the aqueous phase obtained in the first back extraction step. The combined aqueous phase containing the trace metals was stored in a deep freezer until their analysis by AAS (Perkin – Elmer 2380). The samples were analyzed within three hours of extraction.

2.2.3. Extraction of Cd from marine Crab

The researcher extracted metals from the tissue of the Crab following the method of Helen *et al.*, (1991).

5g dry weight of crab tissue (from each individual Crab) was taken in the crucible and dried at 135°C for 2 hrs. The dry weight was noted first, and then they were transferred to a cool muffle furnace, and slowly, the temperature was raised to 450°C – 500°C and kept overnight. Then the samples were removed and cooled to room temperature. Next, cautiously 2 ml of HNO₃ was added and swirled. They were held on a hot plate and allowed to evaporate until dry. The dry sample was then transferred to a coal furnace, and the temperature slowly was raised to 450°C – 500°C. This temperature was maintained for one hour. Crucible was removed from the furnace and cooled. Nitric Acid treatment was repeated three times to obtain clean, carbon-free ash. To this, 10 ml of 1 N HCl was added and heated cautiously on the hot plate to dissolve the ash. These contents were transferred to a 25 ml volumetric flask made up of 1 N HCl. The samples were kept until they cooled and injected into AAS (Perkin – Elmer 2380) for estimation of different heavy metals.

2.3 Bio-Chemical Methods

In the present study, the researcher has estimated the total content of carbohydrates and lipids measured in the muscle tissue of a marine Crab (*Portunus sanguinolentus*), as followed by the below standard procedure.

2.3.1. Total Carbohydrates from marine Crab

The carbohydrate content in the dried tissues was estimated by adopting the technique of Dubois *et al.*, 1956, which involves the hydrolysis of di and oligosaccharides into monosaccharides into furfural or furfural derivatives. These two compounds react with a number of phenolic compounds. One such is phenol, which produces a complex light orange colour product, the intensity of which is

proportional to the total carbohydrates or saccharides present in the sample. Then the optical density of the colour developed was measured at 490 nm, and the carbohydrate content of the sample was calculated with the help of a standard graph prepared by known quantities of glucose λ .

2.3.2. Total Lipids from marine Crab

The lipids were estimated by Bligh and Dyer's 1959 method. The total lipids were extracted from the dry tissues following the gravimetric method. 1 g of fine dry powder was homogenized in 2:1 chloroform and methanol in the ratio of 30 ml of mixture to one gram of the tissue. The sample was centrifuged for 10 minutes at 2,500 pm. And the supernatant was filtered into a pre-

weighed aluminum boat. The sample in the aluminum boat was dried in an oven. The weight difference between the initial weight of the aluminum boat and the dry weight of the lipid-containing aluminum boat gives the total lipid content in the sample. Total lipid content was given as $\mu\text{g/g}$.

2.4. Statistical Calculations:

2.4.1. Cadmium water and Crab:

The samples were analyzed by an Atomic Absorption Spectrophotometer (AAS) (Perkin – Elmer 2380) using an air acetylene flame. All the spectroscopic measurements of the standard metal solution, as well as the sample solutions, were done at their respective wavelength of maximum absorption λ_{max} .

2.4.2. Carbohydrates estimation:

$$\% \text{ Composition of Carbohydrates} = \frac{\text{Standard Value X OD of Sample}}{\text{Weight of Tissue}} \times 100$$

2.4.3. Lipid estimation:

$$\% \text{ Composition of Lipid} = \frac{\text{Weight of Lipid}}{\text{Weight of Tissue}} \times 100$$

2.4.4. ANOVA:

Samples were done anova two pare statistical test between the two seasons at $P < 0.05$.

The Cadmium found a high trend of followed Postmonsoon > Monsoon > Premonsoon seasonal to be noticed. MPS site noticed high concentrations of 0.046 $\mu\text{g/ml}$, 0.032 $\mu\text{g/ml}$, and 0.015 $\mu\text{g/ml}$ respectively in three seasons. Another hand side Postmonsoon, Monsoon, and Premonsoon Cadmium was noticed in low concentrations at PRW2 0.030 $\mu\text{g/ml}$, 0.020 $\mu\text{g/ml}$, and 0.006 $\mu\text{g/ml}$, respectively in three seasons. However, the metal cadmium was noticed by Bellow Detection Limits (BDL) at PRW3 in premonsoon.

3. RESULTS

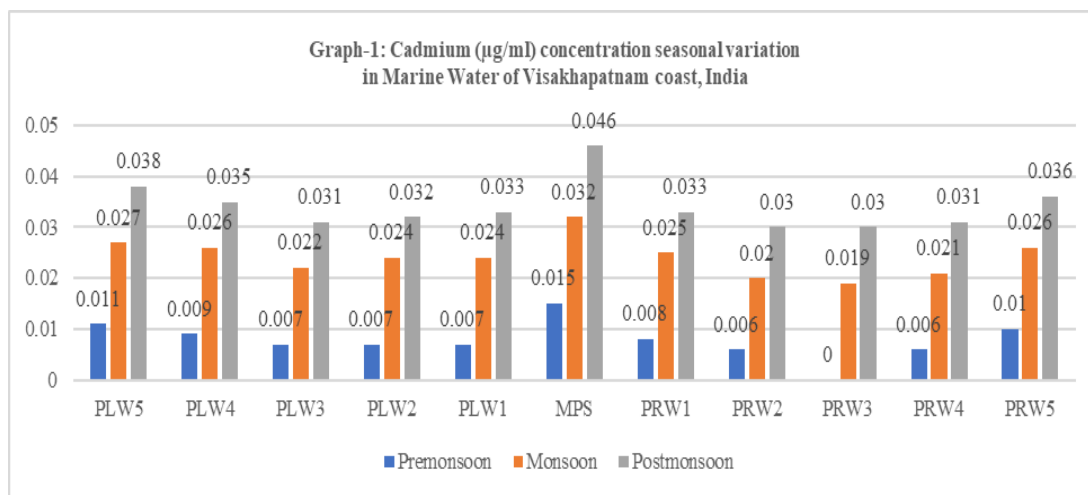
3.1 Cadmium (Cd) Concentration in Marine Water

Table-1 and Graph-1 were shown the data on the concentrations of cadmium metal presented in the 11 selected experimental sites in the marine water of Visakhapatnam coast in different seasons like the premonsoon, monsoon, and postmonsoon.

Sites	Seasons					
	Premonsoon		Monsoon		Postmonsoon	
	Mean	SD (\pm)	Mean	SD (\pm)	Mean	SD (\pm)
PLW5	0.011	0.0059	0.027	0.0041	0.038	0.0042
PLW4	0.009	0.0021	0.026	0.0043	0.035	0.0039
PLW3	0.007	0.0023	0.022	0.0044	0.031	0.0041

PLW2	0.007	0.0025	0.024	0.0046	0.032	0.0035
PLW1	0.007	0.0021	0.024	0.0041	0.033	0.0035
MPS	0.015	0.0035	0.032	0.0045	0.046	0.0045
PRW1	0.008	0.0021	0.025	0.0045	0.033	0.0036
PRW2	0.006	0.0019	0.020	0.0036	0.030	0.0032
PRW3	PDF	-	0.019	0.0036	0.030	0.0036
PRW4	0.006	0.0016	0.021	0.0045	0.031	0.0041
PRW5	0.0100	0.0032	0.026	0.0044	0.036	0.0043

Values are mean (n = 6) ± SD; BDL: Below detection limit; Anova test significant each individual season at P< 0.05



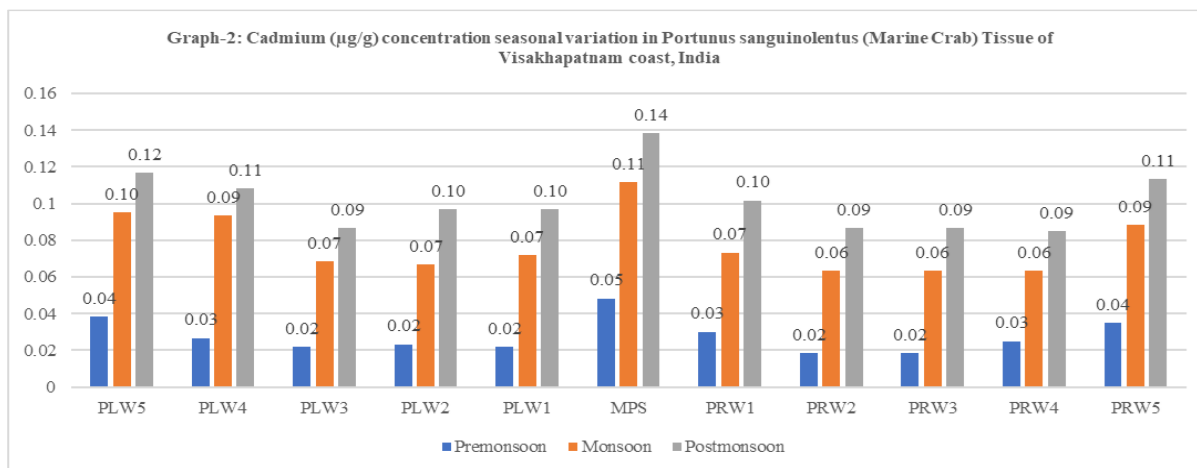
3.2 Cadmium (Cd) Concentration in Dry tissue of Marine Crab

Table-2 and Graph-2 were shown the data on the concentration of cadmium metal presented in the 11 selected experimental sites in the marine water of Visakhapatnam coast in different seasons like the premonsoon, monsoon, and postmonsoon. The Cadmium found a high trend of

following Postmonsoon > Monsoon > Premonsoon seasonal to be noticed. MPS site saw a high concentration of 0.141 µg/g, 0.107 µg/g, and 0.046 µg/g respectively in three seasons, and a low concentration of Cadmium was noticed at PRW3 0.086 µg/g, 0.062 µg/g, and 0.008 µg/g respectively in three seasons.

Sites	Seasons					
	Premonsoon		Monsoon		Postmonsoon	
	Mean	SD (±)	Mean	SD (±)	Mean	SD (±)
PLW5	0.035	0.0158	0.092	0.0251	0.118	0.0261
PLW4	0.029	0.0145	0.088	0.0245	0.107	0.0254
PLW3	0.022	0.0151	0.065	0.0162	0.088	0.0162
PLW2	0.023	0.0132	0.067	0.0143	0.096	0.0153
PLW1	0.023	0.0141	0.071	0.0143	0.096	0.0151
MPS	0.046	0.0152	0.107	0.0215	0.141	0.0252
PRW1	0.025	0.0132	0.074	0.0153	0.101	0.0212
PRW2	0.019	0.0138	0.062	0.0138	0.086	0.0138
PRW3	0.008	0.0038	0.062	0.0141	0.086	0.0153
PRW4	0.021	0.0143	0.063	0.0152	0.087	0.0146
PRW5	0.032	0.0142	0.090	0.0152	0.112	0.0154

Values are mean (n = 6) ± SD; Anova test significant each individual season at P< 0.05



3.3 Total Carbohydrate content in Dry tissues of Marine Crab

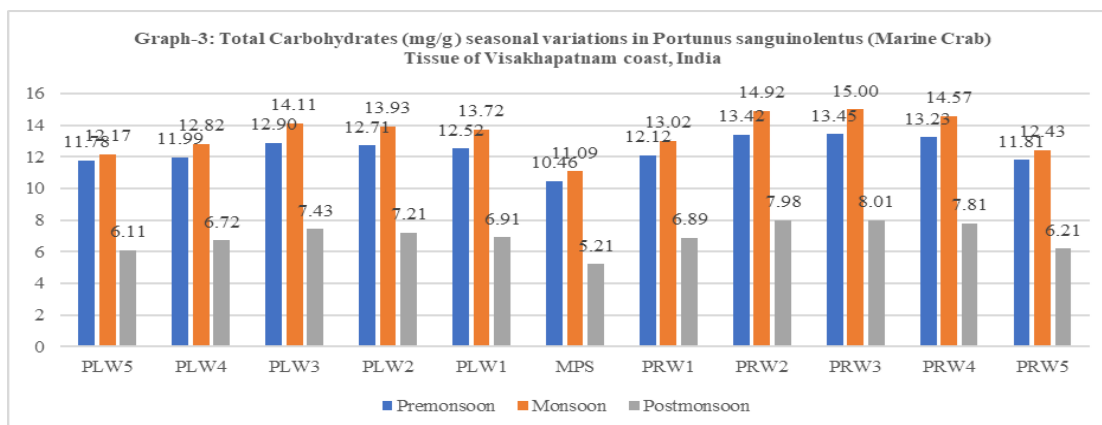
Table-3 & 3.1 and Graph-3 & 3.1 show the data on the amounts of total carbohydrates presented in the dry tissues of marine Crab of 11 selected experimental sites of the Visakhapatnam coast in different seasons like premonsoon, monsoon and postmonsoon. The total carbohydrates deduction in the dry tissues of marine crab trend was followed by Postmonsoon

>Monsoon >Premonsoon and MPS was found more deduction of carbohydrates 5.21 mg/g. The author observed MPS postmonsoon more percentage deduction of 10.46 mg/g (50.21%) and 11.09 mg/g (53.02%) when compared respectively premonsoon and monsoon, and PRW3 high content of carbohydrates was noticed at 3.45 mg/g, 15 mg/g and 8 mg/g respectively in three seasons.

Table – 3: Total Carbohydrates (mg/g) seasonal variations in *Portunus sanguinolentus* (Marine Crab) Tissue of Visakhapatnam coast, India

Sites	Seasons					
	Premonsoon		Monsoon		Postmonsoon	
	Mean	SD (±)	Mean	SD (±)	Mean	SD (±)
PLW5	11.78	3.56	12.17	3.33	6.11	1.33
PLW4	11.99	4.33	12.82	3.45	6.72	1.03
PLW3	12.90	3.13	14.11	3.59	7.43	1.15
PLW2	12.71	3.54	13.93	4.21	7.21	1.02
PLW1	12.52	4.02	13.72	3.25	6.91	1.04
MPS	10.46	3.25	11.09	2.99	5.21	1.90
PRW1	12.12	2.99	13.02	3.25	6.89	1.55
PRW2	13.42	3.22	14.92	4.02	7.98	1.25
PRW3	13.45	3.42	15.00	4.13	8.01	1.55
PRW4	13.23	2.90	14.57	3.88	7.81	1.33
PRW5	11.81	2.59	12.43	3.03	6.21	1.55

Values are mean (n = 6) ± SD; Anova test significant each individual season at P< 0.05



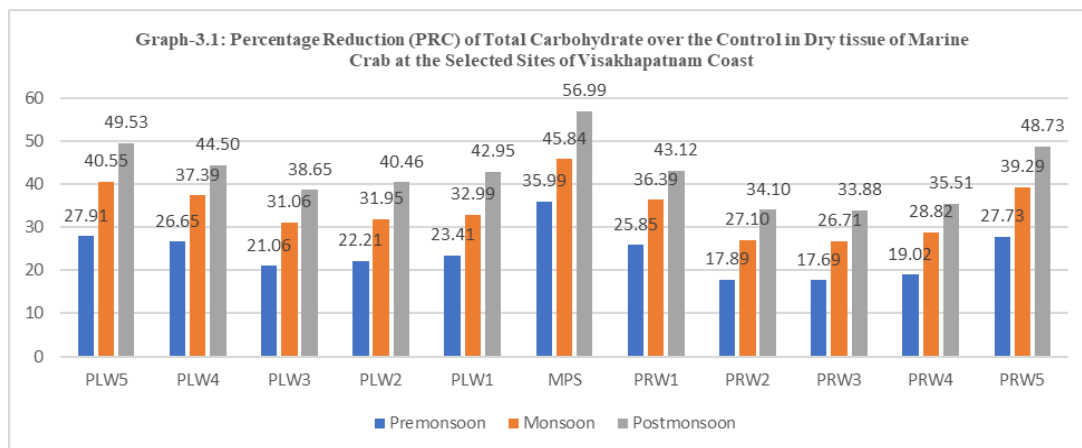


Table – 3.1: Total Carbohydrates Reduction Seasonal Variations in *Portunus sanguinolentus* (Marine Crab) Tissue of Visakhapatnam Coast, India

Sites	Carbohydrates	
	Postmonsoon RCP with	Premonsoon
PLW5	48.13	49.79
PLW4	43.93	47.56
PLW3	42.42	47.35
PLW2	43.28	48.23
PLW1	44.80	49.63
MPS	50.21	53.02
PRW1	43.15	47.09
PRW2	40.52	46.51
PRW3	40.47	46.62
PRW4	40.99	46.39
PRW5	47.43	50.03

3.4 Total Lipids content in Dry tissues of Marine Crab

Table-4 & 4.1 and Graph-4 & 4.1 show the data on the amounts of total Lipids presented in the dry tissues of marine Crab of 11 selected experimental sites of the Visakhapatnam coast in different seasons like pre-monsoon, monsoon and post-monsoon. The total lipids deduction in the dry tissues of marine crab trend was followed by Post monsoon > Monsoon > Pre

monsoon. MPS was found more deduction of lipids 30.13 mg/g in monsoon. The author observed MPS monsoon more percentage deduction of 35.13 mg/g (14.23%) and 33 mg/g (8.71%) when compared with respectively premonsoon and postmonsoon, and high content lipids PRW3 were noticed 42.98 mg/g, 35.92 mg/g and 44 mg/g respectively in three seasons.

Table – 4: Total Lipids (mg/g) seasonal variations in *Portunus sanguinolentus* (Marine Crab) Tissue of Visakhapatnam coast, India

Sites	Seasons					
	Premonsoon		Monsoon		Postmonsoon	
	Mean	SD (±)	Mean	SD (±)	Mean	SD (±)
PLW5	37.68	5.24	32.06	5.25	35.72	5.25
PLW4	38.30	4.24	33.28	4.24	36.93	4.57
PLW3	40.92	4.86	34.88	4.52	42.42	4.26
PLW2	40.00	4.26	34.41	5.25	41.98	5.24
PLW1	39.67	4.13	34.11	4.13	41.01	4.24
MPS	35.13	3.88	30.13	3.90	33.00	3.66
PRW1	38.92	3.76	33.92	4.13	40.67	4.01
PRW2	42.11	4.24	35.67	5.12	43.76	4.24
PRW3	42.98	4.13	35.92	4.59	44.00	3.88
PRW4	41.62	4.37	35.09	4.13	43.00	4.02
PRW5	38.01	3.88	32.93	4.37	36.12	3.55

Values are mean (n = 6) ± SD; Anova test significant each individual season at P< 0.05

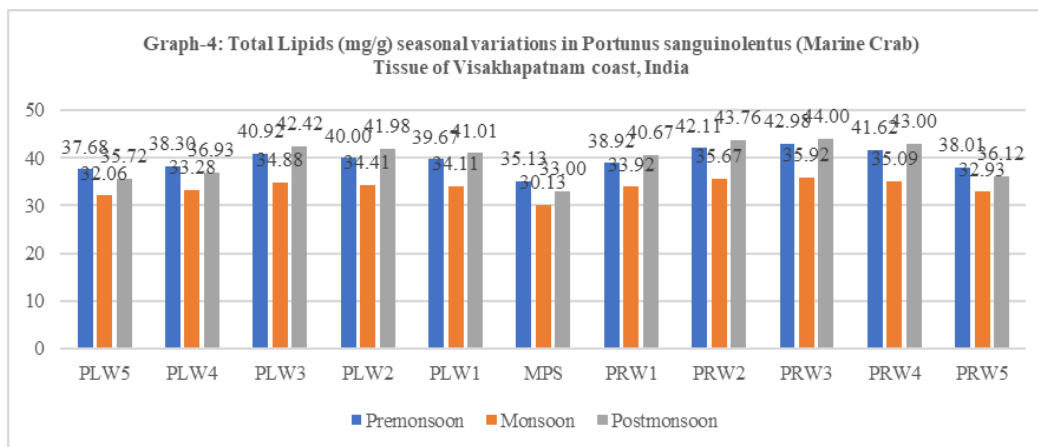
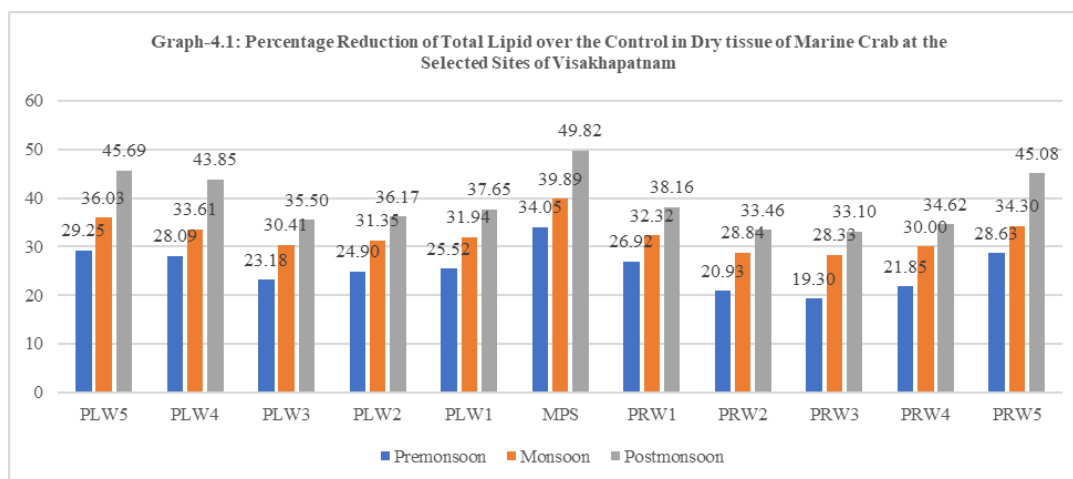


Table – 4.1: Total Carbohydrates and Lipids Percentage Reduction Seasonal Variations in *Portunus sanguinolentus* (Marine Crab) Tissue of Visakhapatnam Coast, India

Sites	Lipids	
	Monsoon RCP with	
	Premonsoon	Postmonsoon
PLW5	14.91	10.25
PLW4	13.12	9.89
PLW3	14.75	17.78
PLW2	13.98	18.03
PLW1	14.01	16.81
MPS	14.23	8.71
PRW1	12.85	16.59
PRW2	15.30	18.49
PRW3	16.42	18.36
PRW4	15.70	18.40
PRW5	13.37	8.83



4. DISCUSSION

4.1 Cadmium (Cd) in Marine Water and Crab

Cadmium, a non-essential element with no biological function (Viarengo, 1985) is highly toxic to man (Dianne *et al.*, 1999; Monisha Jaishankar *et al.*, 2014) and other living organisms. The primary route of Cd exposure is contaminated water or food supplies (Leffel *et al.*, 2003; Satarug and

Moore 2004), smoking (Satarug and Moore 2004), mother's milk (Pillet *et al.*, 2005), work in battery factories (Sahmoun *et al.*, 2005), or through fertilizers or indiscriminate use of pesticide (Weggler *et al.*, 2004). Cadmium is toxic to humans and animals, and excessive exposure to it results in diseases and occasionally death (Othumpangat *et al.*, 2005). The critical effect is the first physiological or

biochemical change that can be detected (Nordberg, 1992; Christopher J. Portier, 2012). According to the Department of Water Affairs and Forestry (DWAFF 1996c), Cadmium is a metal element that is highly toxic to marine and freshwater aquatic life. Paul B Tchounwou *et al.*, 2012 reported the routes of uptake of Cadmium by fish are mainly through ingestion of food, water, and both. Many people suffer due to cadmium toxicity causes a disease in which their bones become fragile and muscles contracted. It has led to osteoporosis including pain, physical impairment, and decreased quality of life (Mehrdad Rafati Rahimzadeh 2017).

The data on the concentration of Cd in all selected sites of marine water and Crab was presented in Tables 1 & 2 and Graph-1 & 2, respectively. The MPS was Cd concentration incremental significant ($P < 0.05$) levels found in postmonsoon with comparing premonsoon and monsoon at all sites in marine water and marine Crab. The MPS site was in premonsoon and noticed significant ($P < 0.05$) to PLW2, PLW3, PLW4, PRW1, PRW2, PRW3, PRW4, and PRW5. The cadmium bio-accumulate found incremental concentration seasonal significance ($P < 0.05$) in marine Crabs at all sites followed by premonsoon, monsoon, and postmonsoon. There were all sites showed similar results in all seasons. However, in premonsoon, Cadmium was not found (BDL) at PRW3, and at the same time, equal amounts of Cd such as 0.007 mg/l and 0.006 mg/l were detected at PLW1, PLW2, PLW3 and PRW2, PRW4 respectively in marine water.

The Cadmium bioaccumulated concentration in marine Crabs has not exceeded the Indian standard limits (Annexure – I) in all sites in three seasons. Cd concentrations in the marine Crab and water were found to be very high in two seasons (monsoon and postmonsoon) at MPS, PLW4, PLW5, and PRW5. Similarly, Asha Raj and Mundanchery 2017 found results in postmonsoon have shown high levels of cadmium concentration at Periyar

River in Ernakulam district. These variations were due to the location of MPS at an area of the Visakhapatnam harbour where the Mehadrigeedda river carrying industrial effluents and urban domestic sewage joined. Gosthani river flow and agriculture runoff entering into the marine environment in between PLW4 and PLW5 and an industrial water storm into PRW5 also could be influencing the level of metal concentration (Ganapathi and Subba Rao, 1958).

Moreover, the concentration of Cd was found more amount water and Crab in postmonsoon 0.046 $\mu\text{g/g}$ and 0.141 $\mu\text{g/g}$ at MPS, respectively, which due to Monsoonal rainfalls loaded various pollutants through urban and industrial runoff and other pollutant washout from agricultural lands. Similarly reported by Sujan Dey *et al.*, 2015 most of the dissolved heavy metals were in slightly higher concentrations during winter than that during the rainy season. This trend indicates that during low flow conditions of the river, the accumulation of the metal concentration increases. Furthermore, untreated industrial wastes contribute to the coastal dead zone (i.e., insufficient oxygen level to support marine life) extension and heavy metal contamination of seafood constantly (Dell Anno *et al.*, 2002; Xiao-e Yang *et al.*, 2008). The takeover led to bio-magnification may ultimately pose a threat to human health, which is eating contaminated fish and other aquatic species (Li Y 2013). The author noticed a similar report of increased levels of cadmium concentration from premonsoon to postmonsoon rest of the all-experimental sites (Asha Raj and Mundanchery, 2017). Daniel P. Loucks and Eelco van Beek (2017) reported these are increments of the amount of metal concentration that has been attributed to an imbalance of the ecosystem, in the form of eutrophication and deficiency of oxygen levels in the water.

Cadmium causes toxic effects chronic and acute exposure duration of the benthic organ of marine Crab (*Portunus sanguinolentus*) exposure to sublethal concentrations of

Cadmium brought about degenerative changes in the hepatopancreas and gill of exposed animals (Bharat Bhusan Patnaik *et al.*, 2011). Extensive cellular proliferation (hyperplasia) of secondary lamellae accompanied by cyst formation and necrotic regions is often seen in the gills of Cadmium-treated crabs (Krishnaja, A.P *et al.*, 1987). However, the Cd concentration in marine waters was found to exceed limits according to Indian standards annexure-1 (Awashthi 2000) in monsoon and postmonsoon at all sites but in premonsoon at MPS and PLW5 in marine water.

4.2 Total Carbohydrates and Lipids in Marine Crab:

The study of the biochemical changes in the marine organisms in polluted areas is necessary to determine the nature of pollutants like those heavy metals and whether they affect the organisms and aquatic system or not. Crabs are a rich source of healthy micro and macronutrients and minerals, in addition owing to their protein content, unsaturated fatty acids, and carbohydrates (Adeyeye EI, 2002). Biochemical studies like estimating carbohydrates and lipids' total contents in marine organisms like fishes, crabs, etc., are treated as bio-indicators. There is a need to assess them to find out the fluctuations in the biological functions of the animals. Szefer *et al.*, 1997 used pacific oysters and crabs as trace metal bio-indicators of the East coast water of Kyushu Island, Japan. In addition, many investigators used marine organisms like fish (Mohamed *et al.*, 1990; Khallaf *et al.*, 1994; Adham *et al.*, 1999). The use of some crustaceans as bio-indicators in the Tamar, Avon, and other estuaries in Southwest England was described by Bryan *et al.*, 1980.

The distribution of heavy metals, particularly within the tissues of marine animals, is of interest in understanding the role they play in the biochemical and physiological mechanisms of the organism (Emara *et al.*, 1993; UNEP 1996 and UNEP, 2001). Mostly Carbohydrates and lipids, and

sometimes proteins serve as energy sources for animals. The TCA cycle is the main pathway for the oxidation of carbohydrates, lipids, and proteins (Hamsford, 1990). The major function of carbohydrates in metabolism is to provide energy for vital activities. The metabolism of carbohydrates has subdivided into glycolysis, glycogenesis, glycogenolysis, oxidation of pyruvate, hexose monophosphate shunt, gluconeogenesis, etc. (Sheline *et al.*, 2000 and Khadija Mounaji *et al.*, 2003).

Carbohydrates act as the chief source of energy in organisms (Weeda and De Kort, 1979). They also reported the utility of carbohydrates in the flight muscle of the *Colorado* potato beetle. Hamsford (1990) may occur through glycogenesis, the storage product of glycogen, which utilizes through gluconeogenesis. Lipids are of great biochemical importance in organisms. These are the energy sources available in food and thus serve as the prime fuel reserve for metabolism (Lehninger, 1982). Besides this, they provide two times more metabolic water on complete oxidation in the body than carbohydrates or proteins.

The present study's aim and objective of this research is to study heavy metal stress on carbohydrates and lipids fluctuations in marine Crabs. The biochemical parameters were observed in three seasons, namely premonsoon, monsoon, and postmonsoon. A gradual decrease in the carbohydrate and lipid contents in the muscle tissue of marine Crabs was observed in seasonal variations (tables-3 & 4 and Graph-3 & 4). The low amounts of carbohydrates were observed compared to lipids in the muscle tissue of marine Crabs in the present work.

Carbohydrate and lipid content were reductions observed in the muscle tissue of the marine Crab highly in postmonsoon followed by monsoon and premonsoon. The carbohydrates MPS was more depletion 5.21mg/g content observed in postmonsoon (tables-3 and Graph-3). The MPS percentage of reduction was noticed at 50.21% and 53.02% when compared respectively with premonsoon and monsoon

(tables 3.1 and Graph-3.1). The author has observed carbohydrate total content significant ($P < 0.05$) between three seasons. In addition, the author noticed incremental levels of Cd concentration at MPS respectively in three seasons. MPS site showed more reduction of carbohydrates and lipids when compared to other sites in the present study, which is due to crab tissue's high Cd levels accumulating. Therefore, when has Cd level has reached an absorbed dose, it starts to damage the cell life functions of an organism (Albergoni and Piccinni, 1983) and leads to decrease levels of carbohydrates and lipids. Even if contaminants are not expected to be bioavailable (such as refractory metal sulfide solids), processes in the environment or the body of those exposed to cadmium compounds may alter or render the metals bioavailable (Laurel A *et al.*, 2007).

The decrease of carbohydrates and lipids observed in the present study is similar to the findings made earlier in various species of Crab and prawns after exposure to steviol, endocal, diesel, naphthalene and pesticides (Khan *et al.*, 1988; Sarojini *et al.*, 1989; 1990). The general decrease in the carbohydrate content in muscle tissue may be due to high energy demand during industrial exposure (Bhagyalakshmi, 1981; Nagabhusanam and Sambasivarao, 1986).

In the present study, the author observed more depletion of lipid content 30.13 mg/g at MPS sites in monsoon (tables 4 & 4.1 and Graph- 4 & 4.1) and the MPS site when compare percentage reduction of 14.23% and 8.71%, respectively compared with premonsoon and postmonsoon (tables-4.1 and Graph-4.1). The author observed lipids' total content significant ($P < 0.05$) levels between three seasons. Long-term Cd exposure indicates induced gluconeogenesis and liver dysfunction. The decreased total lipid content was observed in muscle tissue, maybe due to the enzyme responsible for the breakdown of lipids into free fatty acids and glycerol (West, E.S *et al.*, 1967). As a related matter, it has been shown that while some fish, such as Arctic char, may

eliminate Cadmium (Reinhard *et al.*, 1997), long-term net accumulation still occurs in Arctic char that are exposed to a continuous supply of Cadmium in water (Reinhard *et al.*, 1997). The distinctions between and impacts of water versus food/ingestion cadmium sources have not been clearly identified (Saiki, *et al.*, 2000). Recently The Hindu, (2020) and NCBI 2020 reported one of the industrial tragedies was caused by styrene gas leaked on 7, May 2020 styrene gas leaked from a storage tank on the premises of an LG Polymers unit in Gopalapatnam, Visakhapatnam. A.P, India. This styrene tragedy happened due to improper industrial guidelines for reopening or reoperation industry after the COVID-19 lockdown opened at Visakhapatnam, A.P, India. It is very close to Harbour, as well as the Mehadrigedda River. This tragedy was within a few hours; the gas had killed 11 people and injured hundreds of others. Styrene (C_8H_8) is a chemical compound used to manufacture plastic and rubber. It is toxic acute to the brain and lungs, as well as chronic health effects causes as cancer (CDC, 2020). The author has observed the mystery of water recently and food nickel, lead. Mercury contaminations caused disease outbreaks, which finds clinical symptoms of convulsions, vomiting, headache, dizziness, seizures, nausea, anxiety, loss of consciousness, and other symptoms (Wikipedia 2020; Indian express 2020; BBC, 2020). According to these workers, tissue hypoxia might have played a significant role in the synthesis of lipids for carbohydrate precursors in fish exposed to various toxicants.

CONCLUSION

The results in the present study have demonstrated that water and crab tissue inhabit more cadmium (Cd) concentration in the postmonsoon season. This Cd toxicity post-affected Carbohydrates and Lipids in respective seasons Postmonsoon and Monsoon. The marine crab tissue showed the highest depletion of total carbohydrates and Lipids in postmonsoon and monsoon,

respectively. Marine aquatic Crab showed various levels of biochemical properties based on the inhabiting of cadmium levels in marine water and crab tissue. The author has findings of the present study, which indicates the joining of untreated various industrial, urban, and agricultural waste solid and effluents of various commercial activities discharged into the marine ecosystem with abnormal standards (untreated). The author that industrial accidental and hazardous situations must prevent and control. Those polluters need to follow the environmental, water, and aquatic safety acts and standards before discharging waste from the source into the marine ecosystem. If the polluter were not followed environmental and health actions and standards, these activities would attribute chronic and acute public health issues through bio-magnification. Therefore, the author has requested recommendations to implement and update standards of EPA and WHO rules for all surrounding industries and other activities to state and central pollution control boards.

Declaration by Authors

Acknowledgement: This current research work was entirely carried out at the Dept. of Environmental Science, S.V. University, Tirupati – 517502, A. P., India. I want to say my great thanks to all teaching and non-teaching staff; they have supported all scientific and moral support to complete my current research.

Source of Funding: None

Conflict of Interest: The authors declare no conflict of interest.

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How to cite this article: Dasari Sreeramulu, Perumalla Ratnakumari. Cadmium (Cd) high levels of concentration deplete biochemical metabolism in (*Portunus sanguinolentus* (L)) and it contaminate human food chain at Visakhapatnam Coast, India. *Int J Health Sci Res.* 2024; 14(3):117-132. DOI: <https://doi.org/10.52403/ijhsr.20240319>
