# Heavy metals accumulation in cultured Shrimp in South-West farming regions of Bangladesh and human health risk assessment

A thesis submitted to the Department of Fisheries, University of Dhaka

in partial fulfillment of the requirements for the degree of

Masters of Science (MS) in Fisheries

**Submitted By**


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**Dhaka-1000, Bangladesh**


**12 February 2017**

# Declaration

I hereby declare that the dissertation entitled **"Heavy metals accumulation in culturedShrimp in South-West farming regions of Bangladesh and human health assessment"** submitted to the Department of Fisheries, University of Dhaka for the degree of Master of Science (MS) is based on self-investigation, carried out under the supervision of**Dr Mohammad ShamsurRahman**, Department of Fisheries and **BadhanSaha**, Scientific Officer, Soil and Environmental section, BCSIR Laboratories, Dhaka-1000, Bangladesh.

I also declare that this or any part of this work has not been submitted for any other degree anywhere. All sources of knowledge used have been duly acknowledged.

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# Certificate

We certify that the research work embodied in this thesis entitled **"Heavy metals accumulation in cultured Shrimp in South-West farming regions of Bangladesh and human health risk assessment"** submitted byMd. Monwarul Islam,Exam Roll No: Curzon 810, Session: 2015-16, Registration number: 2011-212-768, has been carried out under our supervision

This is further to certify that it is an original work and suitable for the partial fulfillment of the Degree of Master of Science (MS) in Fisheries from the Department of Fisheries, University of Dhaka.

We wish every success in his life.

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**Author**

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# Abstract

Heavy metals contamination of aquatic environment has attached global attention owing to its abundance, persistence and environment toxicity, especially in developing countries like Bangladesh. They are not only the threat to aquatic environment but also can pose risk to human health through the consumption of aquatic products especially fish and Shrimp.

This study was focused on heavy metals accumulation and estimation of human health risk ofthree Upazilas (Satkhira Sadar, Morrelganj and Mongla) of two districts (Satkhira and Bagerhat) in South-West region of Bangladesh. For the determination of heavy metal concentration using Atomic Absorption Spectrometer (Model No: AA-7000, Shimadzu) a total of 20 freshly caught cultured Shrimp (*Penaeusmonodon*)were randomly sampled from Satkhira Sadar Upazila of Satkhira district, Morrelganj and Mongla Upazila of Bagerhat district during pre-monsoon (May-June) and post monsoon (November-December) of 2016. During pre-monsoon 10 soil and water samples were collected from the same sampling area. To determine the initial concentration of heavy metals in the collected Shrimp, samples wereseparated intoshell, head and muscle.

Concentrations of five heavy metals (cadmium, lead, copper, nickel and chromium) in cultured Shrimp were estimated to evaluate contamination levels and health risks for Bangladeshi people.

The analyzed concentration of metals varied among region to region, pre-monsoon to post monsoon (for Shrimp), organ to organ like shell, head and muscle. Metals like Cd,Pb, Cu and Ni in Shrimp were higher in concentration than the respective maximum allowableconcentrations (MAC), whereas Cr was found below determination level. Health risks associated with this metal intake were evaluated in terms of dietary intake (EDI) and target hazard quotients (THQs). The THQ values for individual metals were below 1, suggesting that people would not experience significant health hazards. Also, the estimation showed that the carcinogenic risk (CR) of lead was exceeded the accepted risk level for Shrimp.From the health concern view, this study showed that the inhabitants who consume contaminatedShrimps are exposed chronically to metal pollution with carcinogenic and non-carcinogenicconsequences.

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# List of Abbreviations

| Serial No. | Symbols | Details |
|---|---|---|
| 1 | Cd | Cadmium |
| 2 | Pb | Lead |
| 3 | Cu | Copper |
| 4 | Ni | Nickel |
| 5 | Cr | Chromium |
| 6 | WHO | World Health Organization |
| 7 | THQ | Target hazard quotients |
| 8 | EDI | Estimated daily intake |
| 9 | CR | Carcinogenic Risk |

# Chapter 1
# Introduction

## 1.1 Background

Shrimp is one of the leading exportable products in Bangladesh. Bangladesh is earning about 500 millions of foreign currency yearly by exporting Shrimp and contributing 3.78% in GDP (DOF, 2015). To gauge the prospects of Shrimp farming, the south-western region of Bangladesh has been considered as the core farming areas. So, this is a concerning issue to determine the heavy metal concentration of Shrimp in this area as it plays valuable role in our economy. On the other hand it's also a significant issue for human health.

Borrellet al.(2016) observed concentration of trace metal in fish and crustaceans at the northern shoreline of the Bay of Bengal and assumed that human health is under concern as the concentration of heavy metal increased and exceeded the proposed health advisory levels. Kwok et al.(2014) observed the muscle and viscera of large tilapia (*Oreochromismossambicus*) and found significant bioaccumulation of Cd (Cadmium). Wu et al.(2005) observed relationship between metallothionein induction and heavy metal accumulation in white Shrimp*Litopenaeusvannamei*and found response to Cd differs from that to Zn. There are several works have been done on heavy metals and bioaccumulation all over the world. Now this is a concerning issue as it is directly related to human health when it crosses the acceptable limit. According to FAO and WHO (2002) each metal has a minimum concentration level. When it crosses the limit then it can be harmful for human health.

## 1.2 Heavy metal

'Heavy metals' is a general collective term, which applies to the group of metals and metalloids with atomic density greater than 4 $g/cm^3$, or 5 times or more, greater than water (Garbarino et al., 1995). The term 'heavy metals'refers to any metallic element which has a relatively high density and toxic or poisonous even at low concentration (Lenntech, 2004). Examples of heavy metals include mercury (Hg), cadmium (Cd), arsenic (As), chromium (Cr), thallium (Tl), and lead (Pb). Heavy metals are dangerous because they tend to bioaccumulate. Bioaccumulation in fish can be predicted by models (Svobodova et al., 2004). The acidic conditions of aquatic environment might cause free divalent ions of many heavy metals to be absorbed by fish gills (Part et al., 1985). Compounds accumulate in living things any time they are taken up and stored faster than they are broken down (metabolized) or excreted. Heavy metal is a loose term usually used to indicate environmentally ''bad'' metals. It is poorly defined with a multitude of often contradictory definitions based on density, atomic weight, atomic number or other properties of the elements or their compounds (Hodson, 2004). Heavy metal is a general collective term which applies to the group of metals and metalloids with an atomic density greater than 4g/cm$^3$ (Duffus, 2002). They are defined by the United Nations Economic Commission for Europe (UNECE) as "those metals or, in some cases, metalloids which are stable and have a density greater than 4.5 g/cm3 and their compounds" (UNECE, 1998). Heavy metals are also defined as all metals of atomic weight greater than sodium with specific gravity of more than 5.0 (Adamo et al., 1994). Alloway (1995) defines heavy metals as "elements which have an atomic density greater than 6 g/cm$^3$". Like fish, Shrimp is also an important source of protein. But now a days due to increasing the urbanization and industrialization are caused of increasing concentration of heavy metals are increasing on fish as well as Shrimp. Consumption of this kind of food is dangerous for human being. There were various works done before on this perspective.

### 1.3 Sources of heavy metals

Heavy metals are elements which occur naturally in the Earth's crust. They are therefore found naturally in soils and rocks with a subsequent range of natural background concentrations in soils, sediments, waters and organisms. Anthropogenic releases can give rise to higher concentrations of the metals relative to the normal background values. The most important anthropogenic releases of heavy metals to the environment come from metalliferous mining and smelting, agricultural materials (pesticides and fertilizers), irrigation and application of sewage water and sludge, fossil fuel combustion and metallurgical industries (Alloway, 1995; Wood and Wang, 1983).

The amounts of most heavy metals deposited to the surface of the Earth by man are many times greater than depositions from natural background sources. Combustion processes are the most important sources of heavy metals, particularly, power generation, smelting, incineration and the internal combustion engine (Hutton and Symon, 1986; Nriagu and Pacyna, 1988; Nriagu, 1989). Industrial processes that release a variety of metals into waterways, almost all industrial processes that produce waste discharges are potential sources of heavy metals to the aquatic environment (Denton et al., 2001). Domestic wastewater, sewage sludge, urban runoff, and leachate from solid waste disposal sites are also obvious sources of heavy metals into rivers, estuaries and coastal waters (Mance, 1987). A proportion of the total anthropogenic metal input in the sediments in near shore waters, adjacent to urban and industrial growth centers comes from the combustion of fossil fuels. Other potential sources include ports, harbors, marinas and mooring sites, also subjected to heavy metal inputs associated with recreational, commercial, and occasionally, military, boating, and shipping activities (Denton et al., 1997). The heavy metal content comes from natural sources (rock weathering, soil erosion, dissolution of water-soluble salts) as well as anthropogenic sources such as municipal wastewater-treatment plants, manufacturing industries, and agricultural activities etc and causes water and sediment pollution as the ultimate sink in the aquatic environment due to discharges or to hydrologic and atmospheric processes (Guven and Akıncı, 2008 and Lagadicet al., 2000).

Heavy metals are emitted both in elemental and compound (organic and inorganic) forms. Anthropogenic sources of emission are the various industrial point sources including former and present mining sites, foundries and smelters, combustion by-products and traffics (UNEP / GPA 2004). Cadmium is released as a by- product of zinc (and occasionally lead) refining; lead is emitted during its mining and smelting activities, from automobile exhausts (by combustion of petroleum fuels treated with tetraethyl lead antiknock) and from old lead paints; mercury is emitted by the degassing of the earth's crust.Generally, metals are emitted during their mining and processing activities (Lenntech, 2004).

Environmental pollution by heavy metals is very prominent in areas of mining and old mine sites and pollution reduces with increasing distance away from mining sites (Peplow, 1999). These metals are leached out and in sloppy areas, are carried by acid water downstream or run-off to the sea. Through mining activities, water bodies are most emphatically polluted (Garbarino et al., 1995; INECAR, 2000). The potential for contamination is increased when mining exposes metal-bearing ores rather than natural exposure of ore bodies through erosion (Garbarino et al. 1995) and when mined ores are dumped on the earth surfaces in manual dressing processes. Through rivers and streams, the metals are transported as either dissolved species in water or as an integral part of suspended sediments (dissolved species in water have the greatest potential of causing the most deleterious effects). They may then be stored in river bed sediments or seep into the underground water thereby contaminating water from underground sources, particularly wells; and the extent of contamination will depend on the nearness of the well to the mining site. Wells located near mining sites have been reported to contain heavy metals at levels that exceed drinking water criteria (Garbarino et al., 1995; Peplow, 1999).

**Table 1.1:** The tolerance limits of some heavy metals are shown in following table: United State Environment Protection Agency (USEPA) maximum contamination levels for heavy metal concentration in air, soil and water.

| **Heavy metal** | **Max conc. in air ($mg/m^3$)** | **Max conc. in sludge ( ppm or mg/kg)** | **Max conc. in drinking water (mg/l)** | **Max conc. in $H_2O$ supporting aquatic life (mg/l or ppm)** |
|---|---|---|---|---|
| Cd | 0.1-0.2 | 85 | .005 | 0.08 |
| Pb | -- | 420 | $0.01^{\pi}$(0.0) | .0058 |
| Zn | 1, 5* | 7500 | 5.00 | .0766 |
| Hg | -- | <1 | 0.002 | 0.05 |
| Ag | 5 | -- | 0.0 | 0.1 |
| As | -- | -- | .01 | -- |

(Value in bracket is the desirable limit; WHO ; 1adapted from U.S. - OSHA; 2 EPA, July 1992; USEPA, 1987; Georgia Code, 1993; Florida Code, 1993; Washington Code, 1992; Texas Code, 1991; North Carolina, 1991; *1 for chloride fume, 5 for oxide fume; - - no guideline available).

**Table 1.2:** Guideline in drinking water by the World Health Organization (WHO) and National Agency for Food and Drugs Administration and Control (NAFDC), Nigeria.

| **Heavy metal** | **Max. acceptable conc. (WHO)** | **Max. acceptable conc. (NAFDC)** |
|---|---|---|
| Zn | 5 mg/l | 5 mg/l |
| Arsenic | .01 mg/l | 0.0 mg/l |
| Magnesium | 50 mg/l | 30 mg/l |
| Calcium | 50 mg/l | 50 mg/l |
| Cadmium | .003 mg/l | 0.0 mg/l |
| Lead | .01 mg/l | 0.0 mg/l |
| Silver | 0.0 mg/l | 0.0 mg/l |
| Mercury | .001 mg/l | 0.0 mg/l |

**Table 1.3:Maximum Allowable Concentration (MAC)**

According to FAO and WHO 2002 the maximum allowable concentration of heavy metal is given below-

| **Heavy metals** | **MAC (mg/kg)** |
|---|---|
| Cr | 1.0 |
| Ni | 0.8 |
| Cu | 4.5 |
| As | 1.0 |
| Cd | 0.1 |
| Pb | 0.5 |

## 1.4 Toxicity of Heavy Metals

The toxicity of a metal is usually defined in terms of the concentration required to cause an acute response (usually death) or a sub-lethal response (Smith, 1986). Environmental pollution with toxic metals is becoming a global phenomenon. As a result of the increasing concern with the potential effects of the metallic contaminants on human health and the environment, the research on fundamental, applied and health aspects of trace metals in the environment is increasing (Vernet, 1991). Heightened concern for reduction in environmental pollution over the past decades has stimulated active continuing research and literature on the toxicology of heavy metals. Virtually all metals can produce toxicity when ingested in sufficient quantities, but there are several of them which are especially important because they produce toxicity at low concentrations (Hoekman, 2008).

Heavy metals become toxic when they are not metabolized by the body and accumulate in the soft tissues. They may enter the human body through food, air, or absorption through the skin when they come in contact. In general, heavy metals produce their toxicity in organisms by forming complexes or "ligands" with organic compounds (Soghoian, 2008; Hoekman, 2008). These modified biological molecules lose their ability to function properly, and result in malfunction or death of the affected cells. Some heavy metals may form complexes with other materials in living organisms. These complexes may inactivate some important enzymes, systems and certain protein structures (Hoekman, 2008). The presence of heavy metals above a certain threshold can be injurious to human health and the environment, particularly communities without alternative sources of drinking water who continue to depend on polluted streams (Akabzaa and Darimani, 2001). Description and toxic effects of some heavy metals are given below.

### 1.4.1 Cadmium (Cd)

Cadmium is a common impurity as complex oxides, sulfides, and carbonates in zinc, lead and copper ores and it is most often isolated during the production of zinc. Some zinc ores concentrates from sulfidic zinc ores contain up to 1.4 % of cadmium (Finkelman, 2005). Cadmium is widely distributed in the Earth's crust, the atmosphere, marine sediment and sea. Environmental levels of cadmium occur following the natural weathering of minerals, forest fires and volcanoes, although larger amounts are released following human activities. These include the application of phosphate fertilisers, fossil fuel combustion, the production of iron, steel and non-ferrous metals, cement production and waste incineration. The anthropogenic sources of cadmium contribute to human exposure to a greater extent due to production, use and disposal of cadmium products and the incineration of cadmium-containing products. Cadmium is prevalent in the three main environmental compartments, namely air, water and soil. The majority of cadmium exposure arises from air and soil, by atmospheric deposition and by the ingestion of vegetables such as lettuce, spinach, celery and cabbage that accumulate cadmium. Foods such as potatoes and peas take up less amounts. Minimal exposure of cadmium arises from water. Cadmium also exists as a number of compounds. Cadmium oxide is of most interest for health effects following inhalation exposure, as it is the main form of airborne cadmium. Both cadmium oxide and cadmium carbonate have similar toxicological profiles as soluble cadmium. Cadmium bound to metallothionein is of interest as they are found in relatively high concentrations of organ meat i.e. liver and kidney. The inhalation of cadmium also contributes to the total cadmium burden, albeit to a lesser extent than oral intake, with the exception of smokers or those undergoing occupational exposure. Cigarette smoke considerably adds to cadmium exposure (HPA, 2010).

Cadmium is extremely toxic to most plants and animal species particularly in the form of free cadmium ions (Denton et al., 1997). The major effects of cadmium poisoning are experienced in the lungs, kidneys and bones. Acute inhalation of cadmium may initially cause irritation of the upper respiratory tract, although symptoms may be delayed for 4-8 hours. Dyspnoea, chest pain and muscle weakness may also occur. Pulmonary oedema, bronchitis, chemical pneumonitis, respiratory failure, toxemia in the liver and death may occur within days of exposure. In the long-term following exposure, progressive pulmonary fibrosis and impaired lung function may occur. Chronic inhalation of cadmium compounds as fumes or dust produce pulmonary emphysema, where the small air sacs of the lungs become distended and eventually destroyed reducing lung capacity (Ansari et al., 2004 and HPA, 2010). According to the Priority List of Hazardous Substances, 2013 Cadmium (Cd) ranking is 7 and its point is 1319 (ATSDR, 2013).

### 1.4.2 Lead (Pb)

Lead is defined by the United States Environmental Protection Agency (USEPA) as potentially hazardous to most forms of life, and is considered toxic and relatively accessible to aquatic organisms (USEPA, 1986). Lead is a naturally occurring toxic metal found in the Earth's crust. Its widespread use has resulted in extensive environmental contamination, human exposure and significant public health problems in many parts of the world. Important sources of environmental contamination include mining, smelting, manufacturing and recycling activities, and, in some countries, the continued use of leaded paint and leaded gasoline. More than three quarters of global lead consumption is for the manufacture of lead-acid batteries for motor vehicles. Lead is, however, also used in many other products, for example pigments, paints, solder, stained glass, crystal vessels, ammunition, ceramic glazes, jewellery, toys and in some cosmetics and traditional medicines. Drinking water delivered through lead pipes or pipes joined with lead solder may contain lead. Much of the lead in global commerce is now obtained from recycling (WHO, 2013). The main routes of systemic exposure are predominantly via ingestion or inhalation. Exposure to inorganic lead occurs primarily through ingestion of food and drinking water, although exposure via soil and dust, air, and chipped leaded paint significantly contributes to the overall exposure (IPCS, 1995).

Lead is toxic and a major hazard to human and animals. Lead has two quite distinct toxic effects on human beings, physiological and neurological. The relatively immediate effects of acute lead poisoning are ill defined symptoms, which include nausea, vomiting, abdominal pains, anorexia, constipation, insomnia, anemia, irritability, mood disturbances and coordination loss. In more severe situations neurological effects such as restlessness, hyperactivity, confusion and impairment of memory can result as well as coma and death (Ansari et al., 2004). According to the Priority List of Hazardous Substances, 2013 Lead (Pb) ranking is 2 and its point is 1529 (ATSDR, 2013).

### 1.4.3 Copper (Cu)

Copper is a metallic element that occurs naturally as the free metal, or associated with other elements in compounds that comprise various minerals. It is an essential micro-nutrient required in the growth of both plants and animals. In humans, it helps in the production of blood haemoglobin. In plants, copper is especially important in seed production, disease resistance and regulation of water. Copper occurs in nature in its metallic form and in ores and minerals. Copper and its alloys are now used extensively in domestic and other plumbing systems and to make cooking utensils. Copper is also used in the production of electrical wire and microelectronic applications, in electroplating and photography, as a roofing material, and as a catalyst in the chemical industry. Exposure of humans to copper occurs primarily from the consumption of food and drinking water. The relative copper intake from food versus water depends on geographical location; generally, about 20–25% of copper intake comes from drinking water (Stern et al., 2007).

Exposure to excessive levels of copper can result in a number of adverse health effects including liver and kidney damage, anemia, immunotoxicity, and developmental toxicity. One of the most commonly reported adverse health effect of copper is gastrointestinal distress. Nausea, vomiting, and/or abdominal pain have been reported, usually occurring shortly after drinking a copper sulfate solution, beverages that were stored in a copper or untinned brass container, or first draw water. The observed effects are not usually persistent and gastrointestinal effects have not been linked with other health effects. Animal studies have also reported gastrointestinal effects (hyperplasia of fore stomach mucosa) following ingestion of copper sulfate in the diet. Copper is also irritating to the respiratory tract. Coughing, sneezing, runny nose, pulmonary fibrosis, and increased vascularity of the nasal mucosa have been reported in workers exposed to copper dust. The liver is also a sensitive target of toxicity. Liver damage (necrosis, fibrosis, abnormal biomarkers of liver damage) have been reported in individuals ingesting lethal doses of copper sulfate. According to the Priority List of Hazardous Substances, 2013 Copper (Cu) ranking is 118 and its point is 807 (ATSDR, 2013).

### 1.4.4 Nickel (Ni)

Nickel occurs naturally in the earth crust and is ubiquitous in air, water, soil and the biosphere. The average concentration of nickel in the earth's crust is 0.008%. Nickel also exists as a number of compounds. Nickel compounds that are soluble in water include nickel chloride and nickel sulphate; insoluble nickel compounds include nickel oxide, nickel sulphide and nickel subsulphide. Nickel carbonyl is a highly toxic, volatile liquid that has specialised industrial uses. Nickel is emitted from natural sources including windblown dust, volcanoes, vegetation, forest fires and meteoric dust. The principle anthropogenic sources of nickel emissions include the combustion of coal and oil, municipal incineration, steel and other nickel alloy production and electroplating (PHE, 2009).

Nickel is moderately toxic to most species of aquatic plants, though it is one of the least toxic inorganic agents to invertebrates and fish. The major source of discharge to natural waters is municipal wastewater followed by smelting and the refining of nonferrous metals (Denton et al., 2001). Also mine drainage effluents are known to be major contributors due to high concentrations of nickel found in the discharges (Finkelman, 2005). Typically, nickel residues in sediments can be up to 100 µg/g or higher but may fall below 1 µg/g in some clean coastal waters (Denton et al., 1997) with the average concentration of nickel in the lithosphere of 55 µg/g (Callender, 2003).

Nickel carbonyl is the most toxic nickel compound following acute inhalation exposure in humans. The effects of nickel carbonyl inhalation occur in two phases, immediate and delayed. The immediate effects include respiratory tract irritation and neurological effects such as dizziness and headache, following which there is often an asymptomatic period before the onset of the delayed pulmonary symptoms, including chest pain, cough and dyspnoea. In severe cases pulmonary oedema, pneumonitis and death may occur. Patients who survive a severe exposure to nickel carbonyl may develop weakness and neurasthenic syndrome. Acute ingestion of nickel compounds may cause nausea, vomiting, diarrhoea, headache, cough and shortness of breath. In severe cases, ingestion of large amounts of a nickel compound may cause death. Chronic oral exposure to nickel or nickel compounds has not been characterised in humans. Dermal exposure to nickel salts can cause skin irritation. Nickel and its water soluble salts are potent skin sensitisers (PHE, 2009). According to the Priority List of Hazardous Substances, 2013 nickel (Ni) ranking is 57 and its point is 996 (ATSDR, 2013).

### 1.4.5 Chromium (Cr)

Chromium is the 21st most abundant element in Earth's crust with an average concentration of 100 mg/kg. Chromium compounds are found in the environment, due to erosion of chromium containing rocks and can be distributed by volcanic eruptions. The concentrations range in soil is between 1 and 3000 mg/kg, in sea water 5 to 800 µg/L, and in rivers and lakes 26 µg/L to 5.2 mg/L. Chromium like zinc, is one of the most abundant heavy metals in the lithosphere with an average concentration of about 69 µg/g and mercury content in carbonate sediments is reported to be 0.03 µg/g (Callender, 2003). Chromium occurs naturally in the Earth's crust, predominately in the trivalent, chromium (III), form, and it is ubiquitous in air, water, soil and biological materials. Chromium (VI) compounds are essentially anthropogenically produced and do not occur naturally in the environment. Large amounts are produced through a range of activities, including the production of chromates and bichromates, stainless steel, welding, chromium plating, ferrochrome alloys and chrome pigment production, material tanning, the combustion of coal and oil, cement works, and waste incineration and released into various environmental media. The general population may be exposed to chromium by inhaling ambient air, or ingesting food and drinking water that contain chromium. Exposure may also occur through skin contact with certain consumer products containing chromium, e.g. some wood preservatives, cement, cleaning materials, textiles and leather tanned using chromium and via cigarette smoke (HPA, 2007).

Chromium is moderately toxic to aquatic organisms. Major contributors of chromium in the aquatic environment are dominated by input urban runoff, domestic and industrial wastewaters and sewage sludge (Denton et al., 1997). Chromium is carcinogenic to humans and the toxicity of chromium depends on the oxidation state, chromium (VI) being more toxic than the trivalent form chromium (III). In addition, chromium (VI) is the more readily absorbed by both inhalation and oral routes.

Although effects on the kidney, gastrointestinal tract and liver have also been reported, acute ingestion of high doses of chromium (VI) compounds, the exact quantity of which is not usually known, results in acute, potentially fatal, effects in the respiratory, cardiovascular, gastrointestinal, hepatic, renal, and neurological systems. Chronic exposure to chromium (III) resulted in weight loss, anaemia, liver dysfunction and renal failure (HPA, 2007). According to the Priority List of Hazardous Substances, 2013 Chromium (Cr) ranking is 17 and its point is 1147 (ATSDR, 2013).

### 1.5 Rationale

The people of Bangladesh, one of the poorest and most densely populated countries in the world with a population of 160 million people and the population is increasing drastically. The most important food crops for this huge population of this country are rice and fish. Fish plays a significant role among the population in Bangladesh for supplying protein, essential vitamins, minerals and fatty acids. Fish accounts for about

70% of the animal protein uptake with annual fish consumption of about 14 kg per person (ADB, 2005). The average per capita fish consumption is lower than the world average of 16.1 kg a year (Hishamunda et al., 2008). Nevertheless, the importance of aquaculture as a source of food has been well recognized in Bangladesh.

Shrimp is called as “white gold” of Bangladesh. Shrimp is the second most important export item toBangladesh. The cultured (cultivated) Shrimp constitutes more than 95% of the total Shrimp export. The main cultured species is the tiger Shrimp (locally known as *bagda*Shrimp) of which the technical name is *Penaeusmonodon*. It is a marine Shrimp and is cultivated in brackish water. Farmers are using water from the nearby river or ground water. In Mongla, farmers are using water from Posur river. Around the river there are lots of industries like cement industry, food industry, chemical industry and others. The wastage of these industries are fall down in the river and the river water is directly used in the gher. So the water has much possibility to have heavy metal accumulation and the Shrimp can accumulate it in their body. On the other hand Shrimp are bottom feeder. The gher are cultivated for many decades. For this large time soil can accumulate heavy metal from various sources. From that Shrimp can accumulate heavy metal. If we don’t consider these circumstances it would have a great impact on our foreign currency and our economy.

Considering all those prospects of demand, Shrimp was selected to conduct this research study. Many researchers have worked on the bioaccumulation of the heavy metals in fish collected from various areas all over the world and determined different heavy metal concentration accumulated from the environment the fish lived but there is not any research regarding heavy metal bioaccumulation on Shrimp by water or soil of the river which is polluted by industrial wastage.

## 1.6 Objectives

The overall objective of the proposed research work was to determine the heavy metals in cultured Shrimp and their impact on human health.

The specific objectives are-

- To determine the heavy metal concentrations of water, soil and Shrimp from sampled area.
- To determine the accumulation of different heavy metal on shell, head and muscle of the Shrimp sample.
- To assess the human health risk due to consumption of heavy metals contaminated Shrimp.

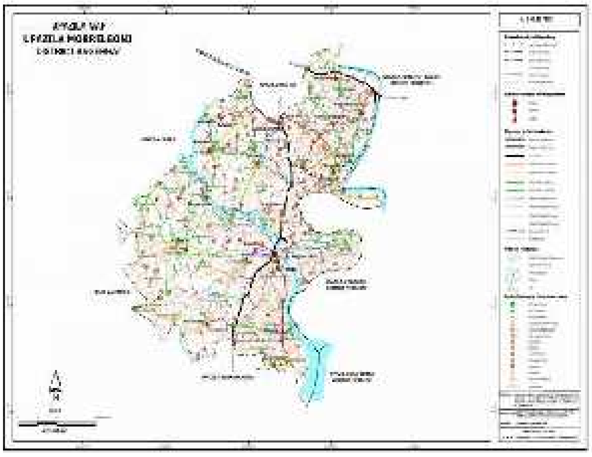
# Chapter 2

# Materials and Methods

## 2.1 Selection of sampling site

The aim of the study was to evaluate the accumulation of heavy metal in Shrimp. For this reason the Shrimp farming area of south-east region (Morrelganj, Mongla and Satkhira Sadar) has been selected to conduct the following research work as there are several chemical, cement industries along to the Posur river. Industrial pollution and consequent heavy metal released into water bodies from these industries can bio-accumulate in fish as well as Shrimp and could be transferred into food chain. These soil, water and Shrimp samples were collected from those areas of Satkhira Sadar, Morrelganj and Mongla.


**A. Map of Satkhira Sadar**



**B. Map of Morrelganj**

**C. Map of Mongla**

**Fig 2.1. Maps of sampling area (Satkhira Sadar, Morrelganj and Mongla)**

## 2.2 Sample collection

Shrimps samples were collected in pre-monsoon and post monsoon season. Soil and water samples were collected only in pre-monsoon season. Soil, water and Shrimp were collected from 10 different places of Satkhira Sadar, Morrelganj and Mongla. 3 samples from Satkhira Sadar, 4 samples from Morrelganj and 3 samples from Mongla were collected. Soil samples were collected into polythene bags. Water was collected into plastic bottles. And Shrimp were collected into plastic bag and kept into ice bag. Then the samples were brought to the laboratory the day after sampling.

**A. Gher of Shrimp**

**B. Collected Shrimp from the gher**

**C. Collected Soil from the gher**

**D. Collected Soil from the gher**

E. Collected water from the gher

F. Collected water from the gher

**Plate 2.1. Photographs of collecting samples from different area (A-F)**

**2.2.1 Soil sample preparation**

Collected soil samples were dried in sun after those were being transported to the laboratory. After sun drying of the samples, the larger aggregates were broken gently, preferably in a mortar and pestle and the ground soil was passed through a 2 mm sieve. The sieved soil was then weighed in to plastic containers and mouths were well capped. Each containers shows location, sample area, sample number, date and gher area. The containers were stored in a cool dry place in the laboratory.

Accurately weighted samples (air-dried) of about 1.0g were digested for heavy metal determination using nitric acid and perchloric acid digestion system (Huq and Didar, 2005). At first 1.0 g dry soil sample was taken into beaker and 10 ml of nitric acid was mixed with the sample. The mixture was then put into fume chamber and digested at 120°C until the solution become clear. Then 5 ml perchloric added to the mixture. Finally mixture was then put into fume chamber and digested at 120°C until the solution become clear. Then the solution was diluted to 50 ml with deionized water and was filtered and collected into clean and sterilized plastic bottles for further analysis of heavy metals in the sample (Huq and Didar, 2005). The process of soil sample preparation is shown in the following flow chart:

Soil sample was kept in sun dry

⇩

Sun dried samples were homogenized using morter pestle

⇩

1.0g of the sun dried soil samples were taken and 10 ml of nitric acid was mixed with the sample

⇩

The mixture was digested at 120˚C in fume chamber

⇩

Then 5.0 ml perchloric acid was mixed with the sample

⇩

The mixture was digested at 120˚C in fume chamber

⇩

Samples were diluted to 50 ml deionized water

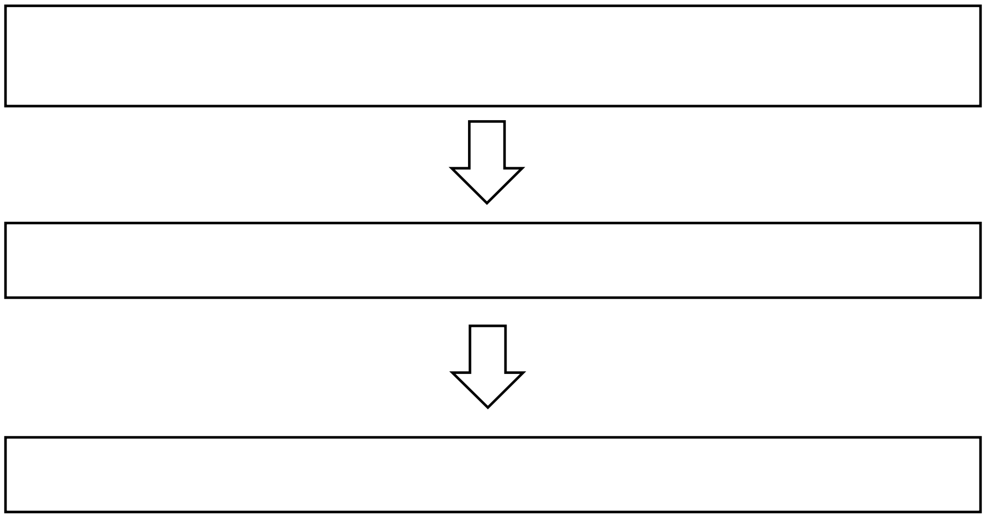
⇩

The solution was filtered and collected into clean and sterilized bottles

**Flow Chart 1. Preparation of soil for heavy metal determination**

### 2.2.2 Water sample preparation

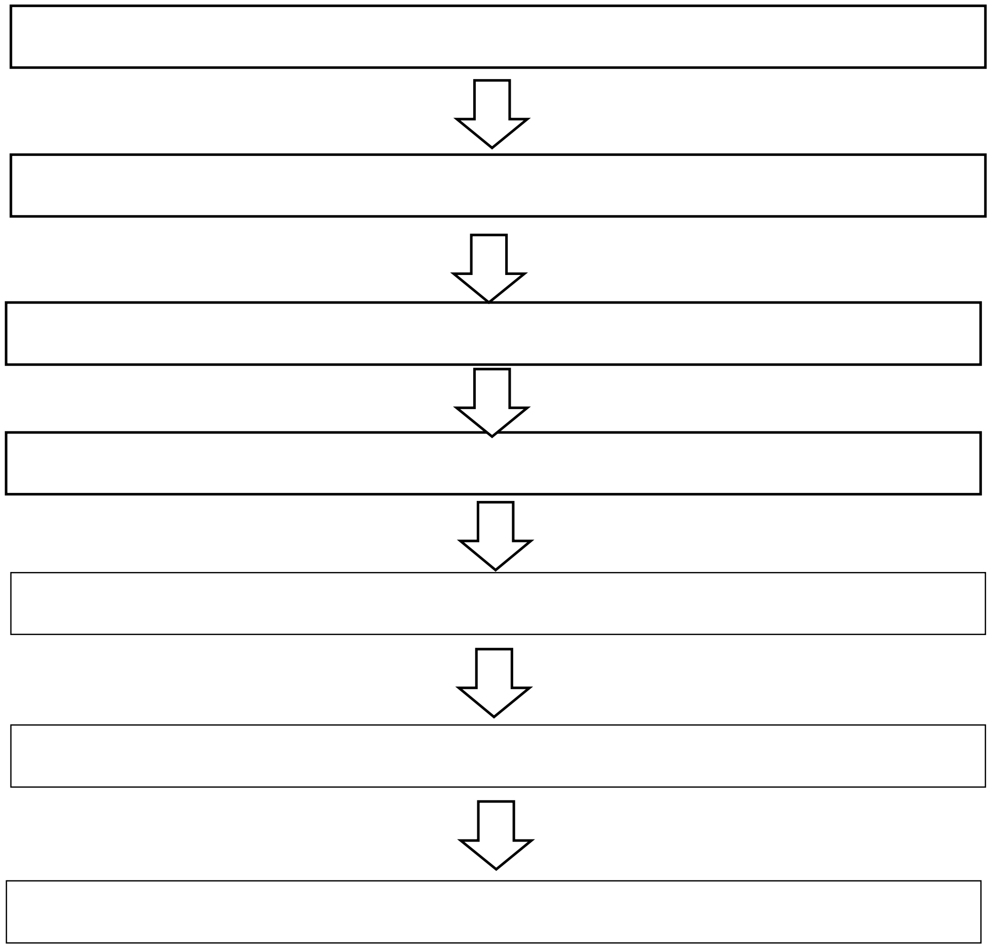
Water sample were collected in clean plastic in clean plastic bottle after rinsing the bottle 2 to 3 times with water being collected and acidified with conc. nitric acid. 80 ml water and 10 ml nitric acid was taken to a beaker and then digested for 30 minute. Then the water was filtered and the filtered water was collected into other clean and sterilized plastic bottle and reserved in cold place in laboratory (Huq and Didar, 2005).



**Flow Chart 2. Water samples preparation for heavy metal determination**

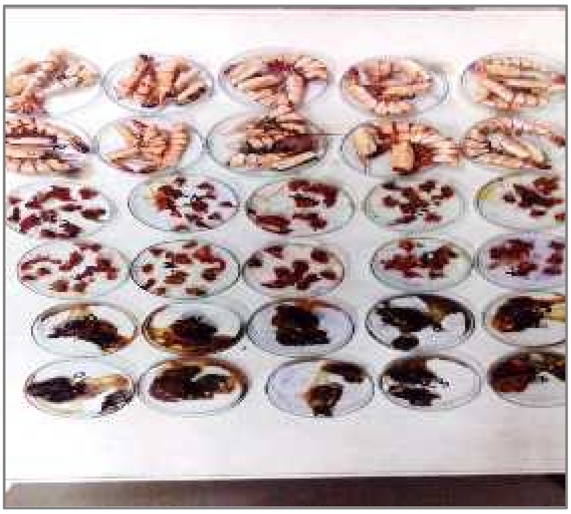
### 2.2.3 Shrimp sample preparation

To determine the initial concentration of heavy metals in the collected Shrimp, samples were taken and separated the muscle, shell and head. Then the muscle, shell and head fish was cut into small part. Then wet sample was taken into an oven and dried at 60°C for 48 hours. After the sample was completely dried then it was grinded. 1.0g of dry sample was taken into a beaker and 10 ml of nitric acid was mixed with the sample. The mixer was then put into Fume Chamber and digested at 12°C until the solution become clear. Then 5 ml perchloric added to the mixture. Finally mixture was then put into fume chamber and digested at 120°C until the solution become clear. Then the solution was diluted to 50 ml with deionized water and was filtered and collected into clean and sterilized plastic bottles for further analysis of heavy metals in the sample (Huq and Didar, 2005). The process of Shrimp sample preparation is shown in the following flow chart:



**Flow Chart 3.Shrimp sample preparation for heavy metal determination**

A.Oven used for drying



B. Dried Shrimp sample

C. Electric balance

D. Electric balance

**Plate 2.2. Photographs of sample preparation for heavy metal determination (A-D)**

## 2.3 Determination of heavy metal

Shrimp, soil and water samples were analyzed at Soil, Agronomy and Environment Section; Biological Research Division, BCSIR, Dhaka. The heavy metals Cr, Pb, Cd, Ni and Cu were analyzed by using Atomic Absorption Spectrometer (Model No: AA-7000, Shimadzu). The equipment was calibrated with chemical standard solutions prepared from commercially available chemicals and reagents used for the preparation for the samples were analytical grade and deionized water was used throughout the study. Heavy metals concentrations were expressed as ppm.

Heavy metal was determined using following formula (Huq and Didar, 2005):

Concentration of heavy metals = (Reading – Blank reading) ×PDF×SDF

Where, Primary Dilution Factor (PDF) = $\frac{\text{Volume}}{\text{Weight of sample}}$

Secondary Dilution Factor (SDF) = $\frac{\text{Secondary Volume}}{\text{Secondary Weight of sample}}$

**A. Digestion of samples in fume chamber**

**B. Filtering of samples**

**C. Collecting samples in plastic bottles**

**D. Determination of heavy metals in Atomic Absorption Spectrophotometer**

**Plate 2.3. Sample preparation for heavy metal determination in Atomic Absorption Spectrophotometer (A-D)**

## 2.4 Estimated Daily Intake (EDI)

The estimated daily intake (EDI) of heavy metals in Shrimps was measured using metal concentration in Shrimp, daily consumption (Rural, Urban and Total) and body weight. Following equation was used for calculation of EDI. (Shaheen at el. 2015)

Estimated daily intake (EDI) = $\frac{FIR \times C}{BW}$

Where,

FIR = Fish/Shrimp ingestion rate (Rural 45.8 g/person/day, Urban 59.9 g/person/day, On average 49.5 g/person/day)

C = Heavy metal concentration in Shrimp (ppm)

BW = Average body weight (60 Kg)

## 2.5 Target hazard quotient

The target hazard quotient (THQ) is an estimate of the risk level of non-carcinogenic due to pollutant exposure. Based on the USEPA (1989), we assumed that the ingestion dose is equal to the adsorbed contaminant dose and that cooking has no effect on the contaminants. In this study, the non-carcinogenic health risks associated with the consumption of Shrimp species by the local inhabitants (low, medium and high fish consumers) were assessed based on the target hazard quotients (THQs) and calculations were made using the standard assumption for an integrate USEPA risk analysis.

THQ = $\frac{EFr \times ED \times FIR \times C}{RfD \times BW \times AT} \times 10^{-3}$

Where THQ is the target hazard quotient

EFr = Exposure frequency (365 days/year)

ED = Exposure duration (70 years)

FIR = Fish/Shrimp ingestion rate (Rural 45.8 g/person/day, Urban 59.9 g/person/day, On average 49.5 g/person/day)

C = Heavy metal concentration in Shrimp (ppm)

BW = Average body weight (60 Kg)

AT = average exposure time for non-carcinogens (EF×ED) (365 days/year for 70 years)

RfD = The oral reference doses were based on 1.5, 0.02, 0.04, 0.0005 and 0.0035 mg/kg/day for Cr, Ni, Cu, Cd and Pb, respectively (USEPA, 2010)

**2.6 Target cancer risk**

According to USEPA (1989), for carcinogens, risks were estimated as the incremental probability of an individual to develop cancer over a lifetime exposure to that potential carcinogen. Target carcinogenic risk (TR) was calculated by using following equation.

$$TR = \frac{EFr \times ED \times FIR \times C \times CSf}{BW \times AT} \times 10^{-3}$$

Where, $CSF_0$ is the oral carcinogenic factor. ($CSF_0$ of Pb = 8.5 ×$10^{-3}$ ) (USEPA, 2010)

**2.7 Statistical analysis**

The data were statistically analyzed using the statistical software, SPSS 20.0 (SPSS, USA) and the graphs were made MS Excel. The data has been presented as mean ± SD (Standard deviation) with 5% level of significance (ANOVA; $p < 0.05$). Tukey's post-hoc tests for multiple comparisons and one way ANOVA were performed. Paired sample t-test was performed (t-test; $p < 0.05$).

# Chapter 3

# Results

### 3.1 Pre-monsoon

Pre-monsoon rain is sharp and intense and gets over for the day, after just one spell.Pre-monsoon season is synonymous with heat and humidity with uncomfortable conditions throughout the day and night.In pre-monsoon (May-June) period Shrimp, soil and water was collected from the sampling area.

#### 3.1.1 Determination of heavy metals in collected Shrimp organs

Fig.3.1 shows that the concentration of Cadmium in Shrimp shell was found highest in Morrelganj (0.20 ppm) and lowest in Mongla (0.10 ppm). In Shrimp head, highest concentration was found in Morrelganj (0.72 ppm) and lowest concentration found in Satkhira Sadar region (0.52 ppm). In Shrimp Muscle, highest concentration was found in Mongla (0.16 ppm) and lowest concentration found in Satkhira Sadar (0.12 ppm). There were significant differences found among Shell, Head and Muscle within region ($p < 0.05$).

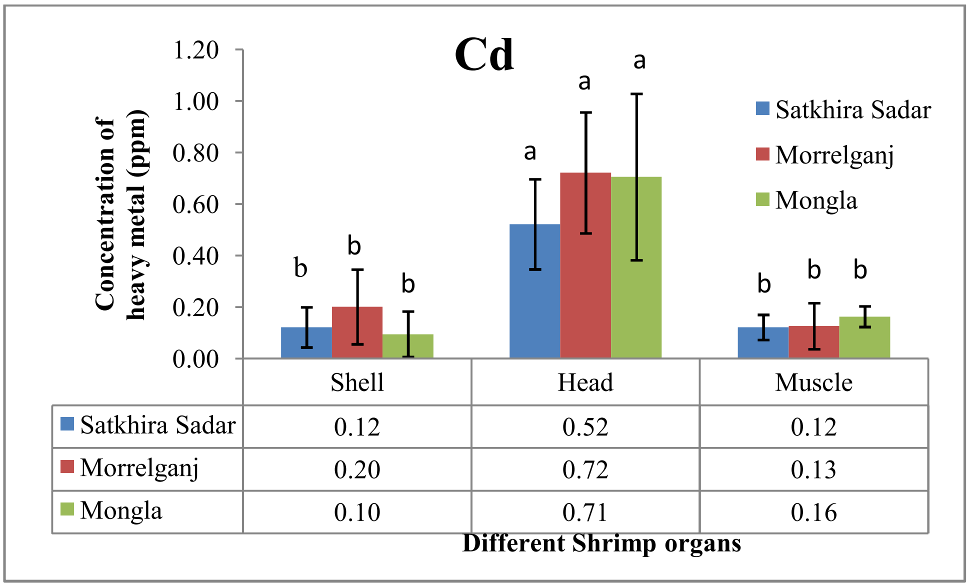



**Fig. 3.1. Mean (± SD) concentration of the Cadmium (Cd) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

Fig. 3.2 shows that the concentration of Lead in Shrimp Shell was found highest in Mongla (8.19 ppm) and lowest in Satkhira Sadar (3.90 ppm). In Shrimp head, highest concentration was found in Mongla (3.22 ppm) and lowest concentration found in Morrelganj (0.54 ppm). In Shrimp Muscle, highest concentration was found in Morrelganj (1.37 ppm) and lowest concentration found in Satkhira Sadar (0 ppm). There were significant differences found among Shell, Head and Muscle within ($p< 0.05$).

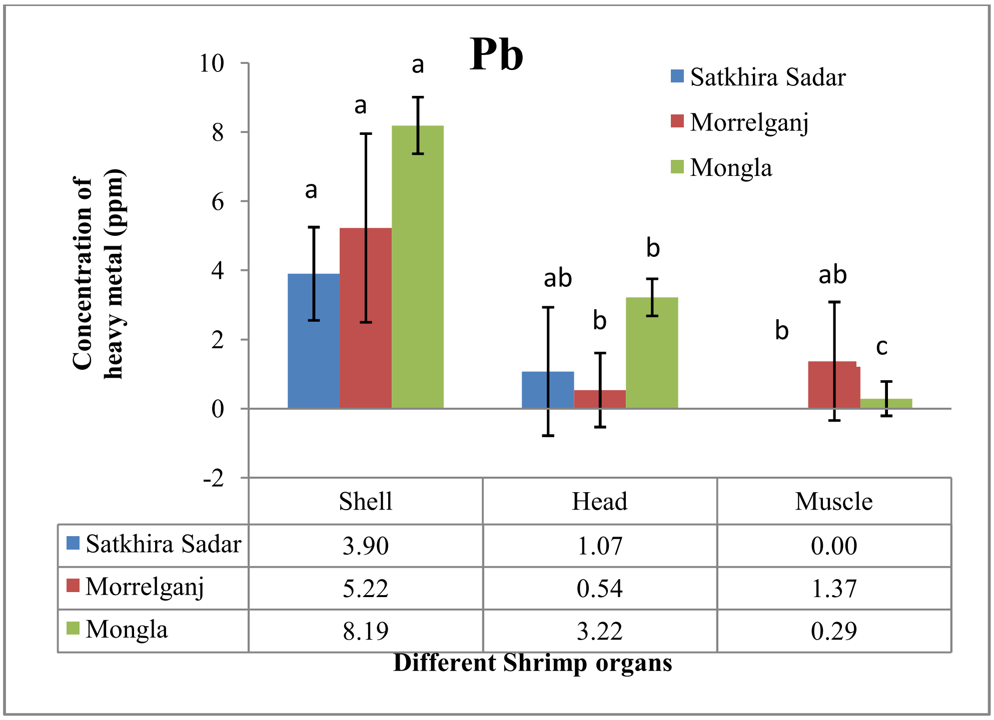
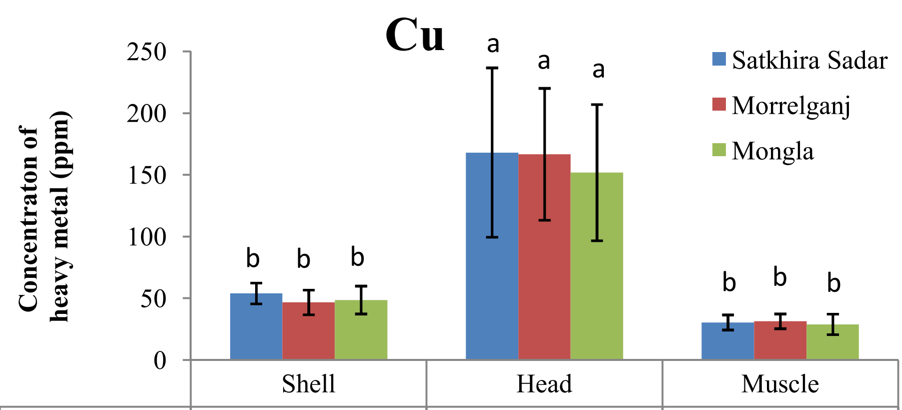



**Fig.3.2. Mean (± SD) concentration of the Lead (Pb) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

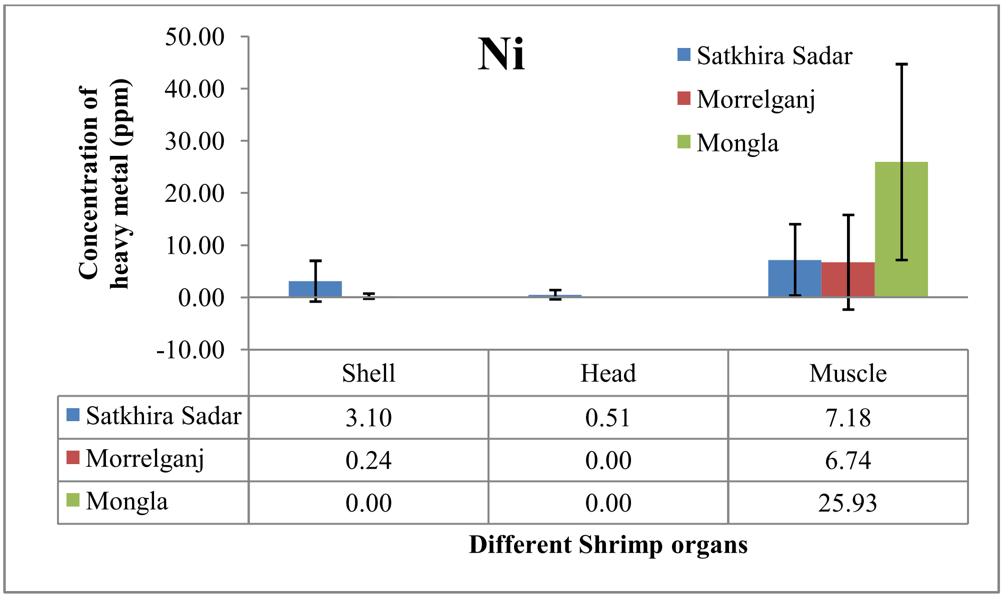
Fig.3.3 shows that the concentration of Copper in Shrimp shell was found highest in Satkhira Sadar (53.94 ppm) and lowest in Morrelganj (46.74 ppm). In Shrimp head, highest concentration was found in Satkhira Sadar (168.05 ppm) and lowest concentration found in Mongla (151.86 ppm). In Shrimp Muscle, highest concentration was found in Morrelganj (31.41 ppm) and lowest concentration found in Mongla (28.86 ppm). There were significant differences found among Shell, Head and Muscle within region ($p< 0.05$).




| | Shell | Head | Muscle |
|---|---|---|---|
| Satkhira Sadar | 53.94 | 168.05 | 30.48 |
| Morrelganj | 46.74 | 166.63 | 31.41 |
| Mongla | 48.65 | 151.86 | 28.86 |

**Fig.3.3. Mean (± SD) concentration of the Copper (Cu) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

Fig.3.4 shows that the concentration of Nickel in shell was found highest in Satkhira Sadar (3.10 ppm) and lowest in Mongla (0 ppm). In head, highest concentration was found in Satkhira Sadar (0.51 ppm) and lowest concentration found in Mongla and Morrelganj (below detectable limit). In Muscle, highest concentration was found in Mongla (25.93 ppm) and lowest concentration found in Morrelganj (6.74 ppm). There was no significant difference found among Shell, Head and Muscle within region ($p< 0.05$).



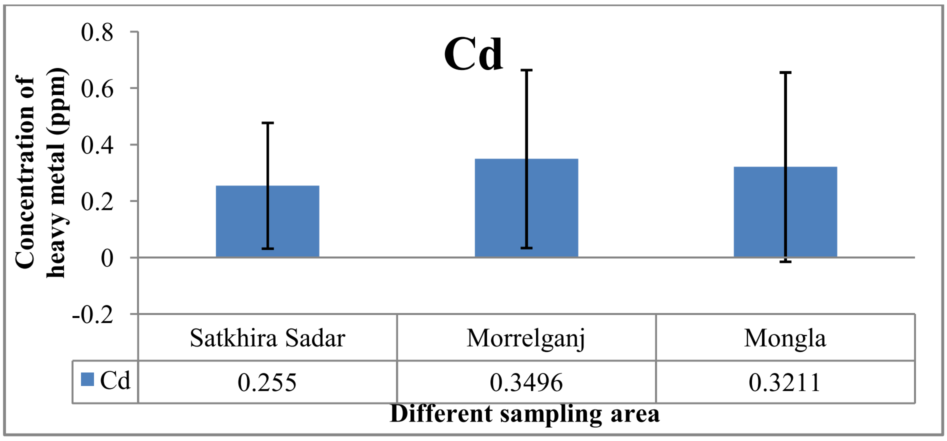

| | Shell | Head | Muscle |
|---|---|---|---|
| Satkhira Sadar | 3.10 | 0.51 | 7.18 |
| Morrelganj | 0.24 | 0.00 | 6.74 |
| Mongla | 0.00 | 0.00 | 25.93 |

**Fig.3.4. Mean (± SD) concentration of the Nickel (Ni) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, p<0.05).**

The concentration of Chromium (Cr) was Below Detection Level (BDL) in Shell, Head and muscle of Shrimp of various regions.

### 3.1.2 Overall concentration of heavy metal in Shrimp of various regions

Fig.3.5 shows that the mean concentration (shell, head and muscle) of Cadmium in Shrimp was found highest in Morrelganj (0.35 ppm) and lowest in Satkhira Sadar (0.26 ppm). There was no significant difference found among regions (p< 0.05).




| | Satkhira Sadar | Morrelganj | Mongla |
|---|---|---|---|
| Cd | 0.255 | 0.3496 | 0.3211 |

**Fig.3.5. Mean (± SD) concentration of the Cadmium (Cd) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, p<0.05).**

Fig.3.6 shows that the mean concentration (shell, head and muscle) of Lead in Shrimp was found highest in Mongla (3.90 ppm) and lowest in Satkhira Sadar (1.66 ppm). There was no significant difference found among regions ($p < 0.05$).

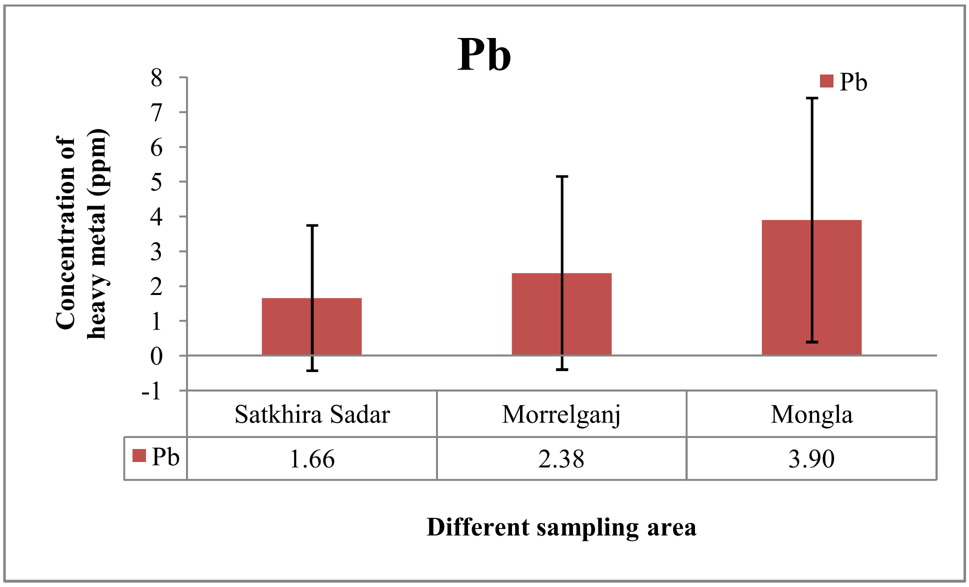



**Fig.3.6. Mean (± SD) concentration of the Lead (Pb) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, p<0.05).**

Fig.3.7 shows that the mean concentration (shell, head and muscle) of Copper in Shrimp was found highest in Satkhira Sadar (84.16 ppm) and lowest in Mongla (76.46 ppm). There was no significant differencefound among regions ($p < 0.05$).

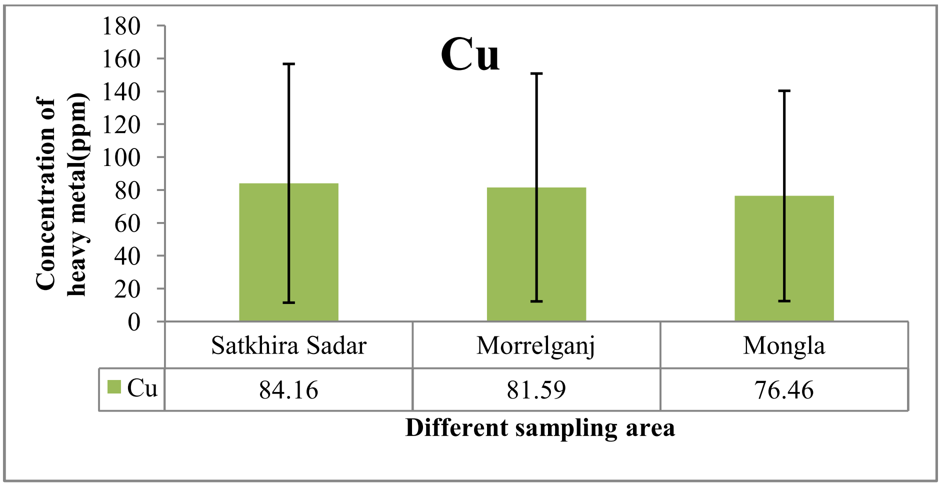



**Fig.3.7. Mean (± SD) concentration of the Copper (Cu) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, p<0.05).**

Fig.3.8 shows that the mean concentration (shell, head and muscle) of Nickel in Shrimp was found highest in Mongla (8.64 ppm) and lowest in Morrelganj (2.33 ppm). There was no significant difference found among regions ($p < 0.05$).

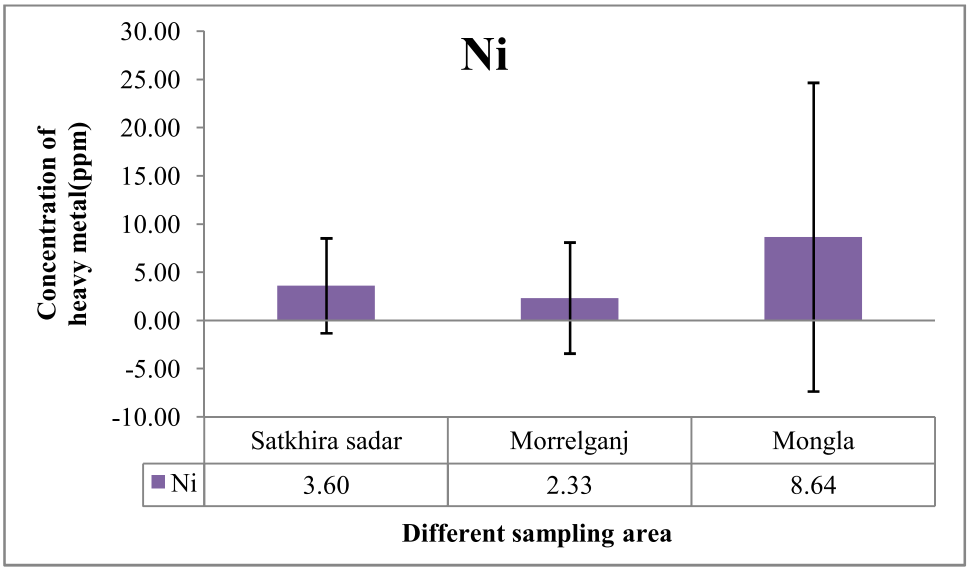



**Fig.3.8. Mean (± SD) concentration of the Nickel (Ni) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, $p<0.05$).**

The concentration of Chromium (Cr) was Below Detection Level (BDL) in Shell, Head and muscle of Shrimp of various regions. So the overall concentration was also below detectable limit.

### 3.1.3 Metal concentration in Soil, Water and Shrimp

Fig.3.9 shows that the concentration of Cadmium in soil was found highest in Morrelganj (0.32 ppm) and lowest in Mongla (0.02 ppm). In water,the concentration of Cadmium (Cd) was Below Detectable Level (BDL) in various. In Shrimp, highest concentration was found in Morrelganj (0.35 ppm) and lowest concentration found in Satkhira Sadar (0.26 ppm). There were significant differences found among Soil, Water and Shrimp within region ($p < 0.05$).

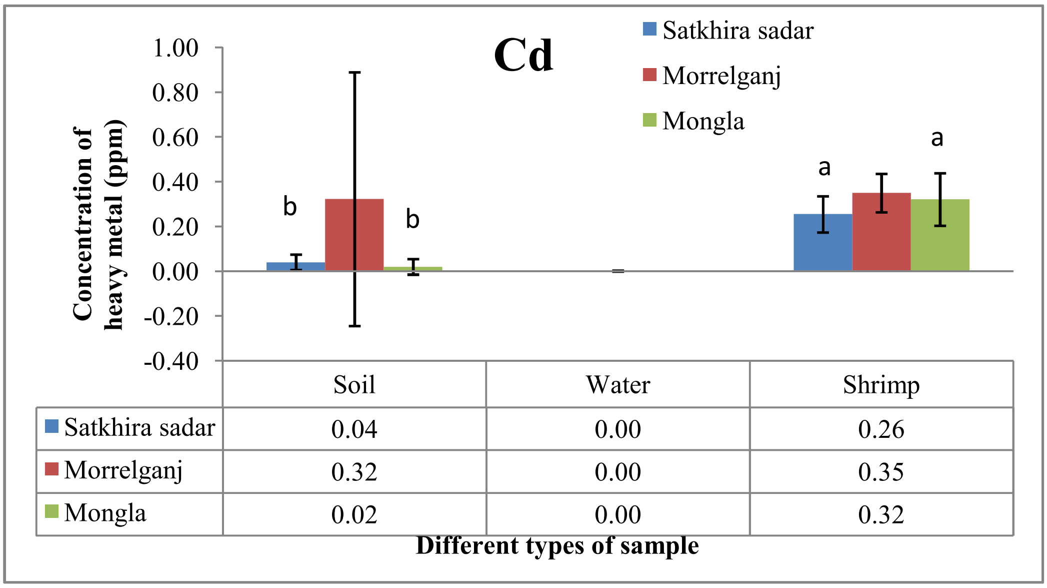
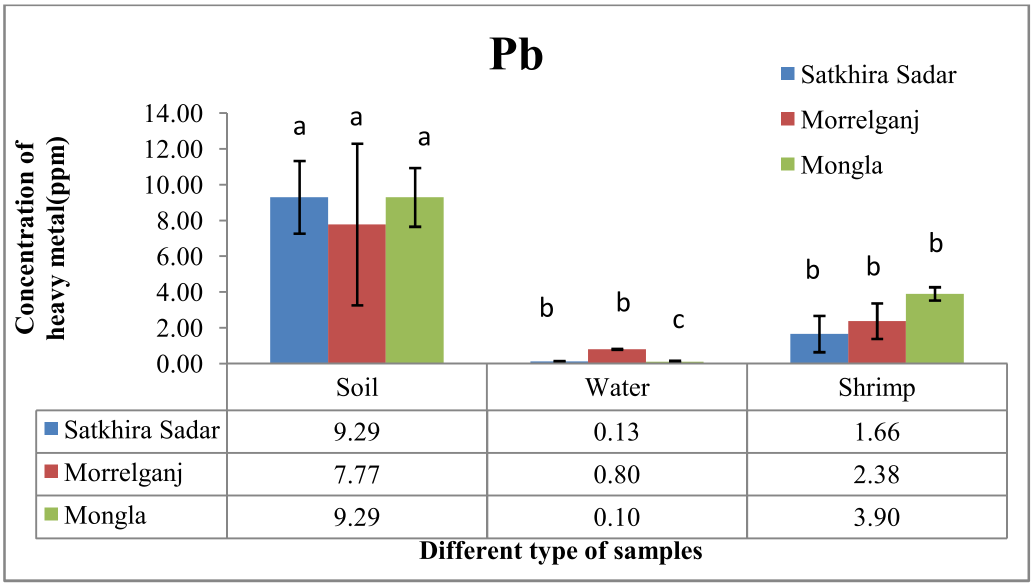



**Fig.3.9. Mean (± SD) concentration of the Cadmium (Cd) in soil, water and Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

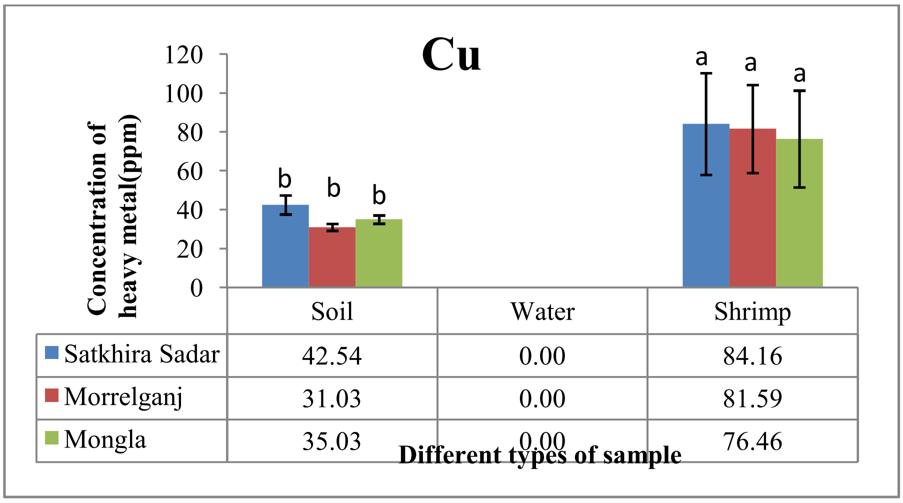
Fig.3.10 shows that the concentration of Lead in soil was found highest in Satkhira Sadar and Mongla (9.29 ppm) and lowest in Morrelganj (7.77 ppm). In water, highest concentration was found in Morrelganj (0.80 ppm) and lowest concentration found in Mongla (0.10 ppm). In Shrimp, highest concentration was found in Mongla (3.90 ppm) and lowest concentration found in Satkhira Sadar (1.66 ppm). There were significant differences found among Soil, Water and Shrimp within region ($p< 0.05$).




| | Soil | Water | Shrimp |
|---|---|---|---|
| Satkhira Sadar | 9.29 | 0.13 | 1.66 |
| Morrelganj | 7.77 | 0.80 | 2.38 |
| Mongla | 9.29 | 0.10 | 3.90 |

**Fig.3.10. Mean (± SD) concentration of the Lead (Pb) in soil, water and Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

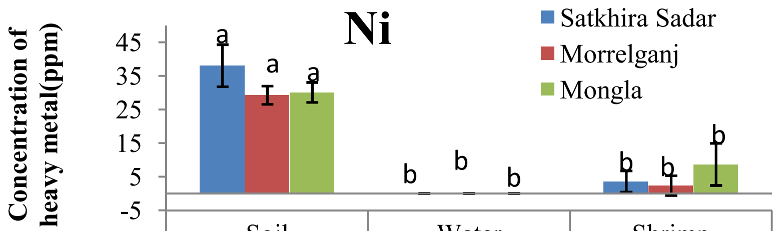
Fig.3.11 shows that the concentration of Copper in soil was found highest in Satkhira Sadar (42.54 ppm) and lowest in Morrelganj (31.03 ppm). In water, the concentration of Copper (Cu) was Below Detectable Level (BDL) in various regions. In Shrimp, highest concentration was found in Satkhira Sadar (84.16 ppm) and lowest concentration found in Mongla (76.46 ppm). There were significant differences found among Soil, Water and Shrimp within region ($p< 0.05$).




| | Soil | Water | Shrimp |
|---|---|---|---|
| Satkhira Sadar | 42.54 | 0.00 | 84.16 |
| Morrelganj | 31.03 | 0.00 | 81.59 |
| Mongla | 35.03 | 0.00 | 76.46 |

**Fig.3.11. Mean (± SD) concentration of the Copper (Cu) in soil, water and Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

Fig.3.12 shows that the concentration of Nickel in soil was found highest in Satkhira Sadar (38.06 ppm) and lowest in Morrelganj (29.27 ppm). In water, highest concentration was found in Morrelganj (0.0181 ppm) and lowest concentration found in Satkhira Sadar (0.0083 ppm). In Shrimp, highest concentration was found in Mongla (8.64 ppm) and lowest concentration found in Morrelganj (2.33 ppm). There were significant differences found among Soil, Water and Shrimp within region ($p < 0.05$).




| | Soil | Water | Shrimp |
|---|---|---|---|
| Satkhira Sadar | 38.06 | 0.01 | 3.60 |
| Morrelganj | 29.27 | 0.02 | 2.33 |
| Mongla | 30.10 | 0.01 | 8.64 |

**Different types of sample**

**Fig.3.12. Mean (± SD) concentration of the Nickel (Ni) in soil, water and Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

Fig.3.13 shows that the concentration of Chromium in soil was found highest in Satkhira Sadar (45.45 ppm) and lowest in Morrelganj (36.80 ppm). In water and Shrimp, the concentration of Heavy Metal was Below Detectable Level (BDL). There were significant differences found among Soil, Water and Shrimp within region ($p < 0.05$).

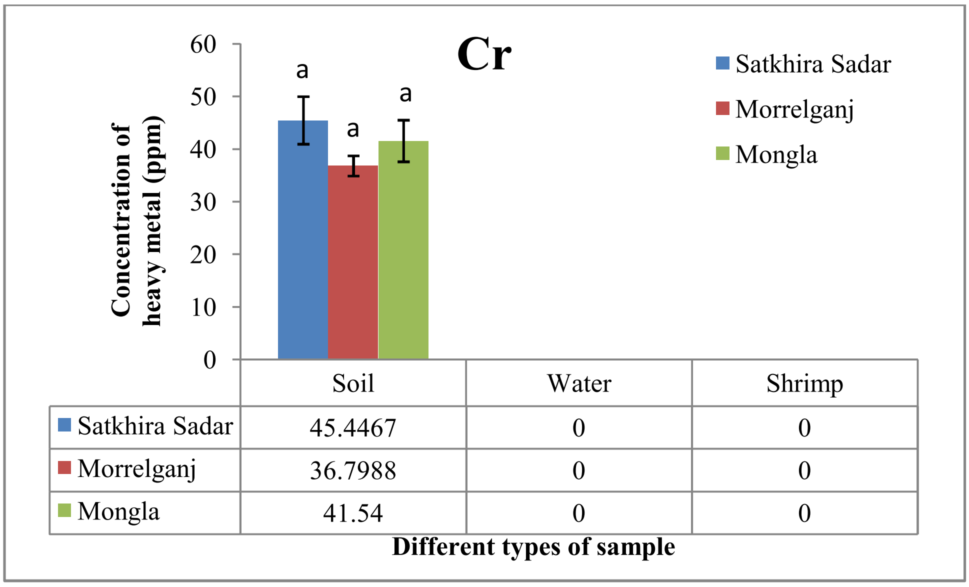
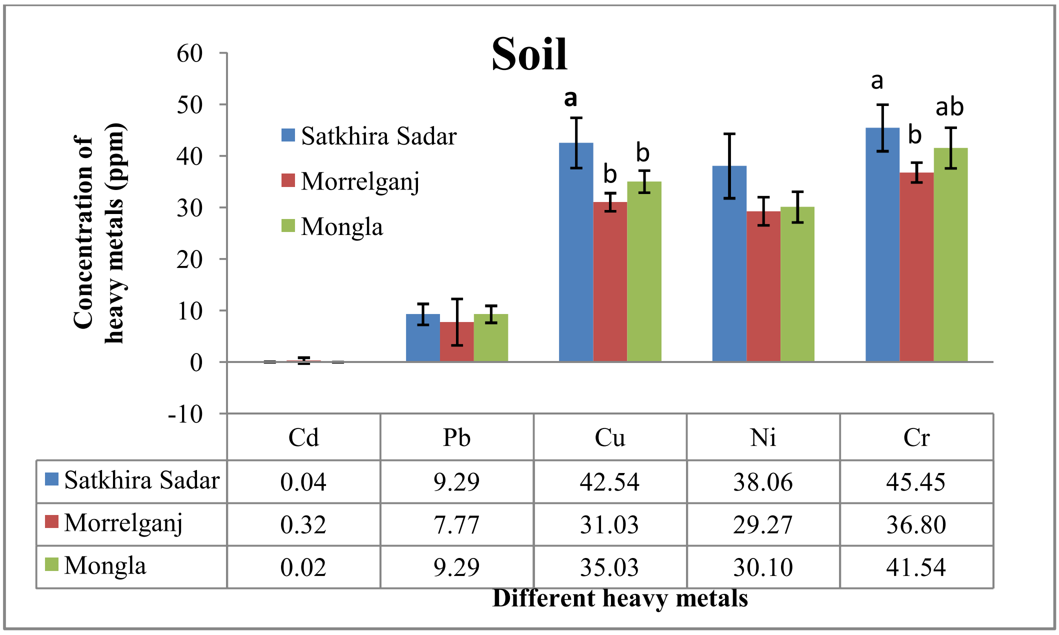



**Fig.3.13. Mean (± SD) concentration of the chromium (Cr) in soil, water and Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

### 3.1.4 Determination of heavy metal concentration in collected Soil, Water and Shrimp

Fig.3.15 shows that the concentration of Cadmium in soil was found highest in Morrelganj region (0.32 ppm) and lowest in Mongla region (0.02 ppm). Lead, highest concentration was found in Mongla (9.293 ppm) and lowest concentration found in Morrelganj region (7.77 ppm).Copper, highest concentration was found in Satkhira Sadar (38.06 ppm) and lowest concentration found in Morrelganj region (29.27 ppm).Nickel, highest concentration was found in Satkhira Sadar (38.06 ppm) and lowest concentration found in Morrelganj region (29.27 ppm).Chromium, highest concentration was found in Satkhira Sadar (45.45 ppm) and lowest concentration found in Morrelganj region (36.08 ppm). There were significant differences found among Soil, Water and Shrimp within region ($p< 0.05$).




| | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|
| Satkhira Sadar | 0.04 | 9.29 | 42.54 | 38.06 | 45.45 |
| Morrelganj | 0.32 | 7.77 | 31.03 | 29.27 | 36.80 |
| Mongla | 0.02 | 9.29 | 35.03 | 30.10 | 41.54 |

**Fig.3.14. Mean (± SD) concentration of the heavy metals in Soil collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different superscripts are significantly different within group (ANOVA, $p<0.05$).**

Fig.3.15 shows that in Water the concentration of Cadmium was Below Detectable Level (BDL). Lead, highest concentration was found in Satkhira Sadar (0.13 ppm) and lowest concentration found in Morrelganj region (0.08 ppm). The concentration of Copper was Below Determination Level (BDL).Nickel, highest concentration was found in Morrelganj Region (0.02 ppm) and lowest concentration found in Mongla region (0.012 ppm).The concentration of Chromium was Below Detectable Level (BDL). There were significant differences found among Soil, Water and Shrimp within region ($p< 0.05$).

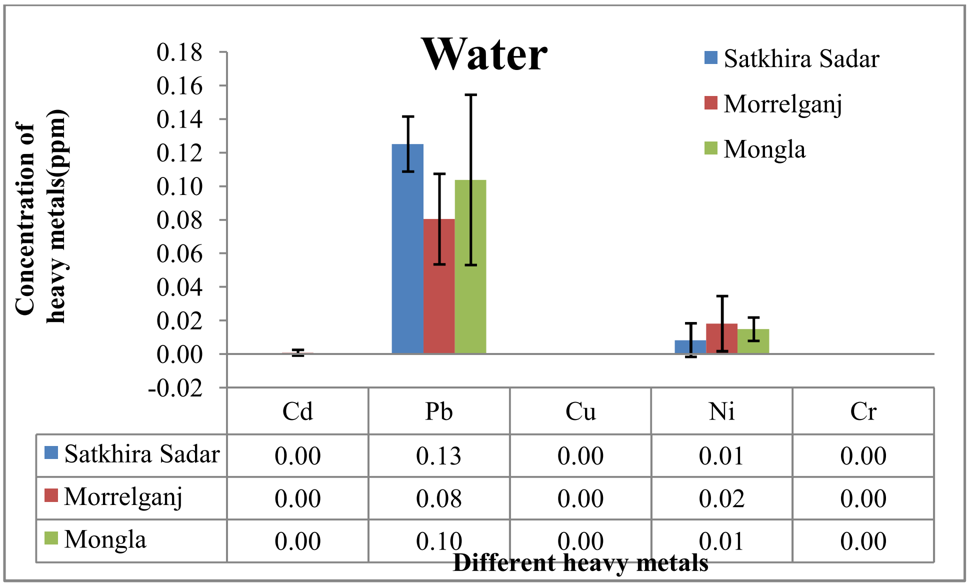



**Fig.3.15. Mean (± SD) concentration of the heavy metals in Water collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different superscripts are significantly different within group (ANOVA, $p<0.05$).**

Fig.3.16 shows that the concentration of Cadmium in Shrimp was found highest in Morrelganj region (0.35 ppm) and lowest in Satkhira Sadar region (0.26 ppm). Lead, highest concentration was found in Mongla (3.90 ppm) and lowest concentration found in Satkhira Sadar region (1.66 ppm).Copper, highest concentration was found in Satkhira Sadar (84.16 ppm) and lowest concentration found in Mongla region (76.46 ppm).Nickel, highest concentration was found in Mongla region (8.64 ppm) and lowest concentration found in Morrelganj region (2.33 ppm).The concentration of Chromium was Below Detectable Level (BDL). There were significant differences found among Soil, Water and Shrimp within region ($p< 0.05$).

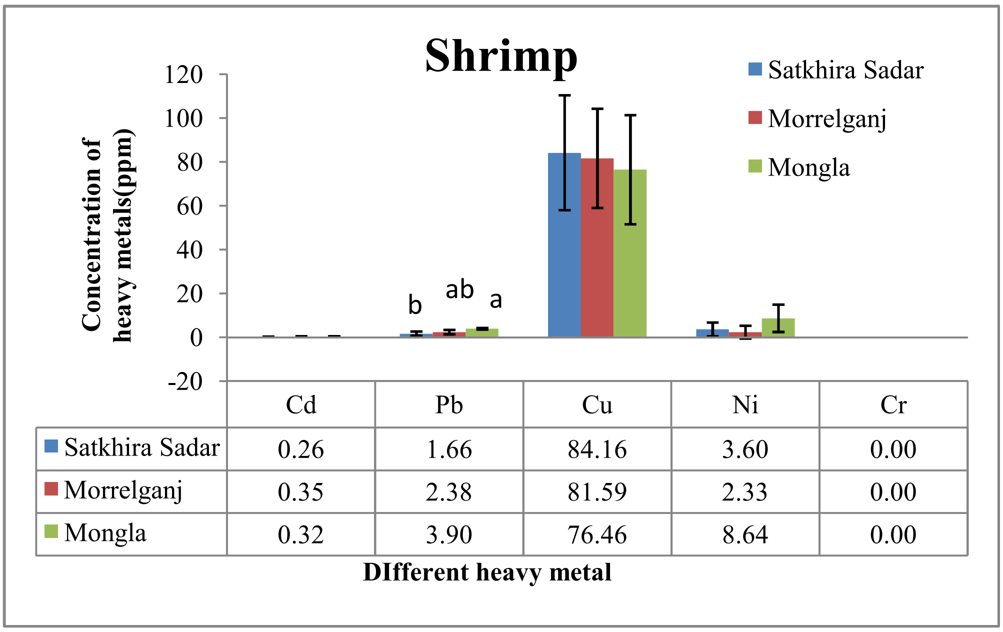



**Fig.3.16. Mean (± SD) concentration of the heavy metals in Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different superscripts are significantly different within group (ANOVA, $p<0.05$).**

### 3.2 Post Monsoon

Post monsoon season is dry season. Rainfall is too less. Water of gherare less than other time being. In post monsoon (November- December) only Shrimp sample was collected from the sampling area.

#### 3.2.1 Determination of heavy metals in collected Shrimp organs

Fig.3.17 shows that the concentration of Cadmium in shell was found highest in Morrelganj region (0.17 ppm) and lowest in Satkhira Sadar BDL (Below Determination Level). In head, Cd was found below determination level (BDL). In Muscle, highest concentration was found in Mongla (0.39 ppm) and lowest concentration found in Mongla region (0.28 ppm). There were significant differences found among Shell, Head and Muscle within region ($p< 0.05$).

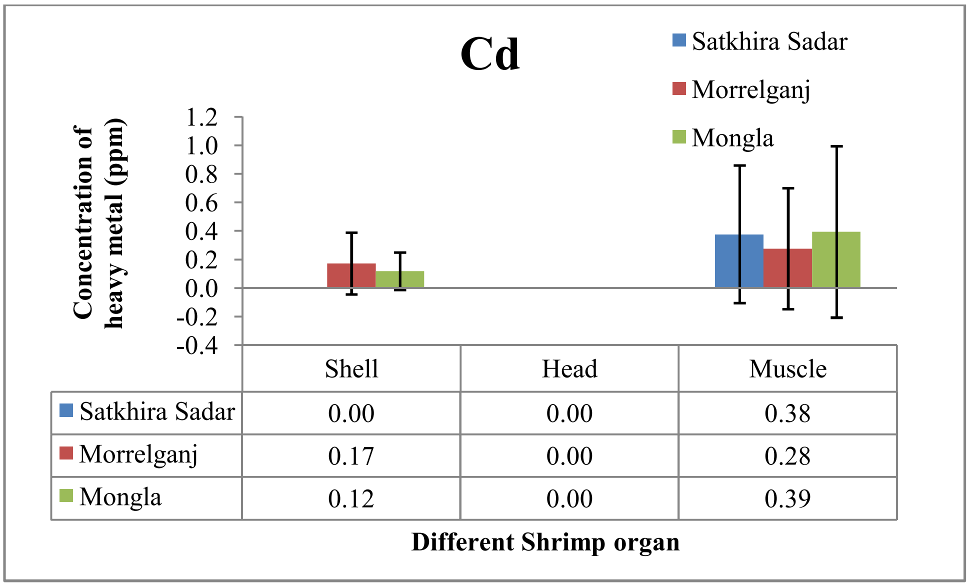



**Fig.3.17. Mean (± SD) concentration of the Cadmium (Cd) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, $p<0.05$).**

Fig.3.18 shows that the concentration of Lead in shell was found highest in Mongla region (2.22 ppm) and lowest in Satkhira Sadar region (Below Determination Level). In head, highest concentration was found in Mongla (9.18 ppm) and lowest concentration found in Satkhira Sadar region (6.45 ppm). In Muscle, highest concentration was found in Mongla (2.67 ppm) and lowest concentration found in Satkhira Sadar region (1.56 ppm). There were significant differences found among Shell, Head and Muscle within region ($p< 0.05$).

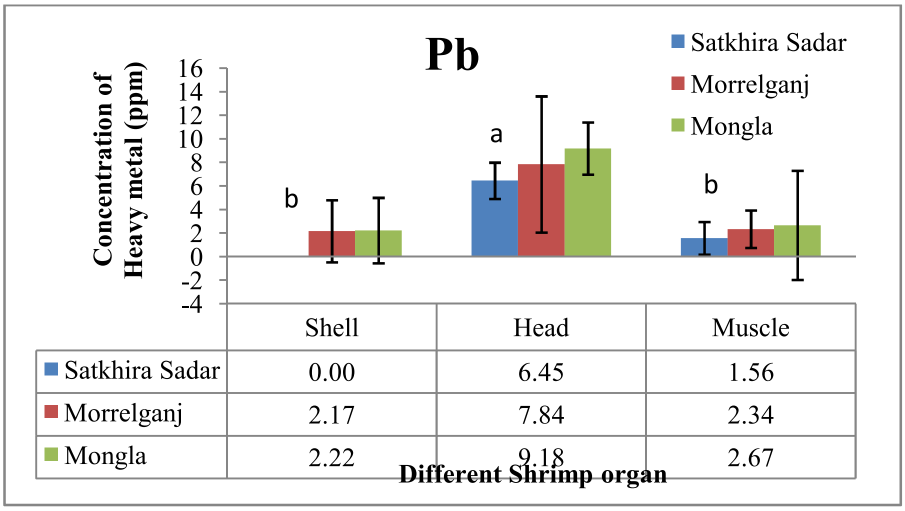



**Fig.3.18. Mean (± SD) concentration of the Lead (Pb) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with**

**different colors with different letters are significantly different within regions (ANOVA, p<0.05).**

Fig.3.19 shows that the concentration of Copper in shell was found highest in Satkhira Sadar (143.96 ppm) and lowest in Mongla (108.18 ppm). In head, highest concentration was found in Satkhira Sadar (32.88 ppm) and lowest concentration found in Mongla (30.50 ppm). In Muscle, highest concentration was found in Satkhira Sadar (29.95 ppm) and lowest concentration found in Mongla region (19.94 ppm). There were significant differences found among Shell, Head and Muscle within region (p< 0.05).

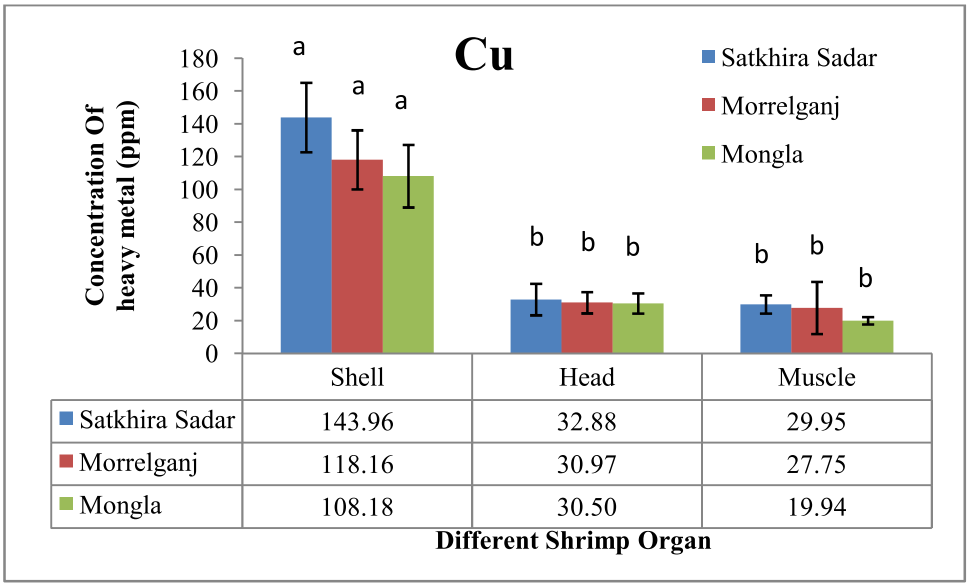



**Fig.3.19. Mean (± SD) concentration of the Copper (Cu) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, p<0.05).**

Fig.3.20 shows that the concentration of Nickel in shell was found highest in Satkhira Sadar region (2.92 ppm) and lowest in Morrelganj region (2.38 ppm). In head, highest concentration was found in Mongla (5.51 ppm) and lowest concentration found in Satkhira Sadar region (1.89 ppm). In Muscle, highest concentration was found in Morrelganj (1.01 ppm) and lowest concentration found in Mongla region (0.62 ppm). There were significant differences found among Shell, Head and Muscle within region (p< 0.05).

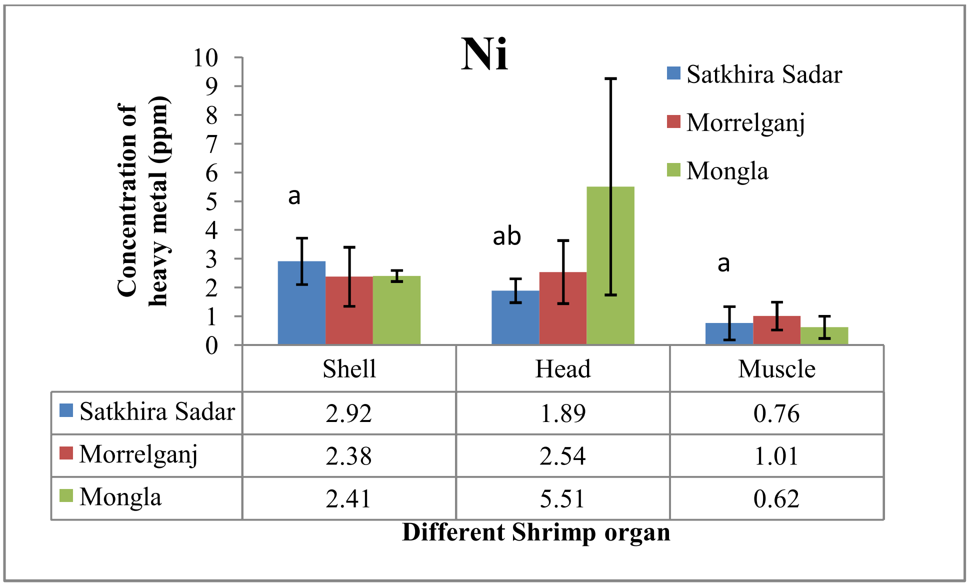



**Fig.3.20. Mean (± SD) concentration of the Nickel (Ni) in shell, head and muscle of Shrimp collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with different colors with different letters are significantly different within regions (ANOVA, $p<0.05$).**

The concentration of Chromium (Cr) was below detectable Level (BDL) in Shell, Head and muscle of Shrimp of various regions.

### 3.1.2 Overall concentration of heavy metal in Shrimp of various regions

Fig.3.21 shows that the mean concentration (shell, head and muscle) of Cadmium in Shrimp was found highest in Mongla region (0.17 ppm) and lowest in Satkhira Sadar region (0.13 ppm). There was no significant difference found among regions ($p< 0.05$).

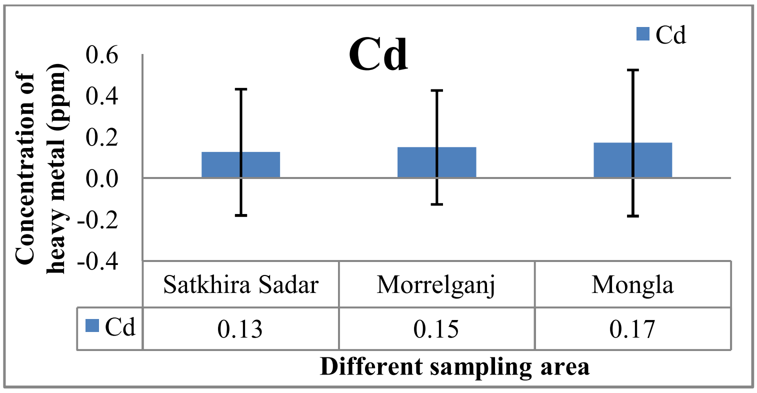



**Fig.3.21: Mean (± SD) concentration of the Cadmium (Cd) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, $p<0.05$).**

Fig.3.22 shows that the mean concentration (shell, head and muscle) of Lead in Shrimp was found highest in Mongla region (4.69 ppm) and lowest in Satkhira Sadar region (2.67 ppm). There was no significant difference among regions ($p < 0.05$).

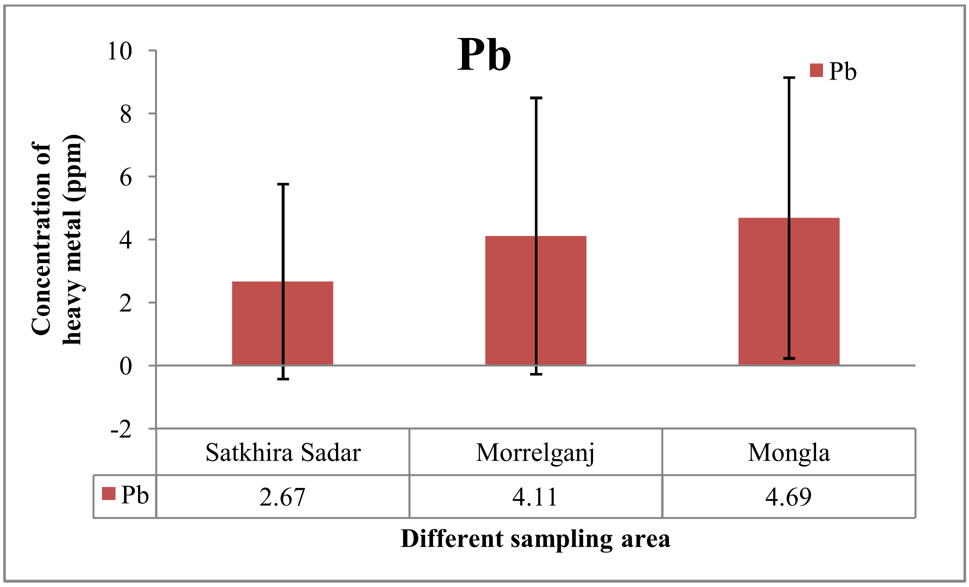



**Fig.3.22. Mean (± SD) concentration of the Lead (Pb) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, $p<0.05$).**

Fig.3.23 shows that the mean concentration (shell, head and muscle) of Copper in Shrimp was found highest in Mongla region (2.84 ppm) and lowest in Satkhira Sadar region (1.86 ppm). There was no significant difference found among regions ($p < 0.05$).

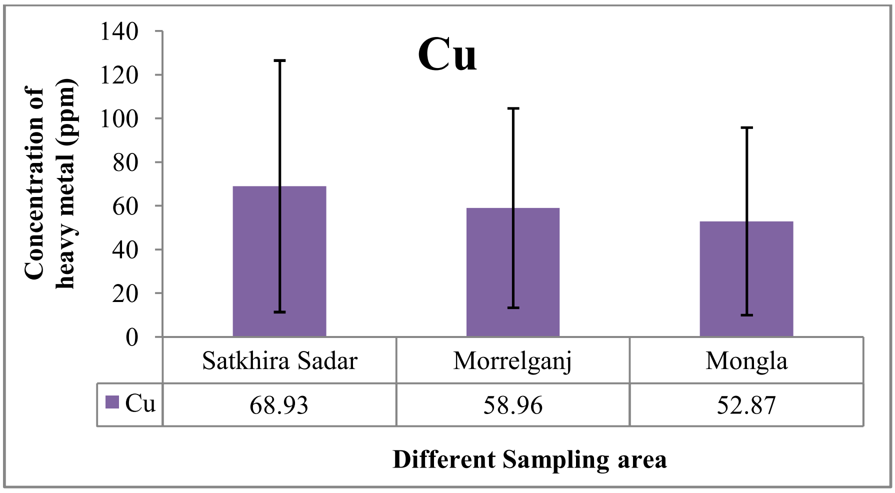



**Fig.3.23. Mean (± SD) concentration of the copper (Cu) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, $p<0.05$).**

Fig.3.24 shows that the mean concentration (shell, head and muscle) of Nickel in Shrimp was found highest in Mongla region (2.84 ppm) and lowest in Satkhira Sadar region (1.86 ppm). There was no significant difference found among regions ($p < 0.05$).

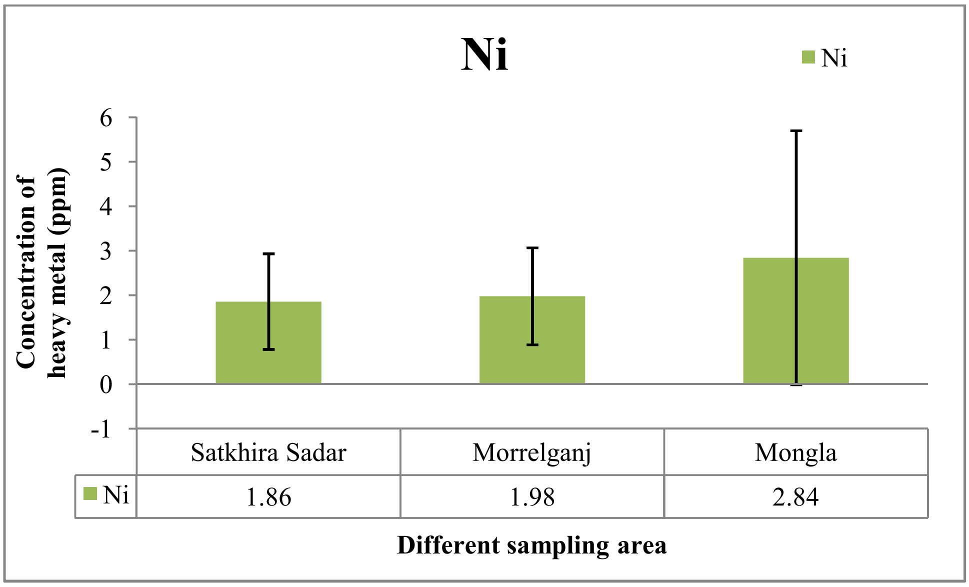
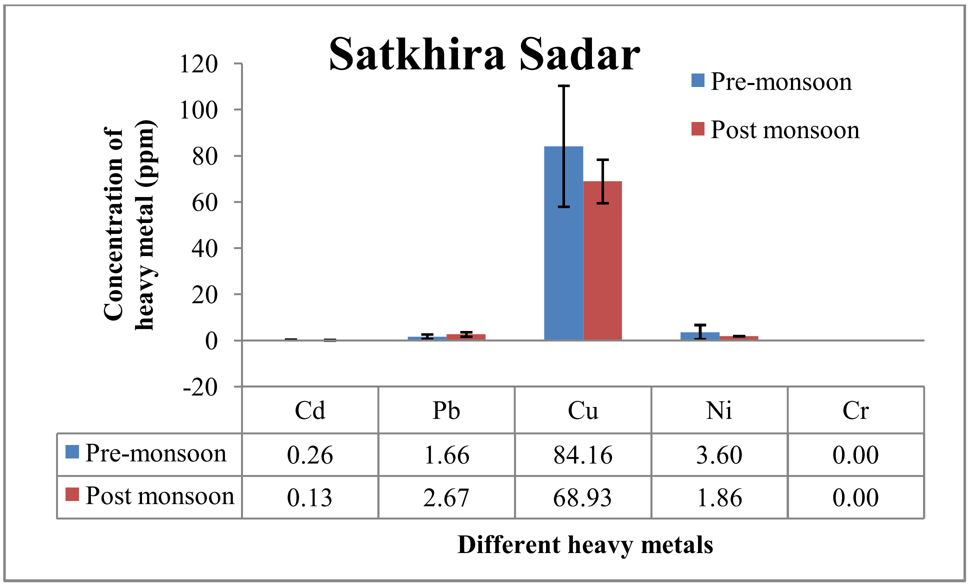



**Fig.3.24. Mean (± SD) concentration of the Nickel (Ni) in Shrimp (mean of shell, head and muscle) collected from ghers of Satkhira Sadar, Morrelganj and Mongla. Bars with no letter denotes no significant difference within regions (ANOVA, p<0.05).**

### 3.3 Comparison of Shrimp samples of pre-monsoon and post monsoon

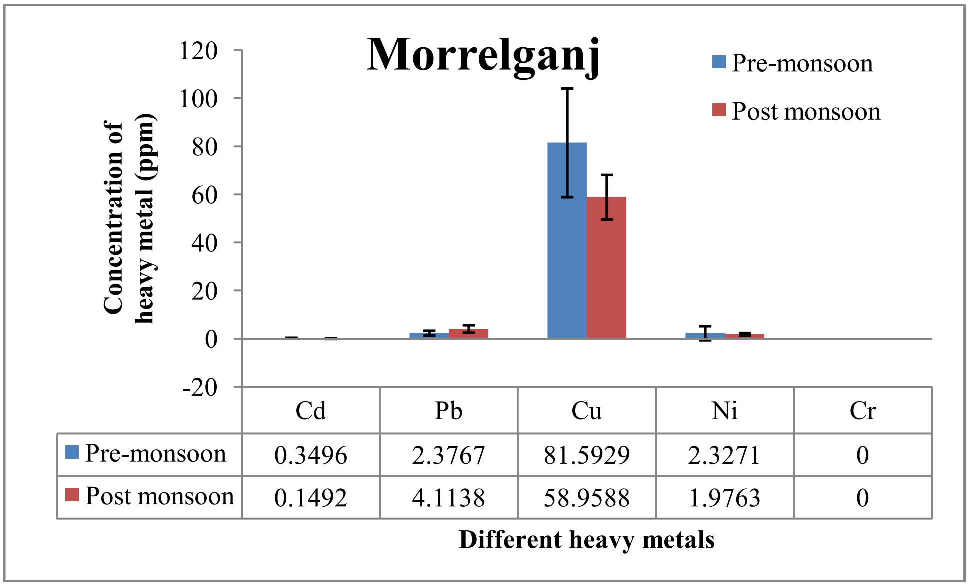
Fig.3.25 shows that the concentration of Cd, Cu and Ni in pre-monsoon were greater than post monsoon concentration and but concentration of Pb in post monsoon was greater than pre-monsoon in Satkhira Sadar. But the concentration of Chromium in both season found below detectable limit.




| | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|
| Pre-monsoon | 0.26 | 1.66 | 84.16 | 3.60 | 0.00 |
| Post monsoon | 0.13 | 2.67 | 68.93 | 1.86 | 0.00 |

**Fig.3.25. Mean (± SD) concentration of different heavy metals in Shrimp collected from ghers of Satkhira Sadar in pre-monsoon and post monsoon. Bars with no letter denotes no significant difference between pre-monsoon and post monsoon (T-test, $p<0.05$).**

Fig.3.26 shows that the concentration of Cd, Cu and Ni in pre-monsoon were greater than post monsoon concentration and but concentration of Pb in post monsoon was greater than pre-monsoon in Morrelganj. But the concentration of Chromium in both season found below detectable limit.




| | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|
| Pre-monsoon | 0.3496 | 2.3767 | 81.5929 | 2.3271 | 0 |
| Post monsoon | 0.1492 | 4.1138 | 58.9588 | 1.9763 | 0 |

**Fig.3.26. Mean (± SD) concentration of different heavy metals in Shrimp collected from ghers of Morrelganj in pre-monsoon and post monsoon. Bars with no letter denotes no significant difference between pre-monsoon and post monsoon (T-test, $p<0.05$).**

Fig.3.27 shows that the concentration of Cd, Cu and Ni in pre-monsoon were greater than post monsoon concentration and but concentration of Pb in post monsoon was greater than pre-monsoon in Morrelganj. But the concentration of Chromium in both season found below detectable limit.

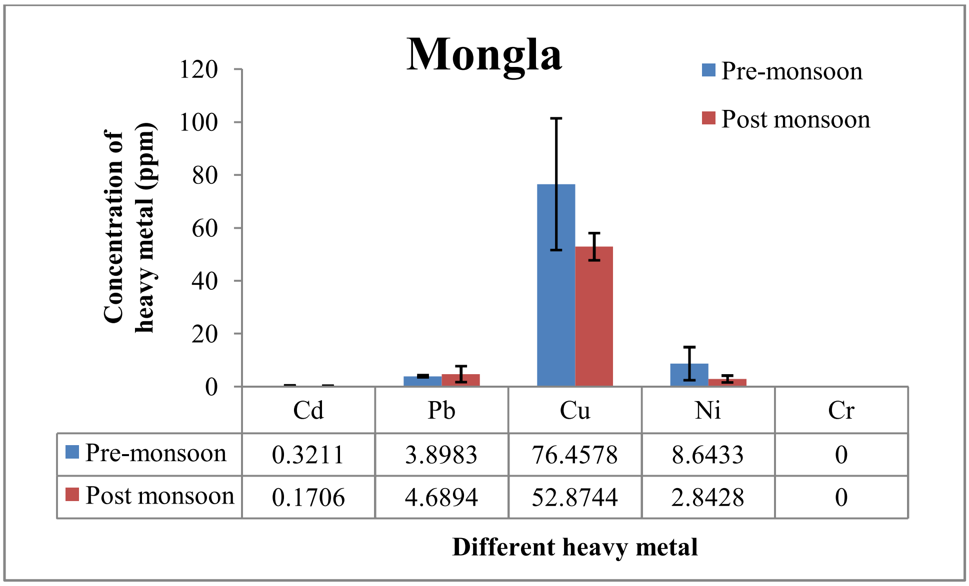



**Fig.3.27. Mean (± SD) concentration of different heavy metals in Shrimp collected from ghers of Mongla in pre-monsoon and post monsoon. Bars with no letter denotes no significant difference between pre-monsoon and post monsoon (T-test, $p<0.05$).**

### 3.4 Human health risk

To assess human health risk estimated daily intake (EDI), target hazard quotients (THQ), carcinogenic risk (CR) are calculated below.

#### 3.4.1 Estimated daily intake (EDI)

In pre-monsoon the EDI of each metal through consumption of fish followed in the descending order of Cu> Ni >Pb>Cb> Cr and in post monsoon each metal through consumption of fish followed in the descending order of Cu> Ni >Pb>Cb> Cr. Table 3.1 and 3.2 show estimated dietary intake (EDI) (mg/day) of heavy metals due to consumption of Shrimps of the present study.

**Table 3.1 Estimated dietary intake (EDI) (mg/day) of heavy metal from pre-monsoon concentration of Shrimp**

| Heavy Metals | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Cd | 0.19465 | 0.266861 | 0.245106 | 0.254575 | 0.349017 | 0.320565 | 0.210375 | 0.28842 | 0.264908 |
| Pb | 1.265454 | 1.814214 | 2.975702 | 1.655037 | 2.372739 | 3.891803 | 1.367685 | 1.960778 | 3.216098 |
| Cu | 64.23832 | 62.28258 | 58.36279 | 84.01474 | 81.45691 | 76.33037 | 69.42788 | 67.31414 | 63.07769 |
| Ni | 2.745481 | 1.776353 | 6.597719 | 3.590706 | 2.323222 | 8.628895 | 2.967278 | 1.919858 | 7.130723 |
| Cr | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

**Table 3.2 Estimated dietary intake (EDI) (mg/day) of heavy metal from post monsoon concentration of Shrimp**

| Heavy Metals | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Cd | 0.095875 | 0.113889 | 0.130225 | 0.125391 | 0.148951 | 0.170316 | 0.10362 | 0.12309 | 0.140745 |
| Pb | 2.037642 | 3.140201 | 3.579575 | 2.664951 | 4.106944 | 4.681584 | 2.202255 | 3.393885 | 3.868755 |
| Cu | 52.61527 | 45.00514 | 40.36079 | 68.81342 | 58.86044 | 52.78628 | 56.86585 | 48.64093 | 43.62138 |
| Ni | 1.417663 | 1.508499 | 2.170004 | 1.854105 | 1.972906 | 2.838062 | 1.53219 | 1.630365 | 2.34531 |
| Cr | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

### 3.4.2 Target Hazard Quotients (THQ)

In pre-monsoon and post monsoon the THQ of each metal through consumption of fish followed in the descending order of Cu> Cd>Pb>Ni> Cr but for urban region Pb was greater than 1 that was denoted as hazard risk in pre-monsoon and for rural, urban and total region pb was greater than1 was denoted as hazard risk in post monsoon. Table 3.3 and 3.4 shows target hazard quotient (THQ) of heavy metals due to consumption of Shrimps of the present study.

**Table 3.3 Target Hazard Quotients (THQ) estimated from pre-monsoon concentration of Shrimp**

| Heavy Metals | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Cd | 0.3893 | 0.533723 | 0.490213 | 0.50915 | 0.698035 | 0.64113 | 0.42075 | 0.57684 | 0.529815 |
| Pb | 0.361558 | 0.518347 | 0.850201 | 0.472868 | 0.677925 | **1.111944** | 0.390767 | 0.560222 | 0.918885 |
| Cu | **1.605958** | **1.557065** | **1.45907** | **2.100369** | **2.036423** | **1.908259** | **1.735697** | **1.682854** | **1.576942** |
| Ni | 0.137274 | 0.088818 | 0.329886 | 0.179535 | 0.116161 | 0.431445 | 0.148364 | 0.095993 | 0.356536 |
| Cr | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Note: Bold indicate THQ > 1

**Table 3.4 Target Hazard Quotients (THQ) estimated from post monsoon concentration of Shrimp**

| Heavy Metals | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Cd | 0.191749 | 0.227779 | 0.260449 | 0.250781 | 0.297903 | 0.340631 | 0.20724 | 0.24618 | 0.28149 |
| Pb | 0.582183 | 0.8972 | **1.022736** | 0.761415 | **1.173412** | **1.337596** | 0.629216 | 0.969681 | **1.105359** |
| Cu | **1.315382** | **1.125129** | **1.00902** | **1.720335** | **1.471511** | **1.319657** | **1.421646** | **1.216023** | **1.090535** |
| Ni | 0.070883 | 0.075425 | 0.1085 | 0.092705 | 0.098645 | 0.141903 | 0.07661 | 0.081518 | 0.117266 |
| Cr | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Note: Bold indicate THQ > 1

### 3.4.3 Carcinogenic Risk (CR)

In pre-monsoon and post monsoon season all the value were between $10^{-4}$- $10^{-6}$ , that is in acceptable limit but still in hazard risk. Table 3.5 and 3.6 shows target carcinogenic risk (CR) of heavy metals due to consumption of Shrimp of the present study.

**Table 3.5**Carcinogenic risk (CR) estimated from the pre-monsoon concentration of Shrimp

| Heavy metal | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Pb | 1.07564E-05 | 1.54208E-05 | 2.52935E-05 | 1.40678E-05 | 2.01683E-05 | 3.30803E-05 | 1.16253E-05 | 1.66666E-05 | 2.73368E-05 |

**Table 3.6 Carcinogenic risk (CR) estimated from the post monsoon concentration of Shrimp**

| Heavy metal | Rural | | | Urban | | | Total | | |
|---|---|---|---|---|---|---|---|---|---|
| | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla | Satkhira Sadar | Morrelganj | Mongla |
| Pb | 1.732E-05 | 2.66917E-05 | 3.04264E-05 | 2.26521E-05 | 3.4909E-05 | 3.97935E-05 | 1.87192E-05 | 2.8848E-05 | 3.28844E-05 |

# Chapter 4
# Discussion

## 4.1 Heavy metals in Shrimp

The concentration of heavy metalswas different in various parts of the Shrimp. In shell, head and muscle concentration of heavy metals were also different in various regions.

In the present study in pre-monsoon we found cadmium in shell varying from 0.1-0.2 ppm, in head 0.52-0.72 ppm and in muscle 0.12-0.16 ppm. The mean concentration of Shrimp found in Satkhira Sadar region was 0.26 ppm, in Morrelganj 0.35 ppm, in Mongla 0.32 ppm. But in post monsoon we found Cd in shell 0.17-0.12 ppm, in head below detectable limit and in muscle 0.39-0.28 ppm. The mean concentration of Shrimp was found All are exceeded the maximum allowable concentration (MAC) according to FAO/WHO(2002). Among them there was a significant difference among shell, head and muscle within region ($p < 0.005$). The mean concentration of shell, head and muscle of Cadmium was found in order of Morrelganj > Mongla > Satkhira Sadar. Ahmed et al. (2015) found the mean concentration of Cd low in the shellfish (0.05±0.00 mg/kg) and fishes (0.01±0.00 to 0.02±0.00 mg/kg) but high in crustaceans (1.51±0.04 mg/kg). Sirelil et al. (2006) found Cd in vacuum packaged smoked fish species (mackerel, *Salmosalar*and *Oncorhynchusmykiss*), which varied from 0.003 to 0.036 mg/kg with a mean of 0.01367 mg/kg. They found Cd in high range due to industrial processes such as smelting or electroplating as well as excess use of fertilizer. But in our study we found Cd high because in the sampling area (Satkhira Sadar, Morrelganj and Mongla) was full of industries and they discharge their wastage in the river especially in Posur river (Mongla).

In pre-monsoon we found Lead in the shell of Shrimp in Satkhira Sadar 3.90 ppm, in Morrelganj 5.22 ppm and highest concentration in Mongla 8.19 ppm. We found 0.54-3.22 ppm in the head of Shrimp. But in muscle the range was 0.29-1.37 ppm and in Satkhira Sadar it was below detectable limit. Themean concentration of Pb in Satkhira Sadar Shrimpwas 1.66 ppm, in Morrelganj 2.38 ppm and in Mongla is 3.90 ppm. In post monsoon we found Pb at 2.17 ppm in Morrelganj, 2.22 ppm in Mongla but in Satkhira Sadar it was below detectable limit in shell. In head we found high concentration of Lead. The range was 6.45-9.18 ppm and in muscle we found 1.56 ppm in Satkhira Sadar 2.34 ppm in Morrelganj and 2.67 in Mongla. Only in Satkhira Sadar there was significant difference among shell, head and muscle. All values exceeded the maximum allowable concentration (MAC) according to FAO/WHO (2002) except in muscle in Satkhira Sadar.Ahmed at el (2015) found the mean value of Pb in Buriganga river were 0.47±0.03 mg/kg in A. coila, 0.54±0.01 mg/kg in *G. youssoufi*, 3.17±0.07 mg/kg in *M. pancalus*, 0.51±0.01 mg/kg in *M. rosenbergii* and 4.55± 0.11 mg/kg in *Indoplanorbisexustus*. In the present study it shows that in shell Pb varies from 3.90-8.19 ppm, in head 0.54-3.22 ppm and in muscle 0-1.37 ppm. There are many cement industries in Mongla region and the wastage of the mill is discharged in the Posurriver. They found high amount of Pb due to accumulation of heavy metal from the water of the river which was polluted by industrial wastage. Satkhira Sadar and Morrelganj hasalso some small industry. The farmers directly use the water of the river. So there is a huge possibility of accumulation of heavy metal from the water.Pb is a ubiquitous pollutant which could find its way into the Posur River through discharge of industrial effluents from various industries such as printing, dyeing, oil refineries, textile around port and other sources.

The present study showed that the concentration of Copper in pre-monsoon was highest in head portion (Satkhira Sadar 168.05 ppm, Morrelganj 166.63 ppm and Mongla 155.86 ppm). In shell the amount of Cu was 53.94 ppm in Satkhira Sadar, 46.74 ppm in Morrelganj region and 48.65 ppm in Mongla region. The mean concentration of cu in Satkhira Sadar was 84.16 ppm, in Morrelganj 81.59 ppm and in Mongla 76.46 ppm. In post monsoon, highest concentration in shell, head and muscle was found in Satkhira Sadar respectively 143.96 ppm, 32.88 ppm and 29.95 ppm and lowest concentration found in Mongla respectively 108.08 ppm, 30.50 ppm and 19.94 ppm.The mean concentration of Shrimp was found in Satkhira Sadar 68.93 ppm, Morrelganj 58.96 ppm and Mongla 52.87 ppm. The present data extremely exceeded the Maximum Allowable concentration (MAC) according to FAO/WHO(2002).Cu concentration in prawn sample was extremely higher compared to the concentration of the other heavy metals that were analyzed in the fish, prawn, and shellfish samples. Islam et al. 2014 found that in fish the highest concentration of copper 0.07–2.5 mg/kg and was lower than that of fish muscle collected from the Bangshi River, Bangladesh.Other studies have shown that Cu is highly toxic in aquatic environments and has effects on fish, invertebrates, and amphibians, with all three groups equally sensitive to chronic toxicity. Copper can accumulate in different organs in fish and mollusks (Kamaruzzam et al., 2010). In soil we found higher amount of copper and it accumulated in Shrimp body.

The present study shows that in pre-monsoon in shell the highest concentration of Nickel is in Satkhira Sadar 3.10 ppm but in Mongla it is below determination level, in head highest concentration shows 0.51 ppm but in Morrelganj and Mongla concentration of Nickel is below determination level, in muscle highest concentration found in Mongla 25.93 ppm and in Satkhira 7.18 ppm as well as in Morrelganj 6.74 ppm. The mean concentration of Nickel in Shrimp is 8.64 ppm in Mongla, 3.60 ppm in Satkhira Sadar and lowest in Morrelganj 2.33 ppm. In post monsoon we found Ni in Satkhira Sadar shell 2.92 ppm, head 1.89 ppm and in muscle 0.76 ppm and mean concentration was 1.86 ppm. In Morrelganj the concentration was 2.38 ppm in shell, 2.54 ppm in head and 1.01 ppm in muscle and mean concentration was 1.98 ppm. The highest concentration was found in Mongla, in shell we found 2.41 ppm in head 5.41 ppm and in muscle 0.62 ppm and the mean concentration was 2.84 ppm. All values exceeded the maximum allowable concentration (MAC) according to FAO/WHO (2002). Due to pollution in water and huge amount of wastage in river, the mean concentrations of Ni in fish samples ranged from 0.5 mg/kg (*C. soborna*) to1.1 mg/kg (*N. notopterus*) reported value of Islam et al (2014). Another study shows that the highest concentration, 5.75± 2.40 mg/kg, was measured in shellfish with the lowest detectable concentration, 0.36±0.10 mg/kg, measured in *A. coila*(Ahmed at el., 2015).

There is a huge amount of nickel whichwas accumulated in soil like in Satkhira Sadar 38.06 ppm, in Mongla 29.27 ppm and in Mongla 30.10 ppm. Shrimp is bottom feeder so the accumulation of nickel in Shrimp is very high which extremely exceeded the Maximum Allowable concentration (MAC) according to FAO/WHO(2002).

In pre-monsoon and post monsoon, we found Chromium below detectable limit in Shrimp in all region. But in soil we found high concentration of Cr in every sampling area. Ahmed at el. (2015) found that Cr in the fish, crustacean, and shellfish samples were in the range of 1.59±0.93 to 16.05±1.48 mg/kg. The lowest concentration, 1.59±0.93 mg/kg,was measured in crustaceans while the highest concentration,16.05±1.48 mg/kg, was measured in shellfish (gastropod Mollusca). These results were found due to denseCr pollution. In present study we found Cr in soil but it did not accumulate in Shrimp.

### 4.2 Heavy metal in Soil

In the present study in soil we found heavy metals in Satkhira Sadar Cd 0.04 ppm, Pb 9.29 ppm, Cu 42.54 ppm, Ni 38.06 ppm and Cr 45.45 ppm; In Morrelganj Cd 0.32 ppm, Pb 7.77 ppm, Cu 31.03 ppm, Ni 29.27 ppm and Cr 36.80 ppm; in Mongla Cd 0.02 ppm, Pb 9.29 ppm, Cu 35.03 ppm, Ni 30.10 ppm and Cr 41.54 ppm. The average concentration of heavy metals in every region were in same decreasing order like Cr > Cu > Ni >Pb> Cd. Islam et al. 2016 found heavy metal in sediment of Korotoa river in the following decreasing order Cr > Ni > Cu >Pb> As > Cd. They found those heavy metals due to urban activities like industrial discharges, municipal waste water, household garbage and urban runoff. We found almost same line in decreasing order due to same reason like industrial pollution, household garbage in the river which is used in the Gher of Shrimp.

**4.3 Heavy metal in Water**

In the present study we found Cadmium, Copper and Chromium below detectable limit in every region. But we found Lead 0.13 ppm in Satkhira Sadar, 0.08 in Morrelganj and 0.10 ppm in Mongla. Nickel was found in very low concentration, 0.01 in Satkhira Sadar, 0.02 in Morrelganj and 0.01 in Mongla. Due to higher use of fertilizer and pesticide and the cumulative impact of numerous industries (printing, dyeing, leather electroplating). Saha et al.(2016) found the mean metal concentrations (mg/L) in ground and surface water, respectively,followed in a decreasing order as: Zn (127.63) > Cu (78.60) >Mn(11.21) > Ni (7.93) >Pb (5.21) > Cr (4.43) > As (0.64) > Cd (0.34) and Zn (2623.34) > Cu (1118.71) >Pb (169.56) > Cr (115.40) >Mn(92.8) > Ni (74.81) > As (18.26) >Cd (8.21). In both cases, thehighest concentrations were observed for Zn and Cu while thelowest for As and Cd. In our study we found Pb and Ni due to industrial pollution and may be accumulated from the soil as we found much amount of lead and Nickel in soil.

**4.4 Analysis between pre-monsoon and post monsoon**

In Satkhira Sadar, the concentration of Cd, Cu and Ni were in order of pre-monsoon >post monsoon but concentration of Pbwas in order of post monsoon (2.67 ppm) >pre-monsoon (1.66 ppm). The concentration of Chromium in both seasons was found below detectable limit.

In Morrelganj, the concentration of Cd, Cu and Ni were in order of pre-monsoon > post monsoon but concentration of Pb was in order of post monsoon (4.11 ppm) > pre-monsoon (2.38 ppm). The concentration of Chromium in both seasonwas found below detectable limit.

In Mongla, the concentration of Cd, Cu and Ni were in order of pre-monsoon > post monsoon but concentration of Pb was in order of post monsoon (4.69 ppm) > pre-monsoon (3.90 ppm). The concentration of Chromium in both seasons found was below detectable limit.

### 4.5 Estimated daily intake (EDI)

According to (WHO) 1985 the dietary exposure approach of fish consumption is a reliable tool by which we can investigate a population's diet in terms of intake levels of nutrients, bioactive compounds, and contaminants, providing important information about the potential nutritional deficiencies or exposure to food contaminants. The intake data can then be used to examine a specific element of interest. This study provides an estimate of the dietary intake and examines the dietary exposure to five trace elements through consumption of fish in the population's daily diet.

The Estimated Daily Intake (EDI**)** of heavy metals (Cr, Ni, Cu, As, Cd, and Pb) were evaluated according to the average concentration of each heavy metal in each food and the respective consumption rate (Santos et al. 2004). Islam et al.(2014) found in fish and vegetable in same descending order Cu > Ni > Cr >Pb> As > Cd. In the present study the EDI value is in order of Cu > Ni >Pb> Cd > Cr for Satkhira Sadar and Mongla region but for Morrelganj region the EDI value is in order of Cu >Pb> Ni > Cd > Cr.

### 4.6Target hazard Quotient

According to USEPA 2011, if the THQ value is greater than 1 then the fish is considered to be unsafe for human consumption. Islam et al.(2016) found that the consumer at high risk due to the exposure of Cu, As and Pb from vegetables and fish in respected area which were detect as non-carcinogenic risk. In the present study we found Cu where THQ > 1 for Satkhira Sadar, Mongla and Morrelganj. Highest value found in Satkhira Sadar 2.10 (Urban consumer) and lowest value in Mongla region 1.45 (Rural consumer). On the other hand the THQ value of Pb is 1.11 for Mongla region (Urban consumer).

### 4.7 Carcinogenic risk (CR)

The target carcinogenic risks (TR) derived from the intake of As and Pb were calculated since these elements may promote both non-carcinogenic and carcinogenic effects depending on the exposure dose. Inorganic As is classified as a known carcinogen (USEPA Group A) and Pb as probable carcinogen based on animal studies (USEPA Group B). In the present study we worked on Lead (Pb). In general, the excess cancer risks lower than $10^{-6}$ are considered to be negligible, cancer risks above $10^{-4}$ are considered unacceptable (USEPA 1989, 2010) and risks lying between 10E−6 and $10^{-4}$ are generally considered an acceptable range. In fish species TR values for Pb were lower than the $10^{-6}$ and regarded as negligible. In the research the TR value for Pb is acceptable range. In all region (Satkhira Sadar, Morrelganj and Mongla) the values is remain$10^{-5}$.

# Conclusion and Recommendation

## 5.1 Conclusion

The present study concludes that Shrimp collected from Satkhira Sadar, Morrelganj and Mongla accumulated various metals at concentration more than maximum allowable concentration (MAC). The investigated Shrimps varied widely in the metal concentration, in region verses region, in representative samples like Soil, Water and Shrimp of any area and also in various parts of Shrimp like Shell, Head and Muscle. In the estimated metal only Chromium was found below determination level in Shrimp and Water but in Soil was it found in higher concentration. Without Cd all the metals were in higher concentration. In water Cd, Cu and Cr were found below detectable limit and Pb and Ni were found in very low concentration. In Shrimp, without Cr all metals were found in higher concentration. In pre-monsoon and post monsoon analysis, it was found that the concentration of Cd, Cu and Ni in pre-monsoon were greater than post monsoon concentration and but concentration of Pb in post monsoon was greater than pre-monsoon and the concentration of Cr remained below detectable limit in both seasons. At current concentration in Shrimp, Cu was found to potential non-carcinogenic risk individually. People who continuously consume Shrimp contaminated with metals as found in the present study are under the target cancer risk in the long run.

## 5.2 Recommendations

Shrimp is called “White Gold” of Bangladesh. Shrimp culture is needed to fulfill the protein demand of growing people of our country. On the other hand it is one of the major sources of foreign currency. But if theconcentration of different harmful metals in Shrimp increase at alarming rate, consumption of those Shrimp can cause serious health risk including cancer, lesions in skin and many other diseases.

1) In this study, concentrations of Cd, Pb, Cu, Ni and Cr were determined in collected Shrimp, Soil and water; the other heavy metal like As, Fe, Zn, Co etc. can be determined. Those metals could accumulate at higher amount than studied metals.

2) Only three Upazila from two districts were selected in this study. The further study can be done in larger scale; more samples can be collected from different areas.

3) Current study is totally laboratory based, so field studies are needed to clarify the actual accumulation of heavy metals on cultured fish.

4) In the present study we used Atomic Absorption Spectrometer (Model No: AA-7000, Shimadzu) which is backdated. Now, scientists are using Inductively Coupled Plasma Mass Spectrometer (ICP-MS) worldwide for heavy metal analysis.

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# Appendices

## Mean and Standard Deviation of shell, head and muscle SPSS output

**Case Summaries[a]**

| | | | | | | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|---|---|---|---|---|
| Sampling_area | Satkhira | Shrimp_organ | Shell | 1 | | .21 | 5.15 | 44.21 | 1.81 | .00 |
| | | | | 2 | | .11 | 2.47 | 58.25 | 7.50 | .00 |
| | | | | 3 | | .05 | 4.08 | 59.36 | .00 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .1217 | 3.9000 | 53.9383 | 3.1017 | .0000 |
| | | | | | Std. Error of Mean | .04512 | .77887 | 4.87720 | 2.26005 | .00000 |
| | | | | | Std. Deviation | .07816 | 1.34904 | 8.44757 | 3.91453 | .00000 |
| | | | Head | 1 | | .46 | 3.22 | 108.25 | 1.53 | .00 |
| | | | | 2 | | .72 | .00 | 242.91 | .00 | .00 |
| | | | | 3 | | .39 | .00 | 153.00 | .00 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .5217 | 1.0733 | 168.0517 | .5100 | .0000 |
| | | | | | Std. Error of Mean | .10093 | 1.07333 | 39.59621 | .51000 | .00000 |
| | | | | | Std. Deviation | .17481 | 1.85907 | 68.58265 | .88335 | .00000 |
| | | | Muscle | 1 | | .11 | .00 | 23.73 | 13.62 | .00 |
| | | | | 2 | | .18 | .00 | 32.14 | 7.92 | .00 |
| | | | | 3 | | .08 | .00 | 35.56 | .00 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .1217 | .0000 | 30.4750 | 7.1783 | .0000 |
| | | | | | Std. Error of Mean | .02804 | .00000 | 3.51363 | 3.94777 | .00000 |
| | | | | | Std. Deviation | .04856 | .00000 | 6.08579 | 6.83773 | .00000 |
| | | | Total | N | | 9 | 9 | 9 | 9 | 9 |
| | | | | Mean | | .2550 | 1.6578 | 84.1550 | 3.5967 | .0000 |
| | | | | Std. Error of Mean | | .07435 | .69626 | 24.18783 | 1.63944 | .00000 |
| | | | | Std. Deviation | | .22306 | 2.08878 | 72.56349 | 4.91833 | .00000 |
| | Morrelganj | Shrimp_organ | Shell | 1 | | .11 | 6.76 | 53.97 | .98 | .00 |
| | | | | 2 | | .19 | 1.15 | 32.38 | .00 | .00 |
| | | | | 3 | | .41 | 6.76 | 53.17 | .00 | .00 |
| | | | | 4 | | .10 | 6.23 | 47.46 | .00 | .00 |
| | | | | Total | N | 4 | 4 | 4 | 4 | 4 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | Mean | .2012 | 5.2238 | 46.7425 | .2438 | .0000 |
| | | | | Std. Error of Mean | .07264 | 1.36376 | 5.00222 | .24375 | .00000 |
| | | | | Std. Deviation | .14528 | 2.72752 | 10.00444 | .48750 | .00000 |
| | | Head | 1 | | .82 | 2.15 | 230.53 | .00 | .00 |
| | | | 2 | | .38 | .00 | 99.83 | .00 | .00 |
| | | | 3 | | .90 | .00 | 165.46 | .00 | .00 |
| | | | 4 | | .80 | .00 | 170.70 | .00 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .7213 | .5363 | 166.6288 | .0000 | .0000 |
| | | | | Std. Error of Mean | .11728 | .53625 | 26.71352 | .00000 | .00000 |
| | | | | Std. Deviation | .23457 | 1.07250 | 53.42703 | .00000 | .00000 |
| | | Muscle | 1 | | .19 | .00 | 36.51 | .00 | .00 |
| | | | 2 | | .13 | 1.94 | 23.57 | 1.39 | .00 |
| | | | 3 | | .00 | .00 | 29.92 | 19.87 | .00 |
| | | | 4 | | .19 | 3.55 | 35.64 | 5.70 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .1263 | 1.3700 | 31.4075 | 6.7375 | .0000 |
| | | | | Std. Error of Mean | .04479 | .85653 | 2.99298 | 4.54063 | .00000 |
| | | | | Std. Deviation | .08957 | 1.71305 | 5.98596 | 9.08126 | .00000 |
| | | Total | N | | 12 | 12 | 12 | 12 | 12 |
| | | | Mean | | .3496 | 2.3767 | 81.5929 | 2.3271 | .0000 |
| | | | Std. Error of Mean | | .09098 | .80056 | 20.00530 | 1.66276 | .00000 |
| | | | Std. Deviation | | .31515 | 2.77322 | 69.30039 | 5.75997 | .00000 |
| Mongla | Shrimp_organ | Shell | 1 | | .00 | 8.37 | 35.64 | .00 | .00 |
| | | | 2 | | .11 | 8.91 | 55.55 | .00 | .00 |
| | | | 3 | | .18 | 7.30 | 54.76 | .00 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .0950 | 8.1900 | 48.6483 | .0000 | .0000 |
| | | | | Std. Error of Mean | .05107 | .47340 | 6.51066 | .00000 | .00000 |
| | | | | Std. Deviation | .08846 | .81995 | 11.27680 | .00000 | .00000 |
| | | Head | 1 | | .61 | 2.68 | 88.33 | .00 | .00 |
| | | | 2 | | .44 | 3.22 | 180.22 | .00 | .00 |
| | | | 3 | | 1.07 | 3.76 | 187.05 | .00 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .7050 | 3.2183 | 151.8633 | .0000 | .0000 |
| | | | | Std. Error of Mean | .18657 | .31033 | 31.83020 | .00000 | .00000 |
| | | | | Std. Deviation | .32315 | .53750 | 55.13152 | .00000 | .00000 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | 1 | | .21 | .00 | 19.29 | 5.28 | .00 |
| | | | 2 | | .16 | .86 | 33.57 | 30.56 | .00 |
| | | | 3 | | .13 | .00 | 33.73 | 41.95 | .00 |
| | | Muscle | | N | 3 | 3 | 3 | 3 | 3 |
| | | | Total | Mean | .1633 | .2867 | 28.8617 | 25.9300 | .0000 |
| | | | | Std. Error of Mean | .02315 | .28667 | 4.78856 | 10.83590 | .00000 |
| | | | | Std. Deviation | .04010 | .49652 | 8.29402 | 18.76832 | .00000 |
| | | | N | | 9 | 9 | 9 | 9 | 9 |
| | | Total | Mean | | .3211 | 3.8983 | 76.4578 | 8.6433 | .0000 |
| | | | Std. Error of Mean | | .11167 | 1.16780 | 21.29330 | 5.33493 | .00000 |
| | | | Std. Deviation | | .33502 | 3.50340 | 63.87991 | 16.00480 | .00000 |
| | N | | | | 30 | 30 | 30 | 30 | 30 |
| Total | Mean | | | | .3127 | 2.6175 | 80.8210 | 4.6028 | .0000 |
| | Std. Error of Mean | | | | .05291 | .52680 | 12.12366 | 1.80226 | .00000 |
| | Std. Deviation | | | | .28979 | 2.88542 | 66.40402 | 9.87140 | .00000 |

a. Limited to first 1000 cases.

## ANOVA test for Satkhira Sadar shell, head and Muscle

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .320 | 2 | .160 | 12.300 | .008 |
| | Within Groups | .078 | 6 | .013 | | |
| | Total | .398 | 8 | | | |
| Pb | Between Groups | 24.352 | 2 | 12.176 | 6.923 | .028 |
| | Within Groups | 10.552 | 6 | 1.759 | | |
| | Total | 34.904 | 8 | | | |
| Cu | Between Groups | 32499.720 | 2 | 16249.860 | 10.131 | .012 |
| | Within Groups | 9623.956 | 6 | 1603.993 | | |
| | Total | 42123.677 | 8 | | | |
| Ni | Between Groups | 67.803 | 2 | 33.901 | 1.618 | .274 |
| | Within Groups | 125.717 | 6 | 20.953 | | |
| | Total | 193.519 | 8 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 6 | .000 | | |
| | Total | .000 | 8 | | | |

## Homogeneous Subset

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Shell | 3 | .1217 | |
| Muscle | 3 | .1217 | |
| Head | 3 | | .5217 |
| Sig. | | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Pb**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 3 | .0000 | |
| Head | 3 | 1.0733 | 1.0733 |
| Shell | 3 | | 3.9000 |
| Sig. | | .608 | .089 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cu**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 3 | 30.4750 | |
| Shell | 3 | 53.9383 | |
| Head | 3 | | 168.0517 |
| Sig. | | .763 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Ni**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |

| | | |
|---|---|---|
| Head | 3 | .5100 |
| Shell | 3 | 3.1017 |
| Muscle | 3 | 7.1783 |
| Sig. | | .253 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

## ANOVA test for Morrelganj shell, head and Muscle

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .840 | 2 | .420 | 14.974 | .001 |
| | Within Groups | .252 | 9 | .028 | | |
| | Total | 1.093 | 11 | | | |
| Pb | Between Groups | 50.026 | 2 | 25.013 | 6.511 | .018 |
| | Within Groups | 34.572 | 9 | 3.841 | | |
| | Total | 84.598 | 11 | | | |
| Cu | Between Groups | 43856.882 | 2 | 21928.441 | 21.999 | .000 |
| | Within Groups | 8971.105 | 9 | 996.789 | | |
| | Total | 52827.987 | 11 | | | |
| Ni | Between Groups | 116.829 | 2 | 58.415 | 2.119 | .176 |
| | Within Groups | 248.121 | 9 | 27.569 | | |
| | Total | 364.950 | 11 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 9 | .000 | | |
| | Total | .000 | 11 | | | |

**Homogeneous Subset**

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 4 | .1263 | |
| Shell | 4 | .2012 | |
| Head | 4 | | .7213 |
| Sig. | | .806 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Pb**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Head | 4 | .5363 | |
| Muscle | 4 | 1.3700 | 1.3700 |
| Shell | 4 | | 5.2238 |
| Sig. | | .823 | .051 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Cu**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 4 | 31.4075 | |
| Shell | 4 | 46.7425 | |
| Head | 4 | | 166.6288 |
| Sig. | | .777 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Ni**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Head | 4 | .0000 |
| Shell | 4 | .2438 |
| Muscle | 4 | 6.7375 |
| Sig. | | .219 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

## ANOVA test for Mongla shell, head and Muscle

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .670 | 2 | .335 | 8.829 | .016 |
| | Within Groups | .228 | 6 | .038 | | |
| | Total | .898 | 8 | | | |
| Pb | Between Groups | 95.775 | 2 | 47.887 | 118.949 | .000 |
| | Within Groups | 2.416 | 6 | .403 | | |
| | Total | 98.190 | 8 | | | |
| Cu | Between Groups | 26174.258 | 2 | 13087.129 | 12.135 | .008 |
| | Within Groups | 6470.884 | 6 | 1078.481 | | |
| | Total | 32645.142 | 8 | | | |
| Ni | Between Groups | 1344.730 | 2 | 672.365 | 5.726 | .041 |
| | Within Groups | 704.500 | 6 | 117.417 | | |
| | Total | 2049.230 | 8 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 6 | .000 | | |
| | Total | .000 | 8 | | | |

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Shell | 3 | .0950 | |
| Muscle | 3 | .1633 | |
| Head | 3 | | .7050 |
| Sig. | | .905 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

Pb

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | | |
|---|---|---|---|---|
| | | 1 | 2 | 3 |
| Muscle | 3 | .2867 | | |
| Head | 3 | | 3.2183 | |
| Shell | 3 | | | 8.1900 |
| Sig. | | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

Cu

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 3 | 28.8617 | |
| Shell | 3 | 48.6483 | |
| Head | 3 | | 151.8633 |
| Sig. | | .751 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

Ni
Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Shell | 3 | .0000 |
| Head | 3 | .0000 |
| Muscle | 3 | 25.9300 |
| Sig. | | .059 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Case summaries of Soil water and Shrimp**

**Case Summaries[a]**

| | | | | | | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|---|---|---|---|---|
| Sampling_area | Satkhira | Sample | Soil | 1 | | .06 | 6.97 | 45.63 | 44.31 | 46.90 |
| | | | | 2 | | .00 | 10.73 | 45.08 | 38.06 | 49.07 |
| | | | | 3 | | .06 | 10.19 | 36.90 | 31.81 | 40.38 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .0400 | 9.2933 | 42.5350 | 38.0600 | 45.4467 |
| | | | | | Std. Error of Mean | .02000 | 1.17208 | 2.82205 | 3.60844 | 2.60963 |
| | | | | | Std. Deviation | .03464 | 2.03010 | 4.88794 | 6.25000 | 4.52002 |
| | | | Water | 1 | | .00 | .14 | .00 | .02 | .00 |
| | | | | 2 | | .00 | .13 | .00 | .01 | .00 |
| | | | | 3 | | .00 | .11 | .00 | .00 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .0000 | .1251 | .0000 | .0083 | .0000 |
| | | | | | Std. Error of Mean | .00000 | .00947 | .00000 | .00577 | .00000 |
| | | | | | Std. Deviation | .00000 | .01640 | .00000 | .01000 | .00000 |
| | | | Shrimp | 1 | | .26 | 2.79 | 58.73 | 5.65 | .00 |
| | | | | 2 | | .34 | .82 | 111.10 | 5.14 | .00 |
| | | | | 3 | | .17 | 1.36 | 82.64 | .00 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .2550 | 1.6578 | 84.1550 | 3.5967 | .0000 |
| | | | | | Std. Error of Mean | .04668 | .58693 | 15.13789 | 1.80435 | .00000 |
| | | | | | Std. Deviation | .08085 | 1.01659 | 26.21959 | 3.12523 | .00000 |
| | | | Total | N | | 9 | 9 | 9 | 9 | 9 |
| | | | | Mean | | .0983 | 3.6921 | 42.2300 | 13.8883 | 15.1489 |
| | | | | Std. Error of Mean | | .04222 | 1.46732 | 12.93479 | 6.17588 | 7.61181 |
| | | | | Std. Deviation | | .12665 | 4.40195 | 38.80437 | 18.52764 | 22.83544 |
| | Morrelganj | Sample | Soil | 1 | | .00 | 6.44 | 31.11 | 26.95 | 35.61 |
| | | | | 2 | | .09 | 5.36 | 30.79 | 28.75 | 34.74 |
| | | | | 3 | | 1.17 | 14.48 | 33.25 | 33.20 | 38.21 |
| | | | | 4 | | .03 | 4.83 | 28.97 | 28.20 | 38.65 |
| | | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | | Mean | .3225 | 7.7738 | 31.0288 | 29.2725 | 36.7988 |
| | | | | | Std. Error of Mean | .28312 | 2.25869 | .87829 | 1.36249 | .96109 |
| | | | | | Std. Deviation | .56624 | 4.51739 | 1.75657 | 2.72498 | 1.92218 |
| | | | Water | 1 | | .00 | .09 | .00 | .01 | .00 |
| | | | | 2 | | .00 | .09 | .00 | .00 | .00 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | 3 | | .00 | .04 | .00 | .03 | .00 |
| | | | 4 | | .00 | .11 | .00 | .03 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .0008 | .0804 | .0000 | .0181 | .0000 |
| | | | | Std. Error of Mean | .00085 | .01349 | .00000 | .00822 | .00000 |
| | | | | Std. Deviation | .00170 | .02697 | .00000 | .01644 | .00000 |
| | | Shrimp | 1 | | .37 | 2.97 | 107.00 | .33 | .00 |
| | | | 2 | | .23 | 1.03 | 51.93 | .46 | .00 |
| | | | 3 | | .44 | 2.25 | 82.85 | 6.62 | .00 |
| | | | 4 | | .36 | 3.26 | 84.60 | 1.90 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .3496 | 2.3767 | 81.5929 | 2.3271 | .0000 |
| | | | | Std. Error of Mean | .04304 | .49646 | 11.31444 | 1.47505 | .00000 |
| | | | | Std. Deviation | .08608 | .99292 | 22.62888 | 2.95009 | .00000 |
| | | Total | N | | 12 | 12 | 12 | 12 | 12 |
| | | | Mean | | .2243 | 3.4103 | 37.5406 | 10.5392 | 12.2663 |
| | | | Std. Error of Mean | | .09867 | 1.19647 | 10.70071 | 4.04957 | 5.23837 |
| | | | Std. Deviation | | .34181 | 4.14471 | 37.06835 | 14.02811 | 18.14624 |
| Mongla | Sample | Soil | 1 | | .00 | 7.51 | 34.21 | 30.14 | 42.77 |
| | | | 2 | | .00 | 9.65 | 37.46 | 33.06 | 44.73 |
| | | | 3 | | .06 | 10.73 | 33.41 | 27.09 | 37.13 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .0200 | 9.2933 | 35.0250 | 30.0950 | 41.5400 |
| | | | | Std. Error of Mean | .02000 | .94649 | 1.23894 | 1.72498 | 2.27850 |
| | | | | Std. Deviation | .03464 | 1.63936 | 2.14591 | 2.98775 | 3.94648 |
| | | Water | 1 | | .00 | .16 | .00 | .01 | .00 |
| | | | 2 | | .00 | .09 | .00 | .01 | .00 |
| | | | 3 | | .00 | .06 | .00 | .02 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .0000 | .1037 | .0000 | .0148 | .0000 |
| | | | | Std. Error of Mean | .00000 | .02928 | .00000 | .00404 | .00000 |
| | | | | Std. Deviation | .00000 | .05072 | .00000 | .00699 | .00000 |
| | | Shrimp | 1 | | .27 | 3.68 | 47.75 | 1.76 | .00 |
| | | | 2 | | .24 | 4.33 | 89.78 | 10.19 | .00 |
| | | | 3 | | .46 | 3.68 | 91.85 | 13.98 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .3211 | 3.8983 | 76.4578 | 8.6433 | .0000 |
| | | | | Std. Error of Mean | .06770 | .21500 | 14.36709 | 3.61197 | .00000 |
| | | | | Std. Deviation | .11726 | .37239 | 24.88454 | 6.25611 | .00000 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | Total | N | 9 | 9 | 9 | 9 | 9 |
| | | Mean | .1137 | 4.4318 | 37.1609 | 12.9177 | 13.8467 |
| | | Std. Error of Mean | .05579 | 1.36225 | 11.80684 | 4.61816 | 6.95451 |
| | | Std. Deviation | .16736 | 4.08676 | 35.42052 | 13.85449 | 20.86352 |
| Total | N | | 30 | 30 | 30 | 30 | 30 |
| | Mean | | .1533 | 3.8013 | 38.8335 | 12.2575 | 13.6052 |
| | Std. Error of Mean | | .04471 | .74519 | 6.55190 | 2.73540 | 3.60708 |
| | Std. Deviation | | .24491 | 4.08156 | 35.88626 | 14.98243 | 19.75678 |

a. Limited to first 1000 cases.

## Satkhira Sadar

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .113 | 2 | .056 | 21.881 | .002 |
| | Within Groups | .015 | 6 | .003 | | |
| | Total | .128 | 8 | | | |
| Pb | Between Groups | 144.707 | 2 | 72.354 | 42.107 | .000 |
| | Within Groups | 10.310 | 6 | 1.718 | | |
| | Total | 155.017 | 8 | | | |
| Cu | Between Groups | 10623.515 | 2 | 5311.757 | 22.401 | .002 |
| | Within Groups | 1422.717 | 6 | 237.120 | | |
| | Total | 12046.232 | 8 | | | |
| Ni | Between Groups | 2648.530 | 2 | 1324.265 | 81.360 | .000 |
| | Within Groups | 97.659 | 6 | 16.277 | | |
| | Total | 2746.189 | 8 | | | |
| Cr | Between Groups | 4130.799 | 2 | 2065.400 | 303.281 | .000 |
| | Within Groups | 40.861 | 6 | 6.810 | | |
| | Total | 4171.660 | 8 | | | |

**Cd**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0000 | |
| Soil | 3 | .0400 | |
| Shrimp | 3 | | .2550 |
| Sig. | | .623 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Pb**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .1251 | |
| Shrimp | 3 | 1.6578 | |
| Soil | 3 | | 9.2933 |
| Sig. | | .384 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cu**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | | |
|---|---|---|---|---|
| | | 1 | 2 | 3 |
| Water | 3 | .0000 | | |
| Soil | 3 | | 42.5350 | |
| Shrimp | 3 | | | 84.1550 |
| Sig. | | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Ni**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0083 | |
| Shrimp | 3 | 3.5967 | |
| Soil | 3 | | 38.0600 |
| Sig. | | .554 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cr**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0000 | |
| Shrimp | 3 | .0000 | |
| Soil | 3 | | 45.4467 |
| Sig. | | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

## Morrelganj

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .301 | 2 | .151 | 1.377 | .301 |
| | Within Groups | .984 | 9 | .109 | | |
| | Total | 1.285 | 11 | | | |
| Pb | Between Groups | 124.785 | 2 | 62.392 | 8.749 | .008 |
| | Within Groups | 64.180 | 9 | 7.131 | | |

| | | | | | | |
|---|---|---|---|---|---|---|
| | Total | 188.965 | 11 | | | |
| Cu | Between Groups | 13569.230 | 2 | 6784.615 | 39.510 | .000 |
| | Within Groups | 1545.456 | 9 | 171.717 | | |
| | Total | 15114.685 | 11 | | | |
| Ni | Between Groups | 2116.280 | 2 | 1058.140 | 196.816 | .000 |
| | Within Groups | 48.387 | 9 | 5.376 | | |
| | Total | 2164.666 | 11 | | | |
| Cr | Between Groups | 3611.061 | 2 | 1805.531 | 1466.008 | .000 |
| | Within Groups | 11.084 | 9 | 1.232 | | |
| | Total | 3622.146 | 11 | | | |

**Cd**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Water | 4 | .0009 |
| Soil | 4 | .3225 |
| Shrimp | 4 | .3496 |
| Sig. | | .339 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Pb**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 4 | .0804 | |
| Shrimp | 4 | 2.3767 | |
| Soil | 4 | | 7.7738 |
| Sig. | | .474 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Cu**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | | |
|---|---|---|---|---|
| | | 1 | 2 | 3 |
| Water | 4 | .0000 | | |
| Soil | 4 | | 31.0288 | |
| Shrimp | 4 | | | 81.5929 |
| Sig. | | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Ni**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 4 | .0181 | |
| Shrimp | 4 | 2.3271 | |
| Soil | 4 | | 29.2725 |
| Sig. | | .377 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Cr**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 4 | .0000 | |
| Shrimp | 4 | .0000 | |
| Soil | 4 | | 36.7988 |
| Sig. | | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

## Mongla

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .194 | 2 | .097 | 19.482 | .002 |
| | Within Groups | .030 | 6 | .005 | | |
| | Total | .224 | 8 | | | |
| Pb | Between Groups | 127.955 | 2 | 63.978 | 67.851 | .000 |
| | Within Groups | 5.658 | 6 | .943 | | |
| | Total | 133.613 | 8 | | | |
| Cu | Between Groups | 8789.217 | 2 | 4394.609 | 21.133 | .002 |
| | Within Groups | 1247.690 | 6 | 207.948 | | |
| | Total | 10036.908 | 8 | | | |
| Ni | Between Groups | 1439.444 | 2 | 719.722 | 44.921 | .000 |
| | Within Groups | 96.131 | 6 | 16.022 | | |
| | Total | 1535.575 | 8 | | | |
| Cr | Between Groups | 3451.143 | 2 | 1725.572 | 332.380 | .000 |
| | Within Groups | 31.149 | 6 | 5.192 | | |
| | Total | 3482.293 | 8 | | | |

**Cd**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0000 | |
| Soil | 3 | .0200 | |
| Shrimp | 3 | | .3211 |
| Sig. | | .936 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Pb**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | | |
|---|---|---|---|---|
| | | 1 | 2 | 3 |
| Water | 3 | .1037 | | |
| Shrimp | 3 | | 3.8983 | |
| Soil | 3 | | | 9.2933 |
| Sig. | | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cu**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0000 | |
| Soil | 3 | 35.0250 | |
| Shrimp | 3 | | 76.4578 |
| Sig. | | .056 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Ni**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0148 | |
| Shrimp | 3 | 8.6433 | |
| Soil | 3 | | 30.0950 |
| Sig. | | .085 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

**Cr**

Tukey HSD

| Sample | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Water | 3 | .0000 | |
| Shrimp | 3 | .0000 | |
| Soil | 3 | | 41.5400 |
| Sig. | | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

## Shrimp

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .016 | 2 | .008 | .872 | .459 |
| | Within Groups | .063 | 7 | .009 | | |
| | Total | .078 | 9 | | | |
| Pb | Between Groups | 7.917 | 2 | 3.958 | 5.226 | .041 |
| | Within Groups | 5.302 | 7 | .757 | | |
| | Total | 13.219 | 9 | | | |
| Cu | Between Groups | 92.843 | 2 | 46.422 | .078 | .925 |
| | Within Groups | 4149.613 | 7 | 592.802 | | |
| | Total | 4242.456 | 9 | | | |
| Ni | Between Groups | 72.730 | 2 | 36.365 | 2.054 | .199 |
| | Within Groups | 123.921 | 7 | 17.703 | | |
| | Total | 196.651 | 9 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 7 | .000 | | |
| | Total | .000 | 9 | | | |

**Cd**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira | 3 | .2550 |
| Mongla | 3 | .3211 |
| Morrelganj | 4 | .3496 |
| Sig. | | .450 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Pb**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Satkhira | 3 | 1.6578 | |
| Morrelganj | 4 | 2.3767 | 2.3767 |
| Mongla | 3 | | 3.8983 |
| Sig. | | .568 | .132 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Cu**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |

| | | |
|---|---|---|
| Mongla | 3 | 76.4578 |
| Morrelganj | 4 | 81.5929 |
| Satkhira | 3 | 84.1550 |
| Sig. | | .915 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Ni**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Morrelganj | 4 | 2.3271 |
| Satkhira | 3 | 3.5967 |
| Mongla | 3 | 8.6433 |
| Sig. | | .203 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

## Soil

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .206 | 2 | .103 | .746 | .509 |
| | Within Groups | .967 | 7 | .138 | | |
| | Total | 1.173 | 9 | | | |
| Pb | Between Groups | 5.542 | 2 | 2.771 | .259 | .779 |
| | Within Groups | 74.838 | 7 | 10.691 | | |
| | Total | 80.380 | 9 | | | |
| Cu | Between Groups | 228.797 | 2 | 114.398 | 12.087 | .005 |
| | Within Groups | 66.250 | 7 | 9.464 | | |
| | Total | 295.047 | 9 | | | |
| Ni | Between Groups | 150.573 | 2 | 75.287 | 4.457 | .056 |
| | Within Groups | 118.255 | 7 | 16.894 | | |
| | Total | 268.828 | 9 | | | |
| Cr | Between Groups | 130.455 | 2 | 65.227 | 5.495 | .037 |
| | Within Groups | 83.095 | 7 | 11.871 | | |
| | Total | 213.550 | 9 | | | |

**Cd**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Mongla | 3 | .0200 |
| Satkhira | 3 | .0400 |
| Morrelganj | 4 | .3225 |
| Sig. | | .577 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Pb**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Morrelganj | 4 | 7.7738 |
| Satkhira | 3 | 9.2933 |

| | | |
|---|---|---|
| Mongla | 3 | 9.2933 |
| Sig. | | .827 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Cu**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Morrelganj | 4 | 31.0288 | |
| Mongla | 3 | 35.0250 | |
| Satkhira | 3 | | 42.5350 |
| Sig. | | .284 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Ni**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Morrelganj | 4 | 29.2725 |
| Mongla | 3 | 30.0950 |
| Satkhira | 3 | 38.0600 |
| Sig. | | .067 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Cr**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Morrelganj | 4 | 36.7988 | |
| Mongla | 3 | 41.5400 | 41.5400 |
| Satkhira | 3 | | 45.4467 |
| Sig. | | .250 | .369 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

## Water

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .000 | 2 | .000 | .700 | .528 |
| | Within Groups | .000 | 7 | .000 | | |
| | Total | .000 | 9 | | | |
| Pb | Between Groups | .003 | 2 | .002 | 1.538 | .279 |
| | Within Groups | .008 | 7 | .001 | | |
| | Total | .011 | 9 | | | |
| Cu | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 7 | .000 | | |
| | Total | .000 | 9 | | | |
| Ni | Between Groups | .000 | 2 | .000 | .520 | .616 |
| | Within Groups | .001 | 7 | .000 | | |
| | Total | .001 | 9 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |

| | | | | | |
|---|---|---|---|---|---|
| Within Groups | .000 | 7 | .000 | | |
| Total | .000 | 9 | | | |

**Cd**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira | 3 | .0000 |
| Mongla | 3 | .0000 |
| Morrelganj | 4 | .0009 |
| Sig. | | .613 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Pb**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Morrelganj | 4 | .0804 |
| Mongla | 3 | .1037 |
| Satkhira | 3 | .1251 |
| Sig. | | .269 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Ni**

Tukey HSD

| Sampling_area | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira | 3 | .0083 |
| Mongla | 3 | .0148 |
| Morrelganj | 4 | .0181 |
| Sig. | | .605 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.273.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Post monsoon**

**Case Summaries[a]**

| | | | | | | Cd | Pb | Cu | Ni | Cr |
|---|---|---|---|---|---|---|---|---|---|---|
| Sampling_region | Satkhira Sadar | Shrimp_organ | Shell | 1 | | .00 | .00 | 167.24 | 3.83 | .00 |
| | | | | 2 | | .00 | .00 | 125.68 | 2.30 | .00 |
| | | | | 3 | | .00 | .00 | 138.98 | 2.63 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .0000 | .0000 | 143.9633 | 2.9150 | .0000 |
| | | | | | Std. Error of Mean | .00000 | .00000 | 12.25244 | .46487 | .00000 |
| | | | | | Std. Deviation | .00000 | .00000 | 21.22185 | .80517 | .00000 |
| | | | Head | 1 | | .00 | 7.34 | 30.28 | 1.42 | .00 |
| | | | | 2 | | .00 | 4.67 | 24.85 | 2.19 | .00 |
| | | | | 3 | | .00 | 7.34 | 43.51 | 2.08 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .0000 | 6.4500 | 32.8767 | 1.8933 | .0000 |
| | | | | | Std. Error of Mean | .00000 | .89000 | 5.54094 | .23879 | .00000 |
| | | | | | Std. Deviation | .00000 | 1.54153 | 9.59718 | .41359 | .00000 |
| | | | Muscle | 1 | | .21 | 2.01 | 30.35 | .11 | .00 |
| | | | | 2 | | .00 | .00 | 24.17 | 1.20 | .00 |
| | | | | 3 | | .92 | 2.67 | 35.32 | .99 | .00 |
| | | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | | Mean | .3767 | 1.5583 | 29.9450 | .7633 | .0000 |
| | | | | | Std. Error of Mean | .27835 | .80247 | 3.22365 | .33497 | .00000 |
| | | | | | Std. Deviation | .48211 | 1.38991 | 5.58353 | .58018 | .00000 |
| | | | Total | N | | 9 | 9 | 9 | 9 | 9 |
| | | | | Mean | | .1256 | 2.6694 | 68.9283 | 1.8572 | .0000 |
| | | | | Std. Error of Mean | | .10197 | 1.03129 | 19.18344 | .35867 | .00000 |
| | | | | Std. Deviation | | .30590 | 3.09386 | 57.55033 | 1.07601 | .00000 |
| | Morrelganj | Shrimp_organ | Shell | 1 | | .24 | 1.34 | 133.54 | 2.63 | .00 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | 2 | | .00 | .00 | 98.16 | .88 | .00 |
| | | | 3 | | .45 | 6.01 | 133.27 | 2.84 | .00 |
| | | | 4 | | .00 | 1.34 | 107.69 | 3.17 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .1713 | 2.1688 | 118.1613 | 2.3775 | .0000 |
| | | | | Std. Error of Mean | .10817 | 1.31690 | 9.01098 | .51322 | .00000 |
| | | | | Std. Deviation | .21635 | 2.63379 | 18.02195 | 1.02644 | .00000 |
| | | Head | 1 | | .00 | 12.01 | 33.51 | 2.30 | .00 |
| | | | 2 | | .00 | 13.34 | 38.81 | 2.41 | .00 |
| | | | 3 | | .00 | 4.67 | 24.04 | 4.05 | .00 |
| | | | 4 | | .00 | 1.34 | 27.53 | 1.42 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .0000 | 7.8375 | 30.9688 | 2.5413 | .0000 |
| | | | | Std. Error of Mean | .00000 | 2.88619 | 3.26268 | .54755 | .00000 |
| | | | | Std. Deviation | .00000 | 5.77238 | 6.52537 | 1.09509 | .00000 |
| | | Muscle | 1 | | .00 | .00 | 25.65 | .66 | .00 |
| | | | 2 | | .90 | 3.34 | 19.14 | 1.53 | .00 |
| | | | 3 | | .21 | 2.67 | 50.76 | 1.31 | .00 |
| | | | 4 | | .00 | 3.34 | 15.45 | .55 | .00 |
| | | | Total | N | 4 | 4 | 4 | 4 | 4 |
| | | | | Mean | .2763 | 2.3350 | 27.7463 | 1.0100 | .0000 |
| | | | | Std. Error of Mean | .21211 | .79396 | 7.95441 | .24198 | .00000 |
| | | | | Std. Deviation | .42421 | 1.58792 | 15.90882 | .48396 | .00000 |
| | | Total | N | | 12 | 12 | 12 | 12 | 12 |
| | | | Mean | | .1492 | 4.1138 | 58.9587 | 1.9762 | .0000 |
| | | | Std. Error of Mean | | .07957 | 1.26608 | 13.17475 | .31523 | .00000 |
| | | | Std. Deviation | | .27565 | 4.38582 | 45.63867 | 1.09198 | .00000 |
| Mongla | Shrimp_organ | Shell | 1 | | .10 | 1.34 | 117.09 | 2.52 | .00 |
| | | | 2 | | .00 | 5.34 | 86.27 | 2.52 | .00 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | 3 | | .26 | .00 | 121.18 | 2.19 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .1183 | 2.2233 | 108.1783 | 2.4050 | .0000 |
| | | | | Std. Error of Mean | .07596 | 1.60285 | 11.01777 | .11000 | .00000 |
| | | | | Std. Deviation | .13156 | 2.77622 | 19.08333 | .19053 | .00000 |
| | | Head | 1 | | .00 | 10.01 | 35.32 | 3.39 | .00 |
| | | | 2 | | .00 | 10.86 | 32.63 | 3.28 | .00 |
| | | | 3 | | .00 | 6.67 | 23.57 | 9.85 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .0000 | 9.1767 | 30.5033 | 5.5050 | .0000 |
| | | | | Std. Error of Mean | .00000 | 1.27713 | 3.55470 | 2.17023 | .00000 |
| | | | | Std. Deviation | .00000 | 2.21205 | 6.15692 | 3.75895 | .00000 |
| | | Muscle | 1 | | .10 | .00 | 19.00 | .22 | .00 |
| | | | 2 | | 1.09 | 8.01 | 22.56 | .66 | .00 |
| | | | 3 | | .00 | .00 | 18.27 | .99 | .00 |
| | | | Total | N | 3 | 3 | 3 | 3 | 3 |
| | | | | Mean | .3933 | 2.6683 | 19.9417 | .6183 | .0000 |
| | | | | Std. Error of Mean | .34692 | 2.66833 | 1.32625 | .22303 | .00000 |
| | | | | Std. Deviation | .60088 | 4.62169 | 2.29713 | .38631 | .00000 |
| | | Total | N | | 9 | 9 | 9 | 9 | 9 |
| | | | Mean | | .1706 | 4.6894 | 52.8744 | 2.8428 | .0000 |
| | | | Std. Error of Mean | | .11791 | 1.48523 | 14.31073 | .95243 | .00000 |
| | | | Std. Deviation | | .35374 | 4.45570 | 42.93218 | 2.85728 | .00000 |
| Total | N | | | | 30 | 30 | 30 | 30 | 30 |
| | Mean | | | | .1485 | 3.8532 | 60.1243 | 2.2005 | .0000 |
| | Std. Error of Mean | | | | .05461 | .73243 | 8.66625 | .32707 | .00000 |
| | Std. Deviation | | | | .29912 | 4.01167 | 47.46702 | 1.79143 | .00000 |

a. Limited to first 1000 cases.

## Satkhira Shell, Head and Muscle

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .284 | 2 | .142 | 1.831 | .239 |
| | Within Groups | .465 | 6 | .077 | | |
| | Total | .749 | 8 | | | |
| Pb | Between Groups | 67.959 | 2 | 33.980 | 23.662 | .001 |
| | Within Groups | 8.616 | 6 | 1.436 | | |
| | Total | 76.576 | 8 | | | |
| Cu | Between Groups | 25349.023 | 2 | 12674.511 | 66.284 | .000 |
| | Within Groups | 1147.298 | 6 | 191.216 | | |
| | Total | 26496.320 | 8 | | | |
| Ni | Between Groups | 6.950 | 2 | 3.475 | 9.019 | .016 |
| | Within Groups | 2.312 | 6 | .385 | | |
| | Total | 9.262 | 8 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 6 | .000 | | |
| | Total | .000 | 8 | | | |

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Shell | 3 | .0000 |
| Head | 3 | .0000 |
| Muscle | 3 | .3767 |
| Sig. | | .295 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Pb**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Shell | 3 | .0000 | |
| Muscle | 3 | 1.5583 | |
| Head | 3 | | 6.4500 |
| Sig. | | .319 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cu**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |

| | | | |
|---|---|---|---|
| Muscle | 3 | 29.9450 | |
| Head | 3 | 32.8767 | |
| Shell | 3 | | 143.9633 |
| Sig. | | .964 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

Ni

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 3 | .7633 | |
| Head | 3 | 1.8933 | 1.8933 |
| Shell | 3 | | 2.9150 |
| Sig. | | .144 | .189 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Morrelganj**

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .244 | 2 | .122 | .969 | .432 |
| | Within Groups | .757 | 6 | .126 | | |
| | Total | 1.001 | 8 | | | |
| Pb | Between Groups | 90.905 | 2 | 45.453 | 4.015 | .078 |
| | Within Groups | 67.921 | 6 | 11.320 | | |
| | Total | 158.826 | 8 | | | |
| Cu | Between Groups | 13930.664 | 2 | 6965.332 | 51.296 | .000 |
| | Within Groups | 814.716 | 6 | 135.786 | | |
| | Total | 14745.380 | 8 | | | |
| Ni | Between Groups | 36.682 | 2 | 18.341 | 3.844 | .084 |
| | Within Groups | 28.631 | 6 | 4.772 | | |
| | Total | 65.312 | 8 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 6 | .000 | | |
| | Total | .000 | 8 | | | |

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Head | 3 | .0000 |
| Shell | 3 | .1183 |
| Muscle | 3 | .3933 |
| Sig. | | .419 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Pb**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Shell | 3 | 2.2233 |
| Muscle | 3 | 2.6683 |
| Head | 3 | 9.1767 |
| Sig. | | .098 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Cu**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 3 | 19.9417 | |
| Head | 3 | 30.5033 | |
| Shell | 3 | | 108.1783 |
| Sig. | | .543 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

**Ni**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Muscle | 3 | .6183 |
| Shell | 3 | 2.4050 |
| Head | 3 | 5.5050 |
| Sig. | | .075 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 3.000.

## Mongla

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .156 | 2 | .078 | 1.029 | .396 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | Within Groups | .680 | 9 | .076 | | |
| | Total | .836 | 11 | | | |
| Pb | Between Groups | 83.253 | 2 | 41.627 | 2.919 | .105 |
| | Within Groups | 128.336 | 9 | 14.260 | | |
| | Total | 211.589 | 11 | | | |
| Cu | Between Groups | 21050.385 | 2 | 10525.193 | 50.890 | .000 |
| | Within Groups | 1861.385 | 9 | 206.821 | | |
| | Total | 22911.770 | 11 | | | |
| Ni | Between Groups | 5.655 | 2 | 2.828 | 3.411 | .079 |
| | Within Groups | 7.461 | 9 | .829 | | |
| | Total | 13.117 | 11 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 9 | .000 | | |
| | Total | .000 | 11 | | | |

**Cd**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Head | 4 | .0000 |
| Shell | 4 | .1713 |
| Muscle | 4 | .2763 |
| Sig. | | .371 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Pb**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Shell | 4 | 2.1688 |
| Muscle | 4 | 2.3350 |
| Head | 4 | 7.8375 |
| Sig. | | .140 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Cu**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Muscle | 4 | 27.7462 | |
| Head | 4 | 30.9688 | |
| Shell | 4 | | 118.1613 |
| Sig. | | .946 | 1.000 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

**Ni**

Tukey HSD

| Shrimp_organ | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Muscle | 4 | 1.0100 |
| Shell | 4 | 2.3775 |
| Head | 4 | 2.5413 |
| Sig. | | .095 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 4.000.

## Average of shell, head and muscle

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Cd | Between Groups | .009 | 2 | .005 | .048 | .954 |
| | Within Groups | 2.586 | 27 | .096 | | |
| | Total | 2.595 | 29 | | | |

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Pb | Between Groups | 19.720 | 2 | 9.860 | .596 | .558 |
| | Within Groups | 446.991 | 27 | 16.555 | | |
| | Total | 466.711 | 29 | | | |
| Cu | Between Groups | 1186.945 | 2 | 593.472 | .250 | .781 |
| | Within Groups | 64153.470 | 27 | 2376.054 | | |
| | Total | 65340.415 | 29 | | | |
| Ni | Between Groups | 5.377 | 2 | 2.688 | .828 | .448 |
| | Within Groups | 87.691 | 27 | 3.248 | | |
| | Total | 93.068 | 29 | | | |
| Cr | Between Groups | .000 | 2 | .000 | . | . |
| | Within Groups | .000 | 27 | .000 | | |
| | Total | .000 | 29 | | | |

**Cd**

Tukey HSD

| Sampling_region | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira Sadar | 9 | .1256 |
| Morrelganj | 12 | .1492 |
| Mongla | 9 | .1706 |
| Sig. | | .945 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 9.818.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Pb**

Tukey HSD

| Sampling_region | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira Sadar | 9 | 2.6694 |
| Morrelganj | 12 | 4.1138 |
| Mongla | 9 | 4.6894 |
| Sig. | | .522 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 9.818.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Cu**

Tukey HSD

| Sampling_region | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Mongla | 9 | 52.8744 |
| Morrelganj | 12 | 58.9587 |
| Satkhira Sadar | 9 | 68.9283 |
| Sig. | | .748 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 9.818.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

**Ni**

Tukey HSD

| Sampling_region | N | Subset for alpha = 0.05 |
|---|---|---|
| | | 1 |
| Satkhira Sadar | 9 | 1.8572 |
| Morrelganj | 12 | 1.9763 |
| Mongla | 9 | 2.8428 |
| Sig. | | .457 |

Means for groups in homogeneous subsets are displayed.
a. Uses Harmonic Mean Sample Size = 9.818.
b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

## Pre-monsoon and post monsoon analysis

**Paired Samples Statistics**

| | | Mean | N | Std. Deviation | Std. Error Mean |
|---|---|---|---|---|---|
| Pair 1 | Pre_Cd | .2550 | 3 | .08085 | .04668 |
| | Post_Cd | .1256 | 3 | .16070 | .09278 |
| Pair 2 | Pre_Pb | 1.6578 | 3 | 1.01659 | .58693 |
| | Post_Pb | 2.6694 | 3 | .97005 | .56006 |
| Pair 3 | Pre_Cu | 84.1550 | 3 | 26.21959 | 15.13789 |
| | Post_Cu | 68.9283 | 3 | 9.41439 | 5.43540 |
| Pair 4 | Pre_Ni | 3.5967 | 3 | 3.12523 | 1.80435 |
| | Post_Ni | 1.8572 | 3 | .06400 | .03695 |
| Pair 5 | Pre_Cr | .0000[a] | 3 | .00000 | .00000 |
| | Post_Cr | .0000[a] | 3 | .00000 | .00000 |

a. The correlation and t cannot be computed because the standard error of the difference is 0.

**Paired Samples Statistics**

| | | Mean | N | Std. Deviation | Std. Error Mean |
|---|---|---|---|---|---|
| Pair 1 | Pre_Cd | .3211 | 3 | .11726 | .06770 |
| | Post_Cd | .1706 | 3 | .16592 | .09579 |
| Pair 2 | Pre_Pb | 3.8983 | 3 | .37239 | .21500 |
| | Post_Pb | 4.6894 | 3 | 3.02516 | 1.74658 |
| Pair 3 | Pre_Cu | 76.4578 | 3 | 24.88454 | 14.36709 |
| | Post_Cu | 52.8744 | 3 | 5.14817 | 2.97230 |
| Pair 4 | Pre_Ni | 8.6433 | 3 | 6.25611 | 3.61197 |
| | Post_Ni | 2.8428 | 3 | 1.29636 | .74845 |
| Pair 5 | Pre_Cr | .0000[a] | 3 | .00000 | .00000 |
| | Post_Cr | .0000[a] | 3 | .00000 | .00000 |

a. The correlation and t cannot be computed because the standard error of the difference is 0.