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ASSESSMENT OF LEVELS OF CADMIUM AND CHROMIUM IN SEAWATER AND TISSUES OF HERRING GULL (*LARUS ARGENTATUS*) FROM SELECTED AREAS IN KARACHI, PAKISTAN

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ABSTRACT

This study focuses on the levels of heavy metals Cadmium (Cd) and Chromium (Cr) in seawater and selected bird Herring gull (*Larus argentatus*) tissues collected quarterly from selected field sites of coastal area of Hawksbay, Sandspit, and Korangi Creek, Karachi. All samples were examined using Atomic Absorption Spectrometer (Perkin Elmer. A. Analyst-700). Peak levels of Cd and Cr were found in seawater samples at Sandspit and Korangi Creek during 3rd and 4th quarter, respectively. While, minimum levels were detected at Korangi Creek and Hawksbay samples during 1st and 3rd quarter, respectively. Levels of Cd (18.758 ug/g) and Cr (18.77 ug/g) were elevated in kidney at Hawksbay and Korangi Creek during 2nd quarter. However, lowest value of Cr (0.66 ug/g) and Cd (2.767 ug/g) were in liver of Herring gull (*Larus argentatus*) at Korangi Creek during 4th and 1st quarter, respectively.

Keywords: Herring gull, metals, coastal areas, body tissues, cadmium, chromium.

INTRODUCTION

Metal contamination is one of the harmful pollutants in natural environment due to their toxicity as it is hazardous to human and aquatic life (Ven *et al*., 2007; Wcislo *et al*., 2008). The coastal contamination by chemicals and heavy metals such as Cadmium, Chromium, Lead, Nickle and Zinc have increased during the last few decades (Mashiatullah *et al*., 2009; Nergis *et al*., 2012). Heavy metals are considered xenobiotic substances because they do not have valuable role in body functions and are damaging even in low levels. Cadmium, Chromium, Lead and Manganese are considered as toxic metals. Aquatic life is exposed to mixture of metals rather than single elements. Metals, such as Copper and Zinc are important in cellular metabolism and their levels inside the body can be regulated by the organism. However, Cadmium and Chromium are toxic even in low concentration and tend to accumulate in body tissues of an animal (Rainbow, 1997, 2002). Heavy metals can enter the body of animal through contaminated food and water and can have lethal effects on different organs (Namroodi *et al*., 2017). Accumulation of metals in marine organisms fluctuates depending on seasonal factors such as diet and temperature (Stewart *et al*., 1994).

As population of Pakistan including Karachi is increasing. That is why; anthropogenic activities in Karachi are increasing due to rapid increase in population. Out of 990 km coastline of Pakistan, 960 km is free of pollution and 30 km of Karachi coast gets contamination through anthropogenic activities (Jilani, 2014). Industries such as tanneries, textile, detergents, dyes, Pharmaceuticals, metallurgy and cement release their dissipate into coastal areas of Karachi via Malir and Lyari river (Qureshi *et al*., 1997). Sea birds get considerable levels of metals due to their position in marine food webs and their long lifespan. Therefore, the present study was carried out to detect the amount of Cd and Cr in seawater samples and body tissues of Herring gull (*Larus argentatus*) from Hawksbay, Sandspit and Korangi Creek coastal areas of Pakistan. From the ecological point of view, these coastal areas provide roosting, feeding and nesting grounds to water birds due to rich mangrove growth. These spots take 90% of unprocessed industrial and domestic waste of Karachi city through Lyari and Malir River. Since only 2% of industries treat their waste matter before discharging into the sea, the level of contamination is increasing.

MATERIALS AND METHODS

Collection of material

During the study period from 2007 to 2010, Seawater (collected in sterilized containers) and 32 Herring gulls (*Larus argentatus*) were caught with the help of local community from the three selected field sites of Hawksbay, Sandspit, and Korangi Creek, Karachi,

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Pakistan at regular interval of every three months i.e from January to March (1st quarter), from April to June (2nd quarter), from July to September (3rd quarter), and from October to December (4th quarter).

Handling of material

To assess levels of Cd and Cr in body tissues of *Larus argentatus,* all these birds were kept in polythene bags after shooting and brought to laboratory. Dissection took place in the Department of Zoology, University of Karachi and tissues (liver and kidney) were collected and stored in deep freeze in polythene bags till acid digestion.

Solvent Extraction Method

Seawater samples were digested by solvent extraction method according to Kremling (1983) to determine the levels of Cd and Cr. In the laboratory, 500 ml seawater was filtered by using Whattman No. 40 filter paper and acidified by using 0.1N 3ml nitric acid. Then acidified filtered water was taken in a beaker and 5 ml of citrate buffer was added to bring pH at 4.5. This water sample was transferred into separatory funnel with 5 ml of ammonium pyrolidine dithiocabamate (APDC) and 20 ml of methyl isobutyl ketone (MIBK) for 10-20 minutes. This mixture was shaken in a shaker and then allowed to settle for 15 minutes. As a result, two layers were formed. Lower aqueous layer is drained in a beaker and the upper organic layer was stored in another 100 ml flask. The procedure was repeated by adding 10 ml MIBK in the resulting aqueous layer and the resulted organic layer was separated and mixed with previous extract. The final extract was then evaporated to semi dryness and dissolved in 1 molar nitric acid and then filtered. The end product was sent to Atomic Absorption Spectrometer for heavy metal analysis.

Acid Digestion

Body tissues (Kidneys and Liver) were selected as study organs to investigate the level of Cd and Cr. Acid digestion technique (Benton and Jr., 1988) was used to digest the 0.5 gm tissue. All tissues were heated at 50 ºC to remove the extra moisture. 2.5ml of concentrated nitric acid was added to 0.5g of tissues in a beaker and the sample left for 24 hours.

After 24 hours, the sample was heated for 1 hour at 80°C on hot plate in the fuming chamber. The beaker was then placed to cool. Concentrated Perchloric acid (2.5ml) was added and beaker was placed again on hot plate in fuming chamber at 180 to 200°C for 3 hours till digestion is clear. Cover was removed from beaker and heated at 80 ºC till fumes disappeared and beaker was removed from hot plate and cooled. Bidistilled water was added to make up the volume up to 10 ml (Raza *et al*., 2012).

RESULTS AND DISCUSSION

Metals in Sea Water

The present study analysis shows maximum levels of Cd (0.068 mg/l, 0.018 mg/l, and 0.46 mg/l) and Cr (0.034 mg/l, 0.497 mg/l and 0.007 mg/l) at Sandspit, Korangi Creek and Hawksbay, respectively. Minimum concentration of Cd (0.04 mg/l, 0.003 mg/l and 0.02 mg/l) and Cr (0.024 mg/l, 0.315, and 0.002 mg/l) was found at Sandspit, Korangi Creek and Hawksbay, respectively (Figs. 1-3; Table 1). Harper (1991) and Ouseph (1992) found Cd ranged between 0.00011-0.0004 mg/l in Tolo harbor and Cochin estuary. Their results are lower compared to our findings may be due to greater anthropogenic activities in the present study area.

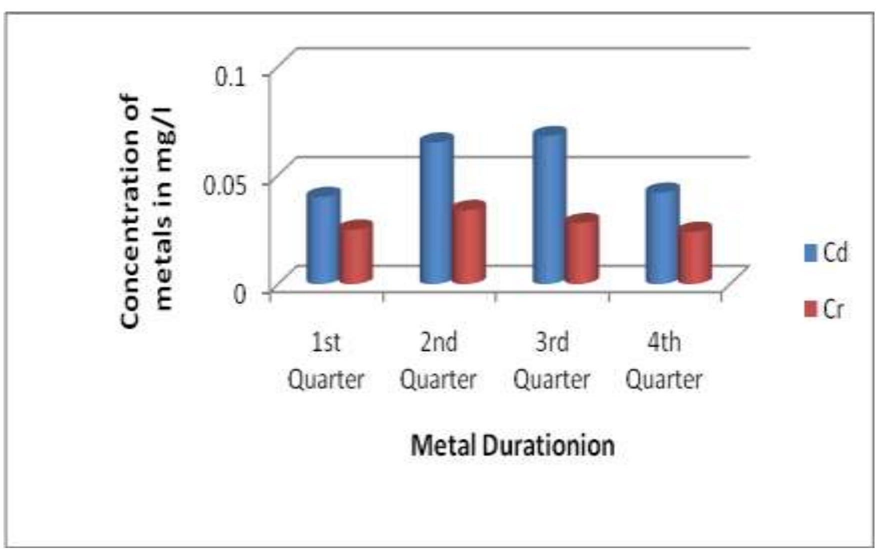

Fig. 1. Concentration (mg/l) of Cd and Cr in water samples at Sandspit.

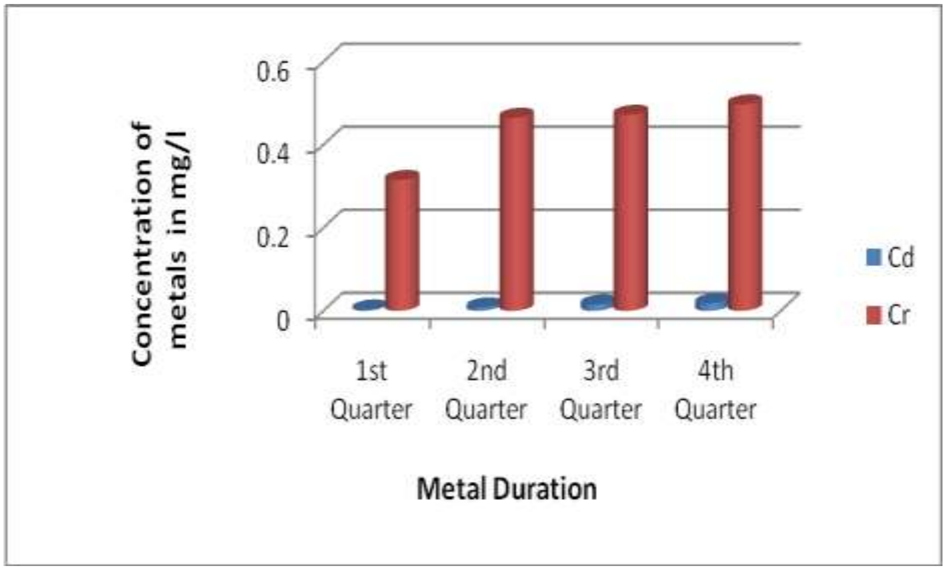

Fig. 2. Concentration (mg/l) of Cd and Cr in water samples at Korangi Creek.

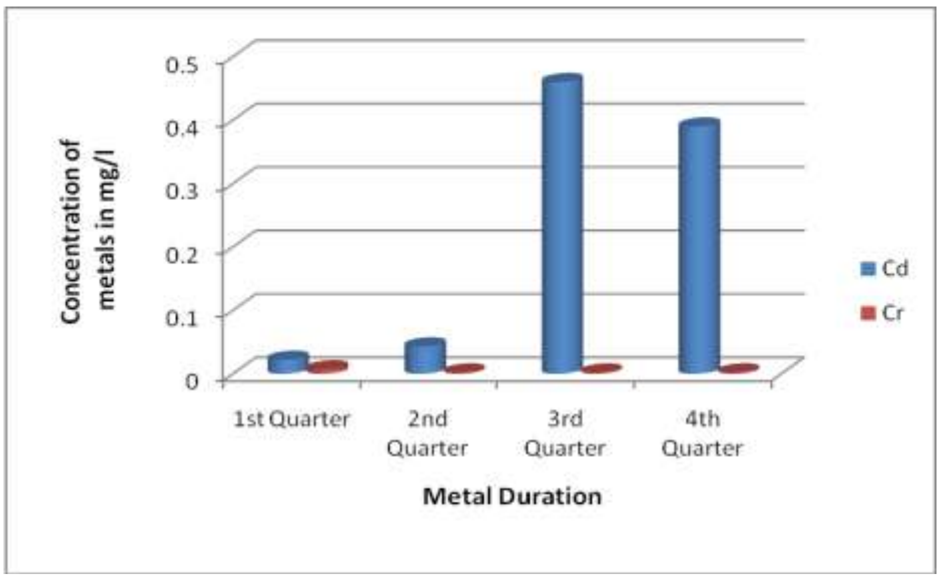

Fig. 3. Concentration (mg/l) of Cd and Cr in water samples at Hawksbay.

Fatoki and Mathabatha (2001) found higher concentrations of Cadmium i.e from 0.6 to 1.4 mg/l in different samples of water from East London harbor. In another research by Qari and Siddiqui (2004) Cadmium in water samples from Paradise point was found to be from 0.02-0.11 mg/l. This similarity in result could be due to contamination from industrial and agricultural waste dumped into sea water.

Srichandan (2016) reported distribution of trace metals in surface seawater of the Bay of Bengal, off Rushikulya estuary, East Coast of India. The rank order distribution of trace metals in terms of their average concentration in seawater was determined as Fe > Ni > Mn > Pb > As > Zn > Cr > V > Se > Cd. In our study, Cr levels were higher than Cd.

In the present study, highest level of Chromium (0.497 mg/l) was recorded in seawater samples from Korangi Creek while lowest level was found in Hawksbay (0.002 mg/l). High levels of Chromium may be due to contaminated waste from tanneries near Korangi creek (Raza *et al*., 2012). Khan *et al.* (2003) found Chromium (0.21-0.54 mg/l) in water samples collected from Gharo creek. In the current study, the similar results were reported for chromium (0.202-0.784 mg/l) in seawater at Korangi creek.

Qari and Siddiqui (2004) found the similar results in water samples from Paradise Point, Karachi i.e. 0.06 to 1.33 mg/l of Chromium, compared to our results from Korangi creek. It is because of untreated industrial waste which is directly dumped to sea. There are only three functional waste treatment plants, Shershah, Mehmood Abad and Mari Poor in Karachi.

Ismail *et al*. (2006) reported Cr 0.068 mg/l in Korangi Creek, 0.062 mg/l in Karachi harbor and 0.291 mg/l in Sandspit. However, in the present study, levels of Cr are high in Korangi Creek samples. Zhao *et al*. (2013) evaluated 2510 mg/l Cr in the seawater samples from Deer Island, Liaoning Province, China during 2010–2011. These results are much higher compared to our results, probably due to less impact of environmental pollutants. Sarinas *et al*. (2014) found 0.545 mg/l Cr at Villa Beach, Iloilo City. This result is in range with present report. In the Iranian coast along the Oman Sea, 201 mg/l Cr was detected in the Gulf Chabahar (Bazzi, 2014). Mortuza and Al-Misned (2017) studied environmental pollution and found 1.36 mg/l of Cr in water of Red Sea Coast of Jizan, Saudi Arabia. These values were higher than the suggested maximum absorption of Cr for drinking water (0.1 mg/l) set by WHO (1993). In the current study, amount of Cr was low but still higher than the recommended maximum level of Cr for drinking water (0.1 mg/l) set by World Health Organization - WHO, 1993.

Metal in Herring gull Body Tissues

Herring Gull (*Larus argentatus*) (Fig. 4) is classified as Least Concern but population trend appears to be decreasing (BirdLife International, 2018).

Fig. 4. Herring Gull (*Larus argentatus*).

In the present work, 18.758 ug/g of Cd and 18.77 ug/g of Cr were found in kidney of Herring Gull (*Larus argentatus*) from Hawksbay and Korangi Creek, respectively. However, in liver, highest value of Cd and Cr was 10.012 ug/g and 3.955 ug/g at Sandspit and Hawksbay, respectively. The minimum amount of Cd (2.767 ug/g) and Cr (0.66 ug/g) was found in liver of *Larus argentatus* at Korangi Creek. While, kidney of *Larus argentatus* had lowest level of Cd (7.319 ug/g) and Cr (1.095 ug/g) at Sandspit (Figs. 5-10, Table 2 and 3).

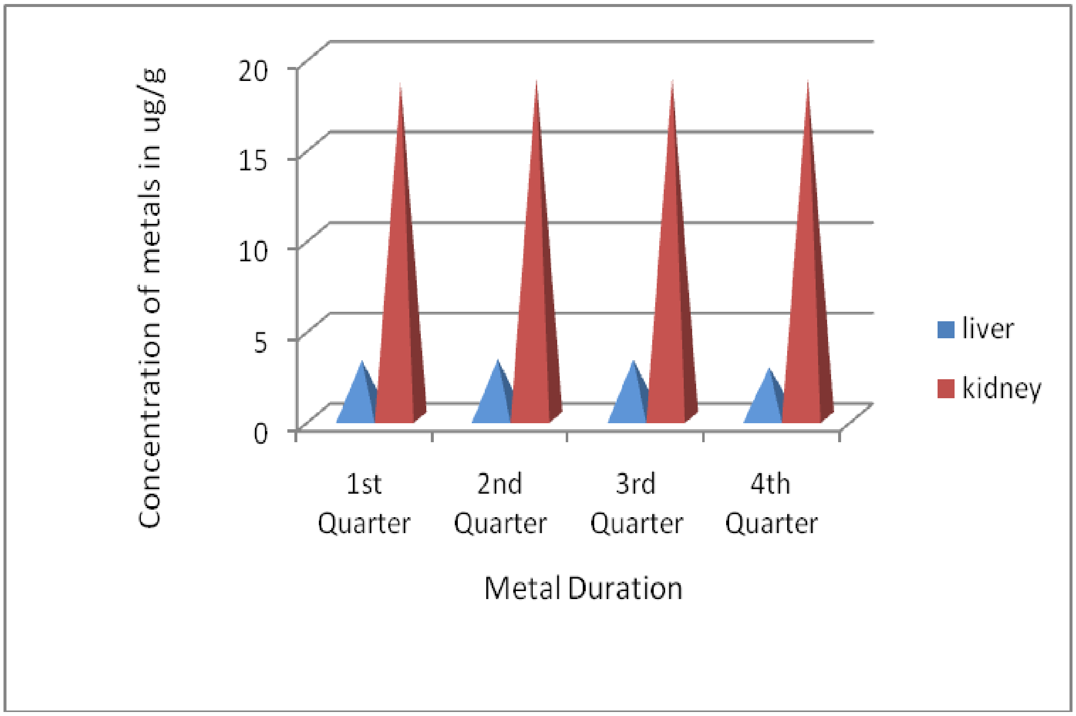

Fig. 5. Concentration (ug/g) of Cadmium in body tissues of Herring gull at Hawksbay.

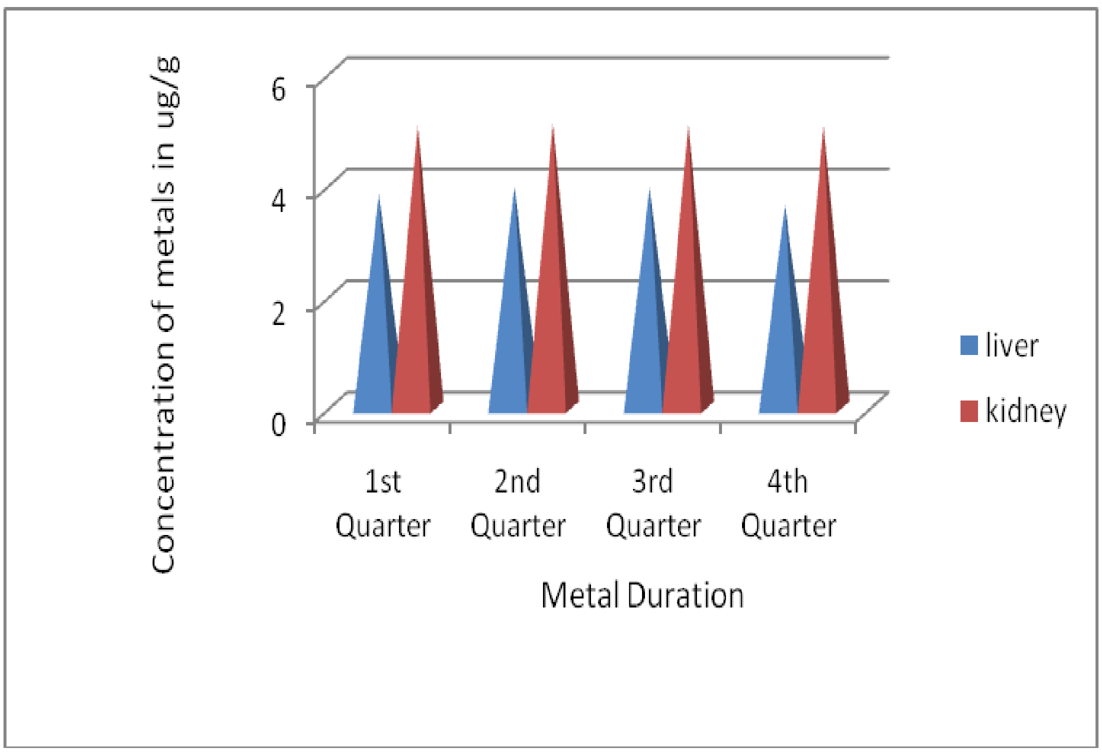

Fig. 6. Concentration (ug/g) of Chromium in body tissues of Herring gull at Hawksbay.

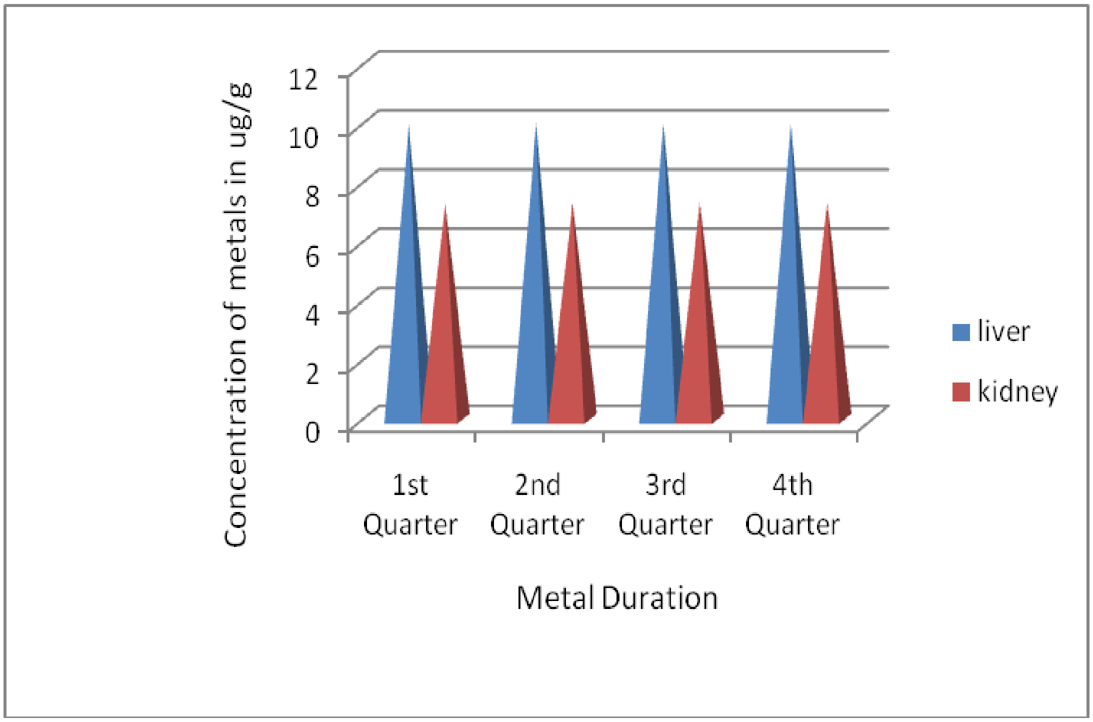

Fig. 7. Concentration (ug/g) of Cadmium in body tissues of Herring gull at Sandspit.

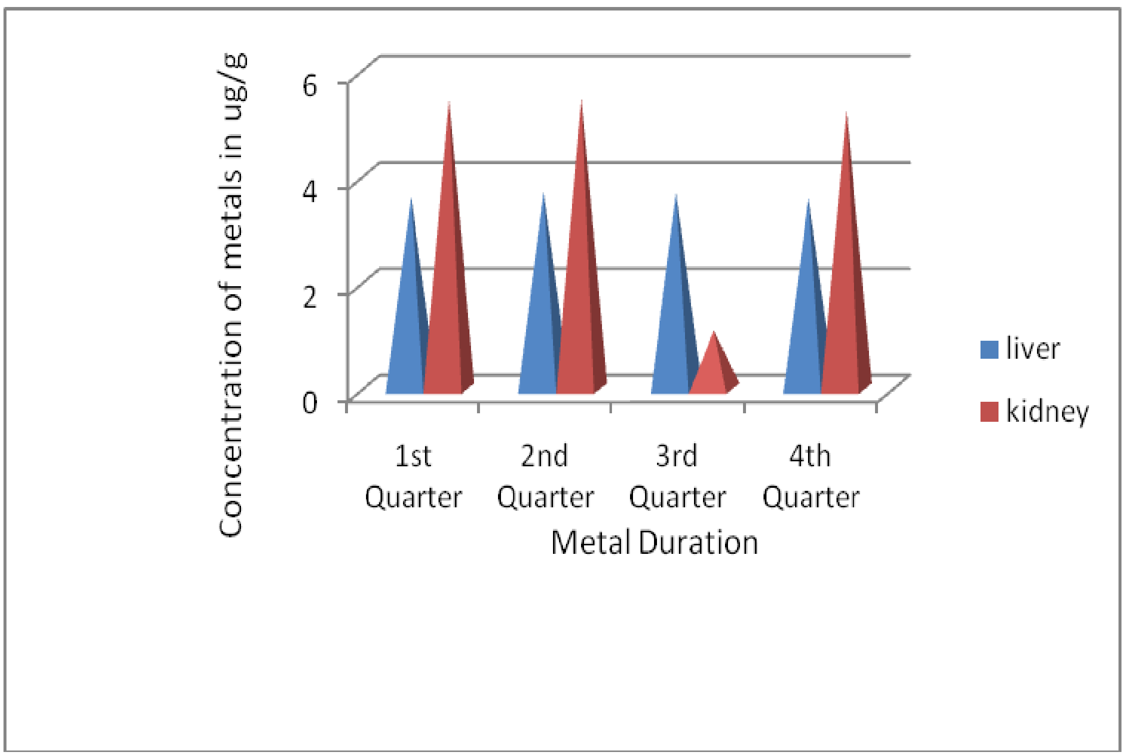

Fig. 8. Concentration (ug/g) of Chromium in body tissues of Herring gull at Sandspit.

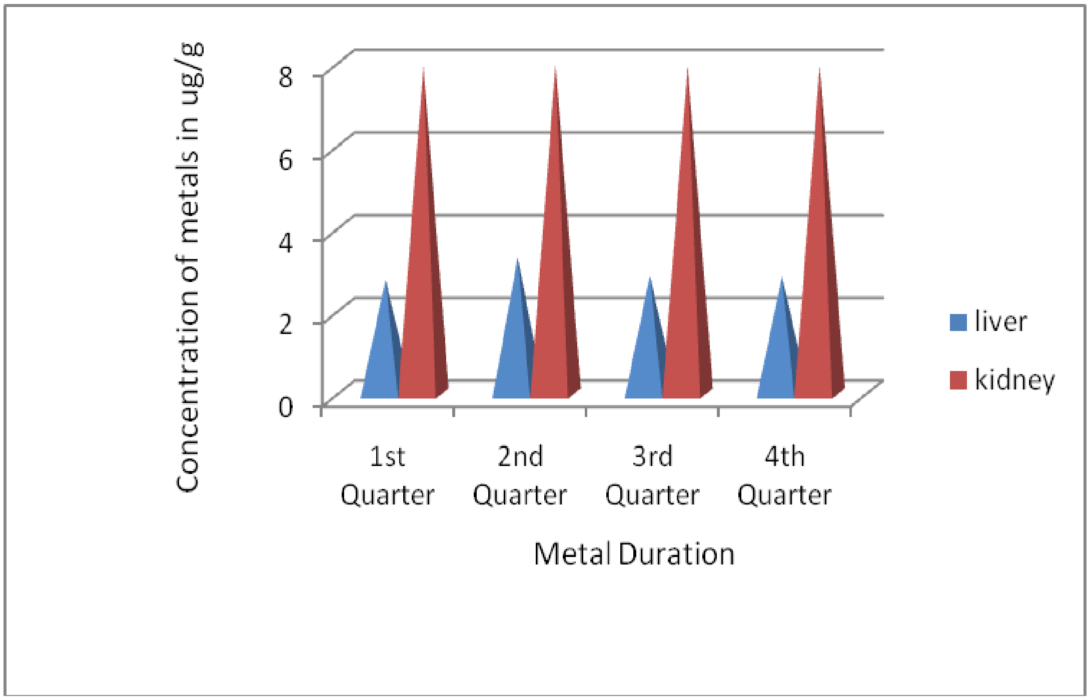

Fig. 9. Concentration (ug/g) of Cadmium in body tissues of Herring gull at Korangi Creek.

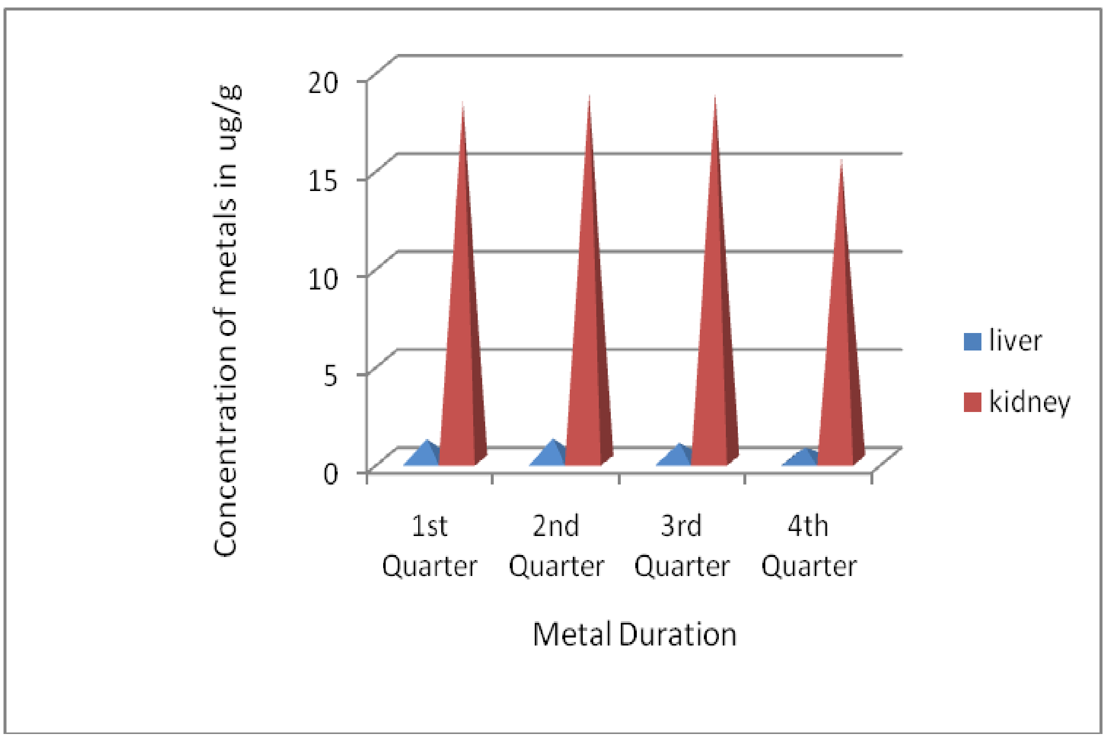

Fig. 10. Concentration (ug/g) of Chromium in body tissues of Herring gull at Korangi Creek.

Elliott *et al.* (1992) studied the concentrations of Chromium and Cadmium in the liver of Atlantic Sea birds. They found high levels of Cadmium (32 ug/g) in liver contents of Leach`s storm. Concentration of Chromium (2.68 ug/g) was also high as compared to our research.

Stoneburner *et al.* (1980) reported the concentrations of Cadmium and Chromium in Sooty terns in Florida. They found high concentrations of Cadmium in their samples, 3790 ug/g in liver and 23500 ug/g in kidneys. Another study by Norheim (1987) investigated the concentration of Cadmium in Seabirds *Larus hyperboreus* of Atlantic and found 9.4 ug/g in liver samples while 4.158 ug/g in kidney samples. Those findings are in the line of current investigations.

Kim and Koo (2007) found 13.4 ug/g of Cd in liver of *Nycticorax nycticorax* (Black crown night Heron). These results are higher as compared to present study, possibly due to less contamination in present study areas. Farreira (2010) collected samples of liver and kidney of *Egretta caerulea* (Little blue Heron) from Sepetiba Bay, Rio de Janeiro, Brazil to analyze Cadmium and Chromium. Cd in level of 6.32 ug/g and 6.57 ug/g and Cr in level of 2.80 ug/g and 4.16 ug/g were found in liver and kidney, respectively. In present findings, high level of Cr (18.77 ug/g) and Cd (18.758 ug/g) was detected in kidney at Korangi Creek and Hawksbay, respectively. However, maximum amount of Cd (10.012 ug/g) and Cr (3.955 ug/g) was found in Liver at Sandspit and Hawksbay, respectively due to high impact of pollution in these areas.

Plessl *et al*. (2017) analyzed liver samples for Cr (0.000001 ug/g) and Cd (0.0000002 ug/g) in Mallard *Anas platyrhynchos* collected from Eastern Austria. These findings are quiet low compared to our study because more than 200 tannery units are functioning in Karachi and the waste material (Cr, Cd, Pb, Mn, and Mg) is directly discarded into the marine water (Raza *et al*., 2012). Aloupi *et al*. (2017) detected six heavy metals (cadmium, chromium, copper, manganese, lead, and zinc) in liver of nine species of waterfowl from the Evros Delta, one of the most important wetlands in Greece, to evaluate metal contamination and possible risk to waterfowl. Significant differences among species were found. For hepatic Cr, Cu, Pb, and Zn contamination, the highest levels of all metals were found in liver of mute swans.

Raza *et al*. (2017) assessed the levels of metals in tissues of Kentish Plover (*Charadrius Alexandrinus*) from the coastal areas of Karachi, Pakistan. They reported the highest level of Cd (10.317 ug/g) in liver of Kentish plover at Sandspit and lowest Cd (0.786 ug/g) in the kidney from Korangi Creek. Maximum and lowest levels of Cr (5.403 ug/g and 0.723 ug/g) were detected in kidney during 2nd and 1st quarter at Hawksbay and Sandspit, respectively. In the present study, highest level of Cr was found in Kidney (18.77 ug/g), possibly due to monthly fluctuation of metal ions and their intake by birds. Cd is the most toxic element after mercury (Hg) for the aquatic life. Its accumulation in the body of the marine organisms is due to its regulatory ability, as reported by Pentreath (1976) and Olafson (1977).

Warren and Wallace (1990) determined levels of Cr in blue winged teal from South plains Texas. Cr was 0.20 ug/g in liver. In the present report, high amount of Cr (3.955 ug/g) was recorded in liver of *Larus argentatus* at Hawksbay and the lowest value was 0.66 ug/g at Korangi Creek. Another study Szefer *et al*. (1993) examined major essential elements in seals, Penguins and other representative fauna of Antarctic. Distinct inter tissue differences in the metal concentration were determined. Szarek *et al*. (2001) investigated Cd concentration in young coots originating from industrial and agricultural regions of North middle Poland. The results showed that in coots from industrial areas, levels of Cd were decreased due to better system of controlling pollutants production. In the present findings, low levels of Cd were detected in kidney at Sandspit, possibly due to less impact of pollution at this area. Agusta *et al*. (2005) observed 4.8 ug/g and 40.7 ug/g Cd in liver and kidney, respectively, of Black tailed gull (*Larus crassirostris*) collected from Rishiri Island, Japan.

In the present work, highest Cd (18.77 ug/g) level was recorded in kidney at Korangi Creek and 10.012 ug/g Cd in liver at Sandspit. Mansouri *et al*. (2012) studied the levels of chromium (0.96 and 2.32 μg/g) in kidney of Western Reef heron (*Egretta gularis*) and Siberian gull (*Larus heuglini*), respectively, whereas in liver they were (1.05 and 2.75 μg/g), respectively. These birds were collected from November to December 2010 throughout the Hara Biosphere Reserve. Present work shows the maximum Cr (18.77 ug/g and 3.955 ug/g) in Kidney and liver at Korangi Creek and Hawksbay, respectively. Coastal areas of Karachi are identified pollution areas of Pakistan (Shahzad *et al*., 2009; Saleem and Kazi, 1998; Hunter *et al*., 2012) and our study also confirmed levels of metal contamination.

CONCLUSION

The present study concluded that untreated waste from the industries and sewage enhances the accumulation of metals and is picked up by the marine organisms which through the food chain end up with marine birds as they are at higher trophic level in a marine environment. Coastal areas of Karachi are getting pollution through several anthropogenic activities. So, regular monitoring of these areas is required. If anthropogenic activities persist, then it will be dangerous in future for biodiversity.

Table 1. Concentration of Cd and Cr in water samples (in mg/l) at Coastal areas of Karachi.

Metal Duration	(Cd at Hawksbay) Mean SD± SE± *Range	(Cr at Hawksbay) Mean SD± SE± *Range	(Cd at Sandspit) Mean SD± SE± *Range	(Cr at Sandspit) Mean SD± SE± *Range	(Cd at Korangi Creek) Mean SD± SE± *Range	(Cr at Korangi Creek) Mean SD± SE± *Range
1st Quarter	0.022 0.002 0.001 0.019 to 0.024	0.007 0.006 0.003 0.0001 to 0.0138	0.04 0.051 0.03 0.018 to 0.098	0.025 0.013 0.007 0.009 to 0.040	0.003 0.001 0.0005 0.001 to 0.004	0.315 0.099 0.057 0.202 to 0.427
2nd Quarter	0.043 0.051 0.029 0.014 to 0.100	0.002 0.001 0.0005 0.0008 to 0.003	0.065 0.066 0.038 0.010 to 0.140	0.034 0.057 0.33 0.030 to 0.098	0.008 0.006 0.003 0.001 to 0.014	0.465 0.231 0.133 0.203 to 0.726
3rd Quarter	0.46 0.272 0.157 0.151 to 0.768	0.002 0.0002 0.0001 0.002 to 0.0024	0.068 0.057 0.033 0.003 to 0.132	0.028 0.012 0.007 0.014 to 0.041	0.015 0.005 0.003 0.009 to 0.020	0.472 0.213 0.123 0.230 to 0.713
4th Quarter	0.39 0.303 0.175 0.046 to 0.733	0.0021 0.002 0.001 0.0001 to 0.004	0.042 0.051 0.03 0.016 to 0.100	0.024 0.015 0.009 0.006 to 0.041	0.018 0.006 0.003 0.011 to 0.024	0.497 0.253 0.146 0.209 to 0.784

(*Range at 95% confidence limit)

Table 2. Concentration of Cd and Cr in Liver (in ug/g) of *Larus argentatus* at Coastal areas of Karachi.

Metal Duration	(Cd at Hawksbay) Mean SD± SE± *Range	(Cr at Hawksbay) Mean SD± SE± *Range	(Cd at Sandspit) Mean SD± SE± *Range	(Cr at Sandspit) Mean SD± SE± *Range	(Cd at Korangi Creek) Mean SD± SE± *Range	(Cr at Korangi Creek) Mean SD± SE± *Range
1st Quarter	3.170 1.051 0.607 1.981 to 4.359	3.821 0.529 0.305 3.222 to 4.419	10.003 0.001 0.005 10.001 to10.004	3.619 1.541 0.889 1.874 to 5.363	2.767 0.576 0.333 2.114 to 3.419	1.08 0.112 0.065 0.952 to 1.207
2nd Quarter	3.234 1.704 0.984 1.306 to 5.162	3.955 2.194 1.267 1.472 to 6.438	10.012 3.605 2.081 5.931 to 14.092	3.709 0.783 0.452 2.822 to 4.595	3.307 0.005 0.003 3.301 to 3.312	1.125 0.014 0.008 1.108 to 1.141
3rd Quarter	3.192 1.663 0.960 1.310 to 5.074	3.941 1.811 1.045 1.892 to 5.990	10.00 4.529 2.615 4.873 to 15.126	3.695 0.552 0.319 3.069 to 4.320	2.857 0.638 0.368 2.134 to 3.579	0.895 0.574 0.331 0.245 to 1.544
4th Quarter	2.780 0.580 0.335 2.123 to 3.437	3.625 0.597 0.345 2.949 to 4.301	9.991 4.919 2.840 4.424 to 15.557	3.597 0.551 0.318 2.973 to 4.220	2.843 0.640 0.37 2.117 to 3.568	0.66 0.051 0.03 0.601 to 0.718

(*Range at 95% confidence limit)

Table 3. Concentration of Cd and Cr in Kidney (in ug/g) of *Larus argentatus* at Coastal areas of Karachi.

Metal Duration	(Cd at Hawksbay) Mean SD± SE± *Range	(Cr at Hawksbay) Mean SD± SE± *Range	(Cd at Sandspit) Mean SD± SE± *Range	(Cr at Sandspit) Mean SD± SE± *Range	(Cd at Korangi Creek) Mean SD± SE± *Range	(Cr at Korangi Creek) Mean SD± SE± *Range
1st Quarter	18.517 5.562 3.211 12.222 to 24.811	5.050 1.527 0.882 3.322 to 6.778	7.32 0.1 0.057 7.206 to 7.433	5.462 0.571 0.33 4.815 to 6.108	7.951 0.640 0.37 7.225 to 8.676	18.36 4.731 2.731 13.005 to 23.714
2nd Quarter	18.758 5.824 3.363 12.166 to 25.349	5.100 2.00 1.155 2.837 to 7.363	7.333 0.095 0.551 7.224 to 7.441	5.478 0.116 0.067 5.346 to 5.609	7.96 0.639 0.369 7.236 to 8.683	18.77 5.776 3.335 12.233 to 25.306
3rd Quarter	18.741 5.741 3.315 12.243 to 25.238	5.070 2.101 1.213 2.693 to 7.447	7.321 0.1 0.057 7.207 to 7.434	1.095 0.054 0.031 1.032 to 1.157	7.946 0.642 0.370 7.218 to 8.673	18.75 5.741 3.315 12.252 to 25.247
4th Quarter	18.717 11.608 6.702 5.581 to 31.853	5.057 1.678 0.969 3.159 to 6.955	7.319 0.010 0.006 1.307 to 7.330	5.254 0.057 0.033 5.189 to 5.318	7.94 0.547 0.316 7.320 to 8.559	15.49 5.828 3.365 8.894 to 22.085

(*Range at 95% confidence limit)

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