

**LEVELS OF METAL CONTAMINATION IN VARIOUS SPECIES
OF BIRDS IN SELECT STATES IN INDIA**

Thesis submitted to Bharathiar University Coimbatore for the award of degree of
DOCTOR OF PHILOSOPHY IN ENVIRONMENTAL SCIENCES



By

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(A Centre of Excellence under the Ministry of Environment, Forests and Climate Change, Govt. of India)

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(Kirubhanandhini.V)

I dedicate this thesis to all the
Birds which lost their lives

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Abbreviations

ABBREVIATIONS

- 1) °C – Degree Celsius
- 2) AA – Atomic Absorption
- 3) AAS – Atomic Absorption Spectrophotometer
- 4) ANOVA – Analysis of variance
- 5) ATSDR – Agency for Toxic Substances and Disease Registry
- 6) BDL – Below Detection Limit
- 7) BF – Breast feather
- 8) Cd – Cadmium
- 9) CE – Critically Endangered
- 10) CNS – Central Nervous System
- 11) CPCB – Central Pollution Control Board
- 12) Cr – Chromium
- 13) Cu – Copper
- 14) df – Degree of Freedom
- 15) DW – Dry Weight
- 16) E – East
- 17) ENVIS – Environmental Information System
- 18) F – Frequency
- 19) H₂O₂ – Hydrogen Peroxide
- 20) HClO₄ – Perchloric Acid
- 21) HNO₃ – Nitric Acid
- 22) JCT – Jivdhaya Charitable Trust
- 23) km² – Kilo Meter Square

24)	L	–	Latitude
25)	m	–	Meter
26)	mL	–	Milli Liter
27)	MSL	–	Mean Sea Level
28)	N	–	North
29)	n	–	Number of samples
30)	NA	–	Not Analyzed
31)	NGO	–	Non-Government Organization
32)	Ni	–	Nickel
33)	NSAID	–	Non-Steroidal Anti-Inflammatory Drug
34)	NT	–	Near Threatened
35)	Pb	–	Lead
36)	PCBs	–	Poly Chlorinated Biphenyls
37)	PAHs	–	Poly Aromatic Hydrocarbons
38)	PF	–	Primary feather
39)	POPs	–	Persistent Organic Pollutants
40)	PPB	–	Parts Per Billion
41)	PPM	–	Parts Per Million
42)	SACON	–	Sálim Ali Centre for Ornithology and Natural History
43)	SE	–	Standard Error
44)	SEZ	–	Special Economic Zone
45)	SF	–	Secondary feather
46)	SISCO	–	Saudi Industrial Services Company
47)	SPCB	–	State Pollution Control Board

- 48) SPSS – Statistical Package for the Social Sciences
- 49) TF – Tail feather
- 50) TML – Total Metal Load
- 51) TNAU – Tamil Nadu Agricultural University
- 52) UK – United Kingdom
- 53) USA – United States of America
- 54) USEPA – United States Environmental Protection Agency
- 55) USFWS – United States of Fish and Wildlife Service
- 56) VUL – Vulnerable
- 57) WW – Wet Weight
- 58) Zn – Zinc

Introduction

1. INTRODUCTION

Metals rank among the main groups of toxic pollutants which are capable of damaging and threatening ecosystem stability even in low concentrations. The contaminants of most concern are predominantly metals such as cadmium, lead and copper which can accumulate and some of them can also magnify in the food chain (Albergoni and Piccinni 1983; Battaglia et al. 2005; Ali et al. 2013). The extinction of several species of fish, amphibians, reptilian, mammalian and avian groups has increased in recent decades due to metals (Wang and Murphy 1982; Purdom et al. 1994; Catherine and Gloria 2000; Baillie et al. 2004).

Over the years, among wildlife, birds are being used for monitoring the impact of environmental contaminants since the 1960s (Morrison and Gastol 1986; Koskimies 1989; Newton et al. 1993; Grasman et al. 1998; Van Wyk et al. 2001; Mochizuki et al. 2002; Rattner 2009 and Gómez-Ramírez et al. 2014). Roles of birds in tracing the origin of contaminants and for serving as indicators for evaluation were documented (Furness and Greenwood 2013).

Birds have the most diverse ecological functions among vertebrates (Holbrook et al. 2002). This is because birds are visible, widely distributed in the ecosystem, sensitive to toxins, high on the food chain, ecologically versatile, they live in all kinds of habitats and belong to various feeding guilds (Jarvinen and Vaisanen 1979; Jarvinen and Vaisanen 1983). Birds feeding on animals accumulate higher metal levels than those feeding on plants (Lindberg and Odsjo 1983; Gochfeld and Burger 1987) and thus they are particularly more exposed to metals. Birds have been ecologically well-researched and hence, can help in making well-founded ecotoxicological interpretations (Berghlund 2010).

Environmental pollution and exposure to toxic substances are a growing problem worldwide. The usage of metals and also pesticides has increased the pace of extinction of various species of amphibians, fish, reptilian, avian and mammalian groups in recent decades (Wang and Murphy 1982; Purdom et al. 1994; Catherine and Gloria 2000; Baillie et al. 2004). Gilyazov (1992) found that the density of typical forest birds fall up

to 80 % in a heavily polluted area around a copper-nickel smelter complex at Monchegorsk, Russia, in the early 1990s.

Waterfowl in the united states were reported dying from lead poisoning, caused by the ingestion of lead shotgun pellets as early as 1874 (Feierabend 1983). A high mortality of Magpie Goose was associated with the ingestion of lead pellet which had accumulated in the sediment at the Bool Lagoon Game Reserve, South Australia (Harper and Hindmarsh 1990). Food is likely the major sources for cadmium observed in waterfowls tissues (White and Finley 1978; Di Giulio and Scanlon 1984).

1.1 Sources of metals in the environment

Metals enter the environment from natural and anthropogenic sources. Natural phenomena such as weathering, earthquakes, volcanic eruptions, erosions, forest fires and dust contribute to metal pollution (Nriagu 1989; Fergusson 1990; Shallari et al. 1998 and Bradl 2002). Agriculture, industries and mining are primary the anthropogenic sources of metal pollution. Chromium in sodium chromate used to prevent leaf fall, copper and cadmium as fungicides and algicides and zinc phosphide as rodenticides are some of the metals that contaminate the agro ecosystem (Dogan et al. 2014; Shakoori et al. 2016), Zinc phosphate in the form of ammonium phosphate, zinc sulphate, and zinc oxide are used as fertilizers (Mortvedt and Gilkes 1993; Tamil Nadu Agricultural University Agritech 2016). Agricultural, Pharmaceutical, fertilizer, pesticide, automobile and paint industries particularly let out effluents in to the environment with many toxic metals (Pohanish 2002). While, Pb, Zn, Cd, Cu and Ni are the backbones of the world's metallurgical industries mining and smelting are the activities that have greatly altered natural cycles centuries. (Holland and Turekian 2010).

Metals are absorbed from the soil by primary receptors such as microbes, plants and macro invertebrates before transferred to organisms at higher level (Blakely et al. 2002). Sediments crucially store metals released into water bodies. Having sequestered metals, sediments can reflect the quality of the overlying water and the effects of anthropogenic emissions (Forstner 1990). Metal compounds or species and the environmental compartment they exist in determine the availability of metals to organisms inhabiting the ecosystem.

Once bound with soil, metals are persistent, and they cannot degrade further unlike complex organic pollutants. However, they can undergo various reversible changes including speciation depending on the chemical environment (Campbell 2007).

1.2 Transformation and magnification of metals in organisms

Magnification is a process through which pollutants are transferred from food to an organism resulting in higher concentrations compared with the source. As metals are a natural part of the environment, organisms, particularly those are subjected to naturally high levels of metal exposure, have developed mechanisms to sequester and excrete them. Problems arise when organisms are exposed to higher concentrations of metals than usual, and which they cannot excrete or detoxify rapidly to prevent potential damage. Some metals are transformed into their organic forms, such as methylmercury, organo-selenium and organo-tin, and they readily bioaccumulate similar to Persistent Organic Pollutants (POPs) (Mann et al. 2011).

Sometimes climate variability (wet, dry years) can also influence inputs and transport of trace metals in aquatic ecosystems, which may pose potential risks to aquatic organisms, particularly fish and man through food chain by metal bioaccumulation and biomagnifications (Kibria et al. 2013).

Many trace elements are essential for life functions (Mertz 1981), and plants and animals possess various mechanisms for acquiring of sufficient amounts of trace elements from their environment. The same mechanism can also facilitate the uptake of non-essential metals (Ballatori 2002; Zalups and Ahmad 2003). The uptake and retention of a metal by an organism is termed bioaccumulation. Bioaccumulation of essential as well as non-essential elements is dependent on both the chemical availability of the metals within the environment and the organism's capacity for uptake and subsequent excretion (Mann et al. 2011).

Human activities can lead to deposition of metals in high concentrations for which biomagnification is not required before symptoms of toxicity can be observed in wildlife, result in the direct poisoning of higher vertebrates with metals (Mann et al. 2011).

1.3 Impact of metals on birds

Birds being at the top of the food pyramid, can yield information over a large area around each sampling site, not only on bioavailability of contaminants but also their possible course of biomagnification in the food web (Battaglia et al. 2005; Perez - Lopez et al. 2008). Birds in many cases appear to be more sensitive to environmental contaminants than other vertebrates (Furness 1993). Waterbirds and raptors may serve as sentinels for natural and man-made toxicological problems in the environment (Koster et al. 1996).

High concentrations of metals can cause teratogenic, mutagenic and carcinogenic effects in organisms (Eisler 2000) including birds. Continued uptakes of food contaminated by metals are stressors to birds. Differences in metal accumulation among different species of birds are related to their foraging habitats (Burger 2002).

Internal tissues and soft tissues are broadly used as bio indicators by many researchers; one or several tissues of muscle, liver, kidney, spleen, heart, lung, as well as fat, blood, brain and bone are usually used to investigate the concentrations of metals in the environment. There are content and concentration differences of the same metal element in different tissues of a species, so it's essential to measure different tissues for the population level evaluation (wei Zhang and Zhang 2011).

Metal accumulation in birds is mostly related to metal species, its bioavailability and exposure pathways (Burger et al. 2008). Moreover, over a period of time it is proved that accumulation of metals in birds is influenced by a variety of parameters such as trophic position, taxonomic group, genetic variability, age, gender, size of bird and feather types (Burger and Gochfeld 2000a; 2000b; Dauwe et al. 2002; Burger et al. 2008; Burger and Gochfeld 2009; Zolfaghari et al. 2009; Lucia et al. 2010a; 2010b; Castro et al. 2011).

Synanthropic species, namely Rock Dove and House Crow are the best indicators to understand the level and impact of contamination in human habitations. Mourning Doves serve as prey for a variety of predators, (Mirarchi and Baskett 1994). Although Mourning Doves are relatively low on the food chain because they eat primarily seeds, they also pick up pollutants from the soil which they pickup along with food as grit to aid in digestion (Mirarchi and Baskett 1994). Information on contaminant levels in dove

tissues can be an indicator of exposure or health of the dove population themselves, as well as of the potential well-being of organisms that consume them such as raptors, predatory mammals, and also humans.

Seabirds are also often considered as bioindicators of elemental levels in marine environments due to their trophic positions and bioaccumulation capacities (Walsh 1990; Furness et al. 1993; Monteiro and Furness 1995; Gray 2002).

Regular monitoring of metal contamination in birds may not be possible, if birds were to be sacrificed. In environmental studies, feathers can be used to evaluate the ecological health of a local ecosystem, and can be used as a non-destructive bio-monitoring tool (Burger and Eichhorst 2007).

The concentrations of metals in feathers of birds have been documented worldwide over the years (Dauwe et al. 2005a; 2005b; Malik and Zeb 2009; Hashmi et al. 2013), and have also been shown to affect bird populations (Graganiello et al. 2001; Muralidharan et al. 2004). Bird feathers offer several advantages as bioindicators of metal exposure and feather collection is non-invasive. Concentration of metals is higher in feathers than in other tissues, and hence easier to detect and quantify. This is because birds excrete considerable amounts of metals through feather moult (Malik and Zeb 2009; Zamani-Ahmadmahmoodi et al. 2010). Moreover, feathers from newly born chicks indicate local contamination, derived mostly from food collected locally by their parents during the short period of egg formation and chick development (Boncompagni et al. 2003).

1.4 Incidences of metal poisoning

Not only birds, metals also affect other organisms including human beings. Itai-Itai disease, Minamata disease and lead poisoning caused by metals had a world-wide impact. First noticed in the year 1930 and identified as cadmium poisoning in 1960, Itai-Itai was traced to the cadmium discarded by the Kamioka mine in Japan. It led to bone weakness resulting in painful spines and legs apart from waddling gaits in human beings.

The liquid wastes of the Kamioka mine, owned by the Mitsui Mining and Smelting Company Limited (Mitsui Kinzoku), were eventually incriminated as the source of the cadmium. Kamioka was one of the richest zinc mines in Japan. Cadmium occurs as a minor component in most zinc ores and therefore is a byproduct of zinc production.

In June 1961, in his report to the head of the Fuchu-machi branch of the JMPPC, Yoshioka concluded that the cause of Itai-Itai disease was due to cadmium discarded by the Kamioka Mine (Yoshioka 1961; 1964).

Minamata disease is yet another disease that was caused by the release of methyl mercury along with the industrial wastewater from the Chisso Corporation's chemical factory, which continued from 1932 to 1968. In human beings, the most notable target of chronic exposure to elemental mercury is the Central Nervous System (CNS). This elicits neurological and psychological symptoms such as neurological degradation, erythrim (increased sensitivity), irritability, excessive shyness, and tremors which intensify and may become irreversible as the duration of the exposure and concentration increases (ATSDR 1999; USEPA 2001; Park and Zheng 2012).

Further, studies also documented wildlife especially birds being affected by methyl mercury. An environment contaminated by mercury is reported to have affected the survival and reproduction of birds such as American Kestrel, Osprey, Snowy Egret and Tricoloured Heron (Ljunggren 1968; Borg et al. 1970; Heinz et al. 2009; 2010).

In India, mercury poisoning has also been reported from Kodaikanal in Tamil Nadu. It originated from the waste dumped by a thermometer factory for 17 years. Around 2001, a number of workers at the factory began complaining of kidney related ailments. 290 tonnes of dumped mercury waste from the shola forest were sent back to the United States for recycling in 2003.

An analysis of water, sediment and fish from the Kodaikanal Lake by Department of Atomic Energy, Government of India showed an elevated mercury level even four years after the stoppage of emissions (Karunasagar et al. 2006). However, there are no studies on the impacts of mercury on birds in India.

Waterfowl, in the United States of America were reported to have died due to lead poisoning caused by lead shot gun pellets way back in 1874 (Feierabend 1983). Lead ammunition can act as secondary poison in wildlife that feed on hunted species with scavengers and predators being the most susceptible. Tavecchia et al. (2001) reported decreased survival of mallards *Anas platyrhynchos* from lead ingestion in France.

Other species which suffered due to lead poisoning include Griffon Vulture *Gyps fulvus*, (Mateo et al. 2003); Saltwater crocodile *Crocodylus porosus*, (Hammerton et al. 2003); Human Beings *Homo sapiens*, (Engstad 1932).

Generally, metal contaminants can bio - accumulate over time to reach sub - lethal or even lethal levels in organisms unless they are excreted or detoxified. This problem is particularly severe for metals in organisms that live long and are at the top of their food chains (Van Straalen 1991; Burger and Gochfeld 1992; Sundlof et al. 1994). Comprehensive studies to assess and understand the impact of metals on birds are limited in India. The present work is an effort to document the current status of metal contamination in select species of birds in the above referred perspective in India.

Literature Review

2. LITERATURE REVIEW

Over the past few decades, excessive deposition of metals in the environment and their negative impacts on ecosystems and organisms has attracted the attention of ecotoxicologists throughout the globe (Frantz et al. 2012). Industrialization resulted in increased toxic metal pollution in the environment (Ayangbenro and Babalola 2017). Masindi and Muedi (2018) stated that increasing metal concentration in the environment is a serious ecological problem which requires immediate attention. Most of the metals are global contaminants that biomagnify in food chains (Mathews and Fisher 2008) and are excreted in eggshells (Mora 2003), internal organs such as bird liver and kidneys (Braune and Gaskin 1987a; 1987b) and feathers (Veerle et al. 2004) via blood proteins.

2.1 Status of metal contamination in tissues of different species of birds

2.1.1. International context

World metal production in the 1970s was stable, but increased in the 1980s due to technological advances and increased use of metal based articles (Faust and Aly 1981). These pollutants, on entering aquatic systems, can persist in the environment and undergo environmental transformation into toxic substances. This affects plants, causes biomagnifications and bioaccumulation besides chronic and sub lethal effects to living organisms (Burger and Gochfeld 1985).

Metal accumulation in different bird tissues depends on the intensity and timing of exposure, forms and types of metals, feeding habits, growth or age, reproduction, moulting (Honda et al. 1986). Accumulation and magnification of metals in animal tissues have received considerable attention in recent years due to increased understanding of their lethal and sub-lethal effects (Ololade et al. 2008; Cardwell et al. 2013; Vadlamani et al. 2018).

Birds generally get exposed to environmental contaminants, including metals, through air, food and water. The contaminants accumulate in the tissues, get metabolized and get excreted from each organism. If the organism cannot eliminate the metal contaminant, it may reach dangerous concentrations in its body or in the food web (Drouillard 2008). Waders are particularly good bioindicators of environmental contamination (Kushlan 1993; Custer and Custer 2000; Bryan et al. 2001). They being

predatory, consume fish, insects, and annelids, and thus become susceptible to bioaccumulation of contaminants.

Until the mid 1980s, two to three per cent of the U.S waterfowl population was dying every year from lead poisoning (von Fuchs 1842). Poisoning was caused by the ingestion of lead shotgun pellets (USFWS 1986). Lead poisoning was first identified as a disease in wild birds as early as 1842 in Berlin, Germany (von Fuchs 1842). The first record of lead poisoning was reported in Tundra Swan by Grinnell (1894). It increased subsequently and was common in waterfowl and grew into a geographically widely distributed phenomenon (Phillips and Lincoln 1930; Shillinger and Cottam 1937; Bellrose 1959 and Mississippi Flyway Council Planning Committee 1965).

Patuxent Wildlife Research Centre's environmental contaminants programme incidentally unearthed the first case of lead poisoning in a Bald Eagle (Mulhern et al. 1970). Bagley et al. (1967) reported lead poisoning in Canada Geese in the USA. They noted that sick geese were usually emaciated and had just enough strength to make feeble escaping efforts, running a short distance in a marsh or by making short flights. Many of these geese showed marked cephalic edema with sub-mandibular swelling, edema in the eyelids and profuse discharge of clear to cloudy liquid from the eyes and nostrils. Their hearts were flabby and their gizzards were dark green with regurgitated bile. The lead level in the liver tissues of the geese ranged from 6 to 20 $\mu\text{g/g}$.

More than 15 countries reported lead poisoning in waterfowl. Thousands of birds in 1969, died after ingesting lead shot gun pellets, fishing weights or less common environmental lead from mining and smelting operations (Bellrose 1959; Simpson et al. 1979; USFWS 1986 and Blus et al. 1991; 1995).

Environmental pollution due to cadmium is worldwide. Cadmium is known to produce a broad spectrum of toxic effects in mammals and birds (Underwood 1971). It has unusual biological properties great accumulation tendency in kidney tissues of mammals and birds, low rates of excretion and consequently long biological half-life (Frank and Borg 1979). An experimental study by Mayack et al. (1981) in Wood Ducks showed that cadmium accumulation in liver tissues was probably the best measure for monitoring the total exposure or to understand the body burden of cadmium.

Hutton (1981) reported levels of cadmium, lead and zinc in select tissues of three species of sea birds. The study recorded positive correlation between zinc and cadmium in the kidney and between selenium and mercury in the kidney and liver. It suggested that these associations may reflect antagonistic interactions between zinc and cadmium toxicity as also selenium and mercury toxicity.

Nicholson and Osborn (1983) reported varying degrees of kidney damage in many species of birds in the U.K. These were associated with mean levels of 22 to 53 $\mu\text{g/g}$ of cadmium in the kidney. The study further stated that the kidney is the most appropriate tissue for studying chronic cadmium exposure. Di Giulio and Scanlon (1984) held that ducks accumulated high concentrations of non-essential metals such as cadmium.

Ohlendorf et al. (1985) studied the tissues which contained the highest concentration of metals and the relationship among tissues and metals in the Brown Pelican. Concentrations of chromium and zinc were most frequently correlated with other metals, and sometimes with each other. The concentrations in other tissues were most frequently related to those in the liver. Lee et al. (1987) studied the distribution of mercury, cadmium and lead in the tissues of 16 species of birds in Korea and related the metal levels to their feeding habits and habitats.

Although lead shots were banned in 1991, DeStefano et al. (1991) found higher exposure rates based on elevated blood lead concentrates ($>0.18 \mu\text{g/g}$) than on ingested shot in the gizzards of Canada Geese in nontoxic shot areas and those where lead was still used.

Wayland and Bollinger (1999) noted the prevalence of high lead exposure levels and lead poisoning reported in Bald Eagles and Golden Eagles from the Canadian Prairie Provinces from 1990 to 1996. Of the 127 Eagles investigated, 12 per cent were lead-poisoned based on lead concentrations of $>20 \mu\text{g/g}$ in the kidney or $>30 \mu\text{g/g}$ in the liver. Further 5 per cent of birds exhibited elevated lead concentrations based on kidney lead concentrations ($>8 \mu\text{g/g}$) or liver lead concentrations ($>6 \mu\text{g/g}$).

Hérons and Egrets are regarded as bioindicators of environmental health as they forage in aquatic environments on large fish and integrate exposure over time and space (Erwin and Custer 2000; Henny et al. 2002; Kim and Koo 2007)

The Bald Eagle suffered from toxicity and population decline due to secondary poisoning. Stauber et al. (2010) reported mortality of 28 out of 46 Bald Eagles which were admitted to the Washington State University's Raptor Rehabilitation Programme due to lead poisoning during 1991 to 2008. The situation slowly improved when lead shots were banned for hunting and steel pellets were introduced.

Burger and Gochfeld (2003) reported levels of arsenic, cadmium, chromium, lead, manganese, mercury and selenium in eggs of Common Terns nesting in five salt marsh islands in Barnegat Bay, New Jersey from 2000 to 2002. The levels of most of the metals were not high enough to cause harm except for those pertaining to mercury. The levels of mercury in some of the eggs were within a harmful range. Such levels were suspected to have reduced the local Common Tern population.

Lead, cadmium and zinc were estimated in Oklahoma, Kansas and Missouri, USA, where mining, milling and smelting were on. The study was to determine whether the habitat was contaminated to the extent of exposing wild birds to toxic concentration of metals (Beyer et al. 2004).

Concentrations of arsenic, lead, cadmium, chromium, zinc and copper were investigated in the muscles, intestines, kidneys, gizzards and livers of 60 Rock Pigeons from five locations in Bangladesh. The study revealed significant differences in metal concentrations among different tissues. But there were no location-wise differences. The study showed the liver to be the main metal storage site while muscles were the lowest (Begum et al. 2005).

Millaku et al. (2014) reported varying concentrations of metals, namely Cd, Pb, Ni and Zn in the lungs and hearts of European House Sparrows. The levels were significantly higher in birds from urban areas as compared to those from rural ones.

The levels of copper and zinc were measured in 18 Great Cormorants from Anzali and Gomishan wetlands in the south of the Caspian Sea. There were relatively high levels

of metals in different tissues. The study recommended the Great Cormorant as a suitable indicator for metal accumulation studies across different wetlands in all seasons (Aazami and KiaiMehr 2018).

Metal pollution was reported in the commercially valuable and edible. Rock Pigeon collected from five different locations in Bangladesh. This edible tissue from Rock Pigeon was considered to pose no health risks for human consumers with respect to these elements. The liver was the main metal storage tissue while muscle had the lowest levels of all analyzed metals. The results confirmed that pigeon muscle (Human consumption) and liver tissues were ideal models for identifying metallic contaminants. On the other hand, internal organs, such as liver and kidney of pigeon had generally unacceptable metal levels which might still be a health hazard for a long period of time (Begum and Sehrin 2013).

The metal levels in synanthropic organisms, namely House Sparrow, House Crow, Rock Pigeon (Johnston 2001) have been assessed in many countries such as Spain (Tejedor and Gonzalez 1992), France (Jenkins 1975; Scheifler et al. 2006), Korea (Nam and Lee 2006), Brazil (Brait and Antoniosi Filho 2011) and Finland (Kekkonen et al. 2012).

A common bird found in human areas is the House Crow which feeds on local wastes. It is therefore an important model species for ecological research, suitable for analyzing zinc, copper, nickel and lead besides being a cost-effective biomonitor for environmental pollution (Janaydeh et al. 2016).

Metal concentrations in omnivorous birds, including *corvids* in Japan have been rarely investigated. Among the few available studies, on *Japanese corvids* provided by Horai et al. (2007) levels of Zn and Cu concentrations in the liver tissues of the Jungle Crow were studied. Orłowski et al. (2013) studied accumulation of Cu, Zn and Pb in the liver tissues of omnivorous *Corvus frugilegus*. Hooded Crows have been used as bioindicators for environmental contaminations by toxic metals in Italy (Giammarino et al. 2014).

2.1.2. Indian context

In India, studies to document metal levels in birds are far and few. To name a few, the study by Husain and Kaphalia (1990) studied metal contamination in a few species in Lucknow. They observed high concentrations of Cd (0.2 µg/g) in liver tissues of Black Kite, Mn (2.7 µg/g) in kidney tissues of Rock Pigeon and Pb (4.75 µg/g) in the House Crow's brain.

Lead contamination was studied in House Sparrows in Jaipur by Bakre and Sharma (1995). They analyzed lead concentrations in bone, brain, heart, intestine, kidney, liver, lung, muscle and stomach. Bone lead levels appeared to have provided reasonably realistic background values for lead in Jaipur city in India.

Muralidharan (1995) documented metal contamination in 16 species of birds in the Keoladeo National Park, Bharatpur. He studied altogether 44 dead birds comprising 16 species during 1988 to 1990. He analyzed cadmium, lead, chromium, copper and zinc in different tissues. The study found the wetland ecosystem's quality to greatly influenced the accumulation of metals in the tissues as the birds depended on the park's aquatic ecosystem and the immediate surroundings for food. While the kidney indicated the contamination of cadmium, lead and zinc the least, the liver indicated copper concentrations.

Kler et al. (2014) documented metal levels in the excreta of the House Crow, Common Myna, Rock Dove and Cattle Egret collected from agricultural fields in Ludhiana district, Punjab. The significant variations in metal levels among the species showed that there were similar fluctuations in the contamination levels of the environment where they fed, roosted and bred. They also suggested the use of excreta as a contamination indicator in wild birds without harming them.

Rajamani and Subramanian (2015) documented metals in the critically endangered Indian White-rumped Vulture in Gujarat, Tamil Nadu and Punjab. Metals were found to differ in their potential for accumulating in the tissues of the birds.

Pannu and Kler (2018) studied the House Sparrow in two villages of Ludhiana district. They found that cadmium, chromium, nickel and zinc levels were above normal in the excreta from both the villages. The cadmium concentrations were particularly

above toxic limits range (0.02 to 1.5 $\mu\text{g/g}$) in Alamgir village. Various agrochemicals used in the fields were likely to have contaminated the food chain of the House Sparrow at different trophic levels. Presence of metals in excreta of House Sparrow has signified the contamination in its preferred food items like grains and invertebrates in agricultural habitat of Ludhiana district.

2.2 Feather as a tool to assess metal contamination in birds

2.2.1. International context

As birds excrete substantial quantity of metals through feather moult, quantifying them become easy (Malik and Zeb 2009; Zamani- Ahmadmah moodi et al. 2010). Several studies also demonstrated a strong correlation between metal concentration in feathers and internal tissues in birds, which suggests that breast feathers are ideal samples for the determination of metal concentration in birds (Gochfeld 1980; Heinz 1979; Goede and Bruine 1986; Dmowski 1999; Ranta et al. 1978; Markowski et al. 2013; Janaydeh et al. 2016; Baker et al. 2017).

Metal levels in feathers of Great Egret were examined from several geographical areas, including the Everglades in Florida (Rumbold et al. 2001; Rumbold and Fink 2006; Herring et al. 2009), Georgia and South Carolina (Bryan et al. 2012), and Korea (Honda et al. 1986; Kim et al. 2009). Heinz (1976) studied mercury levels in feathers of birds. These studies stated that the effects can be explained only for short term exposure but cannot examine long - term impact due to the contamination of the water bodies where they forage.

Burger (1996) examined metals concentrations in breast feathers of adult and fledglings of Franklin's Gulls (*Larus pipixcan*) collected from North Western Minnesota, Eastern South Dakota, Eastern North Dakota and Central Montana. Metal levels in the feathers of the gulls were within the range reported for other species. It also reported difference among locations in levels of metals between the adults and young ones. Out of 24, 19 species showed age related differences in metal levels. Adults significantly had high levels of all the metals tested except, selenium, which is high concentrations in young ones of gulls.

Feather samples of six species of birds collected from Southern Finland were studied for metal contamination by Solonen and Lodenius (1990). Feathers of Common Myna recorded the maximum zinc concentration (153.83 $\mu\text{g/g}$) when compared with other species. Although the results revealed the variation of metal levels in different species, no uniform pattern could be seen except for zinc. Of all the metal, zinc concentration was the highest followed by copper and lead in all the feathers. In the secondary feathers, copper and zinc exhibited the maximum levels of 9.4 and 157.47 $\mu\text{g/g}$ respectively. On the whole the tail feathers ranked first in metal concentration having a uniformly high level of accumulation. Of all the metals, zinc recorded uniformly high levels irrespective of feather type as well as species (100.97 to 188.90 $\mu\text{g/g}$).

Dmowski (1999) examined a few species of birds collected from urban and industrialized areas for assessing exposure to metals in Poland. The distribution pattern of copper was apparently uniform among the primary, secondary and tail feathers of the Cattle Egret and Indian Pond Heron. Nevertheless, the tail feathers of Cattle Egret and Indian Pond Heron recorded high level of copper 219.93 and 235.43 $\mu\text{g/g}$ respectively.

Dauwe et al. (2005b) reported the relation between concentrations of metals in the internal organs and feathers of breeding female Great Tits *Parus major* and concentration in their eggs. Metal concentrations in eggs and eggshells were in most cases much lower than in internal organs. Kidneys were poorly correlated with feathers while cadmium and lead from breast feather were correlated with liver.

Six metals (Cd, Pb, Cr, Ni, Cu and Zn) were assayed in feathers of White Storks collected from Central Poland. Chromium 0.31 $\mu\text{g/g}$, cadmium 0.403 $\mu\text{g/g}$ was recorded in fledglings of White Storks. Their average concentrations were cadmium: 0.049 $\mu\text{g/g}$; nickel: 0.85 $\mu\text{g/g}$; lead: 1.75 $\mu\text{g/g}$; copper: 14.74 $\mu\text{g/g}$ and zinc: 199.9 $\mu\text{g/g}$. While White Stork populations from Central Europe spend only 5 months in their breeding grounds, the levels of lead, copper, nickel, and zinc in feathers of this species are comparable to those in feathers of sedentary European birds and of birds wintering in Europe (Orłowski et al. 2006).

Burger and Gochfeld (2007) investigated concentrations of cadmium, lead and chromium in the down feathers and eggs of Common Eiders *Somateria mollissima* from

Amchitka and Kiska Islands of Alaska to find out whether there were differences between concentrations of cadmium, lead and chromium in down feathers and eggs. Mean levels in eggs were as follow: cadmium (1.49 ng/g), chromium 414 ng/g and lead 306 ng/g. There were no significant correlations between the levels of any metal in down feathers of females and their eggs.

Breast feathers were used to estimate mercury concentrations in birds, namely the Sooty Tern, Brown Noddy, Lesser Noddy, Audubon Shearwater, Barau's Petrel and White-tailed Tropicbird which were nesting in the tropical western Indian Ocean (Kojadinovic et al. 2007). Juveniles consistently had lower mercury (0.39 $\mu\text{g/g}$) than adults. The highest levels were recorded in adult Barau's Petrels from Reunion Island (0.96 $\mu\text{g/g}$). An inter-site analysis of Sooty Tern showed higher mercury levels (0.21 $\mu\text{g/g}$) in nesting birds on Juan de Nova Island. Levels were low in comparison with values reported in the plumage of seabirds worldwide.

Burger and Gohfeld (2009) analyzed arsenic, cadmium, chromium, lead, manganese, mercury and selenium in feathers of Common Eiders and Tufted Puffins from Amchitka and Kiska Islands. Between species, puffins had 10 times higher chromium. Puffins are higher in food chain than Eiders and hence resulted in higher levels of metals in their feathers. The relatively low levels of metals neither indicate the potential for adverse behavioral or reproductive effects in the birds themselves, nor pose concern for other consumers, including subsistence hunters.

Hofer et al. (2010) used passerines as bioindicators of metal contamination. Study site was approximately 100 ha brown field wild area of Liberty State Park located in New Jersey City. In this study they tested feathers of nestlings of two common bird species (House Wren and American Robin) to understand accumulation pattern of Pb, Cr, Cu and Zn between a polluted urban field and a rural unpolluted site. At the polluted site, they monitored nests of 21 House Wren and 45 nests at the control site. In the test site Liberty State Park 19 nests were considered successful (fledged 1 nestling) out of 21, while only one nest was lost to predation and then abandoned. In contrast, at control site there were only nine successful nests out of 45 nests that had at least one egg. House Wrens accumulated higher concentrations of Pb (119.5 ppb), Fe (144.8 ppb) and Zn (3.7 ppb) than American Robins.

Recent studies have investigated the levels of metals in feathers of feral pigeons in many countries, including Korea (Nam and Lee 2006), Israel (Adout et al. 2007) and Brazil (Brait and Antoniosi Filho 2011).

Feathers were sampled from 91 pigeons caught with trap cages in France. The concentrations of 4 metals, namely cadmium, copper, lead and zinc in pigeon feathers from 7 sites were analyzed. Elements were detected in all individuals of pigeons, indicating that metals present in urbanized areas strongly depended on the origin of individuals. Overall, Frantz et al. (2012) suggests that urban feral pigeons may represent a good model system for metal biomonitoring.

The concentrations of metals (Cd, Pb, Ni, Cu and Zn) in feather samples of raptors in three families were determined from southern, central, and northern parts of Punjab Province (Nighat et al. 2013) in Pakistan. Deposition of metals significantly varied among the raptor families. Falconidae deposited the highest concentrations of metals and minimum was observed in Strigidae family. While feathers from birds of central Punjab appeared to have more metal loads, lowest levels of metal contamination was documented in southern Punjab. The concentrations of metals were reported to be alarming.

Narjes (2013) reported concentrations of metals (Cd, Pb, Cu and Zn) in tail feathers of 57 birds in Shadegan wetland in southwest Iran. Concentrations of metals were studied in relation to trophic levels. In this study, bird species were classified into five groups based on their diet habitat, namely herbivorous, omnivorous, invertebrate predators, piscivorous and carnivorous. They found a significant difference between Cd concentration across carnivores and omnivores with other metals, namely Cd, Pb, Cu and Zn. Moreover the results showed significant difference between Cu concentration in feathers of herbivores and other trophic levels. But there was no significant difference for Zn among the trophic levels. From this study, it is observed that the carnivorous and omnivorous species are useful bioindicators for Cd and herbivorous birds for Pb pollution in Shadegan wetland.

Levels of cadmium, chromium and lead in feathers of Great Egret fledglings collected at nesting colonies in Baretgat Bay, New Jersey from 1989 to 2011 were

examined. There were temporal differences in metal levels in feathers of fledglings of Great Egret (Burger 2013).

Abbasi et al. (2015) studied metals in feathers of 48 bird species from Pakistan. Concentrations of metals were compared among different taxonomic groups feeding guilds and widely stretched geographical locations. Inter and intra-specific variations in metal accumulation in different bird species were also reported. They observed taxonomically close species to resemble in terms of metal concentrations in their feathers compared to distantly related species, inferring similar metal accumulation in genetically related organisms. Results from this study also confirmed that metal concentrations are directly associated with the feeding guild. They recommended that the preen gland also alter the metal contamination in feathers, so preen gland should be taken into account while analyzing metal concentrations in feathers.

Zinc, copper, lead and cadmium were measured in feathers of Eurasian Magpie *Pica pica* from urban area of Central Iran in 2013. Correlations between the concentrations of Cu and Zn as well as Pb and Cd were significant. Moreover wing length had the highest correlation with metal concentrations. There were significant differences among genders, ages and size. Pb and Cd contamination was as high as in the industrial complex areas. Authors suggested that in biomonitoring studies combination of effective factors including sex, age, physiological and morphological characteristics and sampling sites should be considered (Zarrintab et al. 2016).

2.2.2. Indian context

A thorough literature survey revealing that there are only few studies available on the levels of metals in feathers of birds in India.

Feathers of 39 birds comprising six species of birds were studied in Nilgiri district, India. Indian Pond Heron had the lowest concentrations of zinc (100.97 µg/g) in primary feathers. The Little Egret had the maximum (196.32 µg/g) in tail feathers. Lead concentrations were below detection limit in secondary feathers of Jungle Babbler and Common Myna. It was suggested that tail feathers could be considered as appropriate indicators of metal contaminations. However the magnitude of contamination could be influenced by several exogenous and endogenous factors (Muralidharan et al. 2004).

Eleven species of birds collected from urban and rural areas of Thiruchirapalli, Tamil Nadu were analyzed for metals, namely iron, chromium, nickel, copper, zinc and cadmium (Manjula et al. 2015)

In India, although, the metal pollution has not created any major episode, the problem exists in varying magnitude in various places. But it is not explored in its totality anywhere as discussed here. The present investigation will document the magnitude of contamination of metals, in birds from states namely Gujarat, Assam, Tamil Nadu and Kerala, India.

Objectives

3. OBJECTIVES

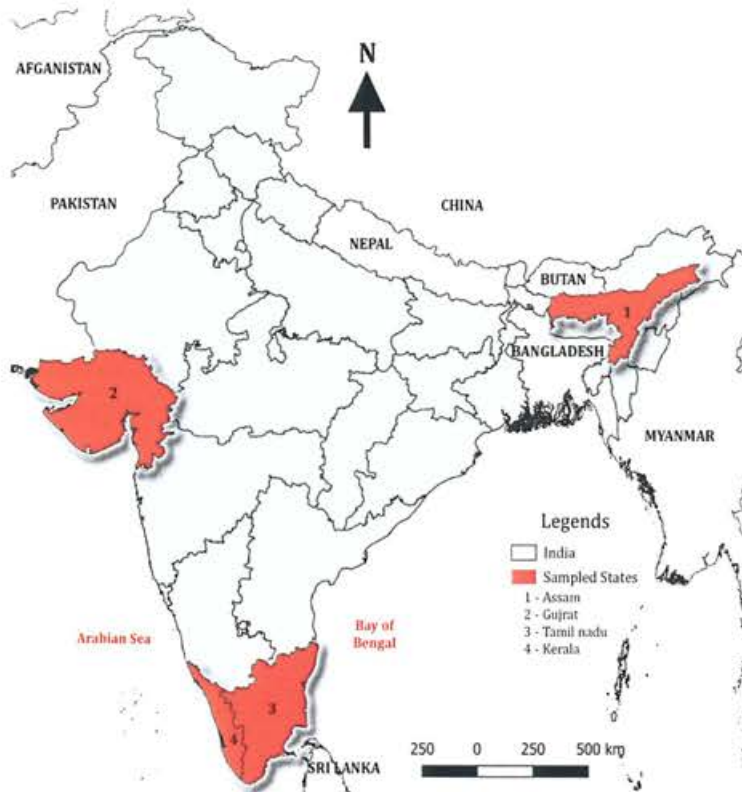
1. Document the levels of metals, namely cadmium (Cd), lead (Pb), chromium (Cr), nickel (Ni), copper (Cu) and zinc (Zn) in tissues of different species of birds in select states in India, and generate a data base.
2. Understand the variation in accumulation pattern of metals among different types of feathers, and check if feathers could be used as a tool to understand metal contamination.
3. Check if metals were responsible for mortality birds during the study period.

Methodology

4. METHODOLOGY

4.1 Study Area

The study was conducted in 4 states, namely Gujarat, Assam, Tamil Nadu and Kerala (Figure 4.1) in India.



Note: This map is not purported to be a political map of India

Figure 4.1 India Map showing the states that have been included in the present study

4.1.1. Assam

Assam is a state ($26^{\circ} 14' 38.9616''$ N and $92^{\circ} 32' 16.2312''$ E) in north-eastern India, situated south of the eastern Himalayas along the Brahmaputra and Barak river valley; Assam covers an area of $78,438 \text{ km}^2$. The state is bordered by Bhutan and Arunachal Pradesh to the North; Nagaland and Manipur to the East; Meghalaya, Tripura, Mizoram and Bangladesh to the South; and West Bengal to the West via the Siliguri Corridor. A 22 - kilometer strip of land connects the state to the rest of India.

Totally, there are 110 industries in Assam. 15- cement; 9 - steels and bars; 5- paper mills; 4-metal alloys; 2- aluminum; 1- cosmetics; 1- crackers; 1- carbon; 3-wires and

cables; 2- fertilizer; 2- mosquito repellent; remaining are food and oil industries. Lead, iron, and arsenic levels have been reported to be elevated in groundwater in 21 districts out of 24 districts of Assam (Singh 2004; Chetia et al. 2008; Borah et al. 2009).

4.1.2. Gujarat

Gujarat (20° 01'N 74° 24' E), situated in western India, has diverse habitats and a varied birdlife. The main regions of Gujarat are Kutch, Saurashtra and mainland Gujarat (which includes South Gujarat, North Gujarat and the area in between, which can be referred to as Central Gujarat). These regions have varied habitats, with moist, deciduous forests of South Gujarat to the deserts of Kutch. There are four National Parks, twenty - three sanctuaries, one conservation reserve in the state. Gujarat has only 11.04% of its geographical area declared as forest, which is much below the national average. Forest and tree cover is in the extent of 11.74 % (7.48 % forest cover and 4.26% tree cover outside forest) of its geographical area. The unique features of the state are the climatic and geomorphologic conditions, the largest coastline in the country, the saline deserts of Rann, grasslands and wetlands. These factors have bestowed the state with the diversity of flora and fauna. The majestic Asiatic Lion and Wild Ass have their last resorts of the world in Gujarat. The faunal biodiversity consists of 14% fishes, 18 % reptiles, 37% avi fauna and 25% of the mammal populations of the country. Hence, there is great diversity in the bird life seen here. The varied habitats, along with a culture of non-violence towards wildlife, have made Gujarat a premier destination for bird watching in India. Gujarat had 611 of bird species (Lepage 2019) in different types of habitats including deciduous and thorn forests, grasslands, wetlands, marine intertidal areas, scrublands and saline deserts (Singh 2001).

Out of 27,892 industries registered under Gujarat Pollution Control Board till 2011-12, more than 60 % of industries fall under Red Category. Totally there are 1240 metal based industries in the state. Out of 1240, 653 industries under Red Category; 166- Pharmaceutical industry; 38- copper; 13 – paints and varnish; 7-chromium; 4- fertilizers; 3-cadmium and 1- electrochemical and electrolytic plant-based industry. Estuarine areas are being polluted due to the high input of domestic sewage being released from Ahmedabad city. 77% of mining areas; 61% of total quarries; more than 90 % *Special*

Economic Zone (SEZ) and over 80 % of Sir Creek is located in the coastal region. Industries are located in and around river bank of Sabarmati (Vediya and Patel 2011a; 2011b). Hence, metal contamination is highest in Gujarat when compared to other states (CPCB 2011).

A unique event in Ahmedabad is the kite fling festival during Uttarayan. People celebrate the sport of flying kites around 14th January, Makar Sankranti, every year when summer replaces winter. Unfortunately, thousands of birds fall victims to kite flying during this period.

4.1.3. Kerala

Kerala is situated between 10° 51' 1.8576" N and 76° 16' 15.8880" E along the windward side of the Western Ghats of Indian Peninsula. The state is located in the tropical region of Indian Peninsula and extends to an area of about 38863 km², which accounts for about 1 to 2 per cent of the total geographical area of the country. The state is bordered by Tamil Nadu on its south and part of the East, Karnataka on the North and part of the east, the Lakshadweep Sea on the west and the Indian Ocean along the south. The state has a total coastline of about 560 km and from sea level; it rises to about 2694 meter above sea level (SL).

The Western Ghats region of Kerala covering nearly 21856 sq km or 56% of the total geographical area of the state and 42.7% of the entire Western Ghats region is the watershed of all the 44 rivers that sustain the agro-economy of the state. Praveen et al. (2018) compiled a list of 524 species of birds occurring in the state of Kerala. Out of 524, sixteen species are endemic to the state.

There are 29 metal producing industries (coils, iron, steel, bars ferroalloys, light metals, metal ash, non - ferrous metals, plates, raw metals, aluminum, and its alloys) in the state. Most of the industrial and commercial establishments in Kerala are concentrated in the coastal zone. Among the coastal districts, Ernakulam and Trivandrum have a fairly large number of industries along the coast, followed by Alappuzha, Kollam, Kozhikode, Kannur, Kasargod, and Malappuram. Eloor- Edayar- Ambalamugal area are the major industrial area located in the coastal zone in the city of Kochi in Ernakulam District along the banks of the Cochin backwaters.

The rivers of Kerala have been increasingly polluted from the industrial and domestic waste and pesticides and fertilizer from agriculture. Industries discharge hazardous pollutants like phosphates, sulfides, ammonia, fluorides, metals and insecticides into the downstream reaches of the river. The river Periyar and Chaliyar are very good examples of pollution due to industrial effluents. It is estimated that nearly 260 million liters of trade effluents reach the Periyar estuary daily from the Kochi industrial belt (ENVIS 2019).

There are local level quality problems faced by all rivers especially due to the dumping of solid waste, bathing, and discharge of effluents. With regard to groundwater, water quality characteristics of wells in Kerala are found to be affected by chemical and biological contaminants (ENVIS 2019).

Not only water, the soil also has turned into dumping grounds of most of the waste products-domestic, human, animal, industrial and agricultural. Every year the solid wastes dumped into the soil are increasing at an alarming rate in the state. The main sources of soil pollution include industrial wastes, urban and domestic waste, radioactive pollutants, agrochemicals, chemical and metallic pollutants, biological agents, acid rain (ENVIS 2019).

4.1.4. Tamil Nadu

Tamil Nadu is situated between 11.1271° 3' 35.3556" N and 78° 23' 14.8236" E. The Protected Areas in the state includes 5 National parks, 15 Wildlife Sanctuaries, 15 Bird Sanctuaries and two Conservation Reserves besides four Tiger Reserves *viz.* Anamalai, Kalakkad - Mundanthurai, Mudumalai and Sathyamangalam. Further, Tamil Nadu also has the distinction of having three Biosphere Reserves, namely Nilgiris, Gulf of Mannar and Agasthiarmalai, which are internationally acclaimed for their rich and unique biodiversity. The Western Ghats are one of the 25 global biodiversity hotspots and one of the three mega centers of endemism in India. The genetic assemblage of Tiger Reserves, Mukurthi, Srivilliputhur, Kanniyakumari and Megamalai owe their richness in flora and fauna to their locations in Western Ghats. The Biodiversity Conservation and Rural Livelihood Improvement Project pursued at Kalakkad-Mundanthurai Tiger Reserve is a new endeavor in perusing conservation at landscape level, covering five districts, namely Kanniyakumari, Tirunelveli, Virudhunagar, Madurai and Theni.

Around 454 species of birds are reported in Tamil Nadu. Out of 454 species of birds 17 species are endemic and 42 species are in schedule – I (ENVIS 2019).

In Tamil Nadu there are 110 industries which fall under 17 categories. tanneries - 9; aluminium -1; pharmaceutical -25; cement - 24; fertilizer -5; pesticide -2; pulp and paper -3; petro chemicals 10; copper sterlite -1; dyeing 1; metal based mercury industry in Kodaikanal (CPCB 2011); Iron and Steel -1 (Ministry of Micro Small and Medium Enterprises 2015).

These industries have been contributing to water, soil and air pollution for years in the state. Further Tuticorin waters are affected by industrialization since past few years (Easterson, 1998; Murugan and Edward, 2000). Cd and Pb pollution in Gulf of Mannar and Palk Bay, Southeast coast of India (Palanichamy and Rajendran 2000) and also Cu and Zn from the Gulf of Mannar, Bay of Bengal (Ganesan et al. 1991) were reported. Further, Pushpam et al. (2014) documented metals to be above the permissible levels in drinking water from temples in Kanyakumari district.

4.2. Collection of samples

Totally 197 individuals comprising 31 species of birds were collected (Table 4.1; Plate 4.1 and 4.2) from four states of India, namely Assam, Gujarat, Tamil Nadu and Kerala.

Table 4.1 List of bird species included in this study

S. No.	Name of the species	Scientific Name	IUCN status	Feeding Guild	Individuals
1.	Asian Koel	<i>Eudynamis scolopacea</i>	Least Concern	Omnivorous	04
2.	Bar-headed Goose	<i>Anser indicus</i>	Least Concern	Herbivorous	03
3.	Barn Owl	<i>Tyto alba</i>	Least Concern	Carnivorous	04
4.	Black Drongo	<i>Dicrurus macrocercus</i>	Least Concern	Insectivorous	03
5.	Black Kite	<i>Milvus migrans</i>	Least Concern	Carnivorous	14
6.	Blue-faced Malkoha	<i>Phaenicophaeus viridirostris</i>	Least Concern	Omnivorous	05
7.	Cattle Egret	<i>Bubulcus ibis</i>	Least Concern	Insectivorous	20

S. No.	Name of the species	Scientific Name	IUCN status	Feeding Guild	Individuals
8.	Common Coot	<i>Fulica atra</i>	Least Concern	Herbivorous	08
9.	Common Kingfisher	<i>Alcedo atthis</i>	Least Concern	Piscivorous	02
10.	Demoiselle Crane	<i>Grus virgo</i>	Least Concern	Omnivorous	03
11.	Emerald Dove	<i>Chalcophaps indica</i>	Least Concern	Granivorous	01
12.	Eurasian Collared Dove	<i>Strepto peliadecaoccto</i>	Least Concern	Granivorous	16
13.	Great White Pelican	<i>Pelecanus monocrotalus</i>	Least Concern	Piscivorous	01
14.	Griffon Vulture	<i>Gyps fulvus</i>	Least Concern	Carnivorous	03
15.	Himalayan Vulture	<i>Gyps himalayensis</i>	Near Threatened	Carnivorous	12
16.	House Crow	<i>Corvus splendens</i>	Least Concern	Omnivorous	11
17.	Indian Black Ibis	<i>Pseudibis papillosa</i>	Least Concern	Carnivorous	06
18.	Indian Peafowl	<i>Pavo cristatus</i>	Least Concern	Omnivorous	05
19.	Indian Pitta	<i>Pitta brachyuran</i>	Least Concern	Insectivorous	01
20.	Indian Pond Heron	<i>Ardeo lagrayii</i>	Least Concern	Carnivorous	13
21.	Indian Roller	<i>Coracias benghalensis</i>	Least Concern	Insectivorous	01
22.	Indian Spotted Eagle	<i>Clanga hastata</i>	Vulnerable	Carnivorous	01
23.	Indian White-rumped Vulture	<i>Gyps africanus</i>	Vulnerable	Carnivorous	13
24.	Painted Stork	<i>Mycteria leucocephala</i>	Near Threatened	Piscivorous	04
25.	Red-wattled Lapwing	<i>Vanellus indicus</i>	Least Concern	Insectivorous	05
26.	Rock Pigeon	<i>Columba livia</i>	Least Concern	Granivorous	20
27.	Rose-ringed Parakeet	<i>Psittacula krameri</i>	Least Concern	Frugivorous	06
28.	Sarus Crane	<i>Antigone antigone</i>	Vulnerable	Omnivorous	03
29.	Shikra	<i>Accipiter badius</i>	Least Concern	Carnivorous	04
30.	Slender-billed Vulture	<i>Gyps tenuirostris</i>	Critically Endangered	Carnivorous	02
31.	Spot-billed Pelican	<i>Pelecanus philippensis</i>	Near Threatened	Piscivorous	03



1) Rock Pigeon



2) Rose-ringed Parakeet



3) Indian Black Ibis



4) Indian Peafowl



5) Painted Stork



6) Common Kingfisher

Plate 4.1 Notable species collected dead from select states in India during 2012 - 2014



1) Indian Pond Heron



2) Asian Koel



3) Barn Owl



4) Demoiselle Crane



5) Black Kite



6) Indian White-rumped Vulture

**Plate 4.2 Notable species collected dead from select states in India during
2012 - 2014**

4.2.a Post mortem and transportation of tissue samples

Samples of dead bird were collected opportunistically by networking with forest officials, bird-watchers, volunteers and public from road kills, electrocution and other accidental deaths. Field visits were organized to collect samples of dead birds during mass mortality due to any incidence of poisoning, diseases, kite flying and any other unknown reasons. While collecting samples, evidences and other supporting data on circumstantial sources of contamination such as water runoff, crops cultivation in nearby farms, type of fertilizers and pesticides used were collected. External injuries on birds, if any were also observed and recorded.

In Gujarat, samples were also collected during “Save Bird Campaign” organized as part of kite festival in Gujarat during January every year from 2010 to 2014. Towards saving birds, a team of researchers from SACON (veterinarians and biologists) supported the Gujarat State Forest Department and Jivdaya Charitable Trust (JCT), Ahmedabad; an NGO actively working to save birds those got injured due to kite strings. On the whole, a group of 30 veterinarians and around 10 biologists from different parts of the country, and hundreds of volunteers from Ahmedabad participated in the “Save Bird Campaign” organized between 2010 and 2014. Treatment such as wound dressing and suturing was done for the birds that got injured by kite strings. Around 2000 birds comprising 60 species were estimated to have died during the referred period. Notable species were Indian White-backed Vulture, Demoiselle Crane, White Ibis, Barn Owl, Indian Cuckoo, Shikra, Rosy Starling and Red-Wattled Lapwing. We used this opportunity to measure the background levels of contaminants. While SACON’s veterinarians and scholars tried to save as many birds as possible, birds that did not survive were subjected to postmortem examination and tissues were stored over ice and brought by air to the Ecotoxicology laboratory at SACON, Coimbatore for contaminant analyses.

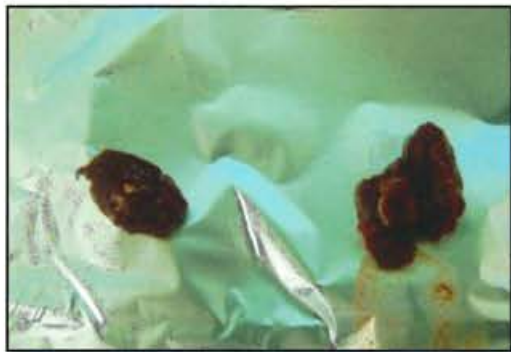
Post-mortem examinations were conducted either in the field or at SACON laboratory (Plate 4.3). During post-mortem, status of the internal organs and gizzard contents were observed. Tissues such as muscle, liver, kidney, and feathers were collected (Plate 4.4), wrapped in aluminum foil, labelled and packed such that there would be no leakage during transportation and storage. All these tissues were transported to SACON over ice, and preserved at -20°C until chemical analyses.



Plate 4.3 Postmortem examination of Black Kite



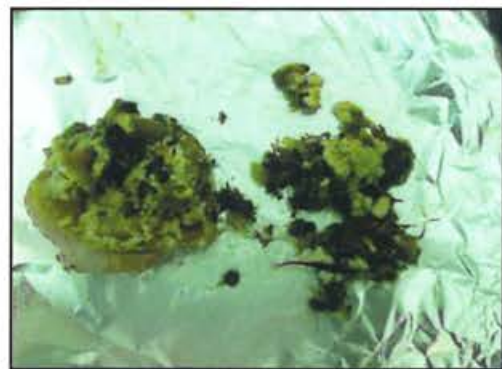
Muscle



Liver

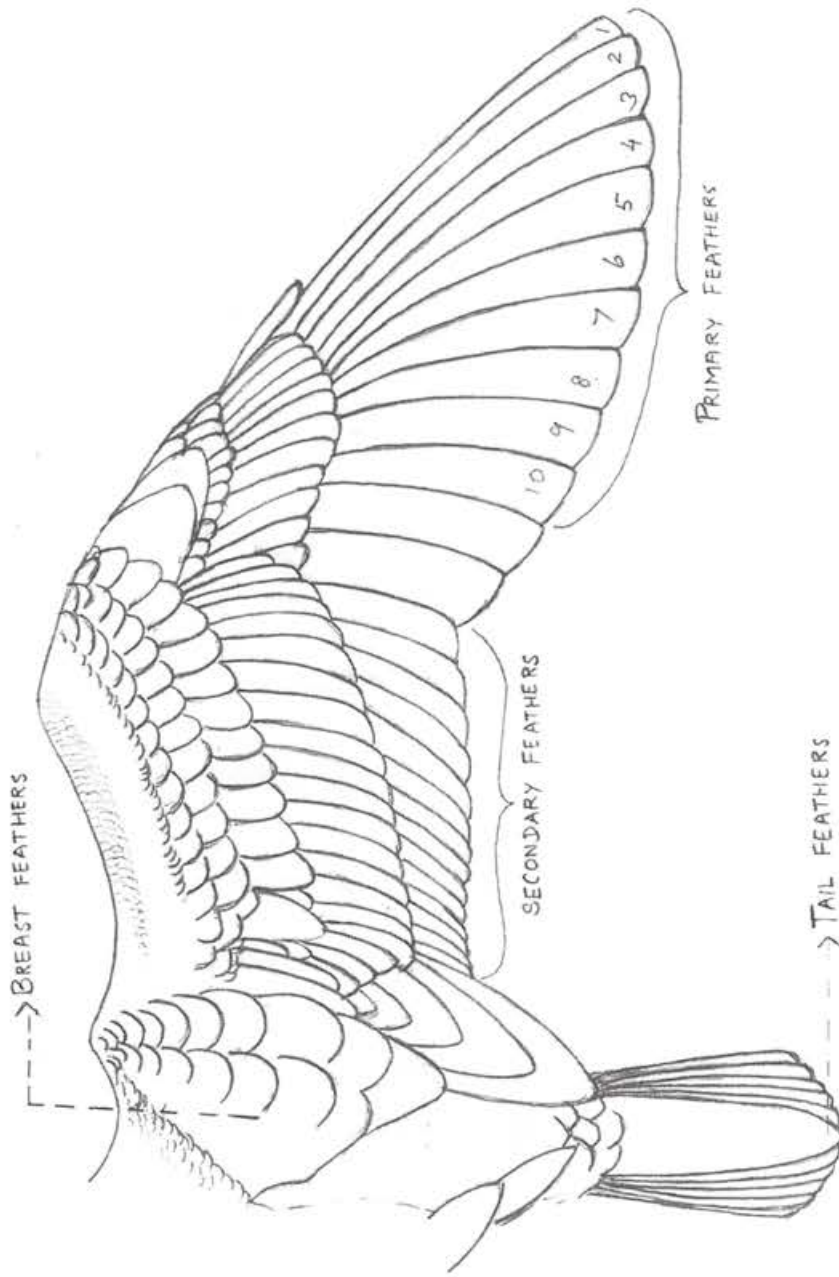


Kidney



Gut content

Plate 4.4 Tissue/gizzard dissected out for further analysis



Source: Book - Birds of the Indian Sub continent, Edition 2014.

Plate 4.5 Different type of feather I bird

4.2.b Collection and transportation of feather samples

Breast feathers, from the breast region, primary feathers from the first 10 feathers from the edge of flight feathers, continued by secondary feathers (next to 10 primary feathers) and tail feathers from tail region were obtained (Plate 4.5). As ambient temperature is good enough to store and transport the samples, all the feathers were packed in clean polyethylene bags separately, labeled appropriately and transported to the lab at the division of Ecotoxicology, SACON.

4.3. Sample processing

4.3.a Digestion of tissue samples

Samples were processed by using triple acid digestion method (Tam and Yao 1999). For digestion, about 1-1.5 g of tissue samples were weighed with the aid of a top loading balance (Mettler AE 240) and digested in Microwave Digestion System (Milestone, MLS 1200) using 10 mL Nitric Acid (HNO₃) (69% GR Merck) for 10 minutes, 1 mL Perchloric Acid (HClO₄) (70% GR - Merck) for 5 minutes and 5 mL Hydrogen Peroxide (H₂O₂) (30% GR Merck) for 10 minutes at 250 Volt magnetron power settings sequentially. These are the most effective acids used in the digestion of tissue matrices and determination of minerals in the detectable form. A blank without sample was prepared as mentioned above. The digested solutions were filtered using Whatman No.1 filter paper, made up to 25 mL using 10 per cent acid water and kept in 4° Celsius until analysis (Plate 4.6).

4.3.b. Digestion of feather samples

Before digestion all the collected feathers were cleaned with running tap water, distilled water, and Millipore water thoroughly to remove external dirt and stains. After complete drying under room temperature, they were washed with acetone to remove external contamination (Gochfeld et al. 1996) and stored in clean polyethylene bags for further digestion.

Triple Acid Digestion method as adopted by Muralidharan et al. (2004) was carried out. About 1-1.5 g of feather samples were weighed with the aid of a top loading balance (Mettler AE 240) and digested in Microwave Digestion System (Milestone, MLS 1200)

Digestion stages of analysis



Addition of acids to the samples



Microwave digester



Filtration of digested samples through filter paper



Sample analysis in AAS

Plates 4.6 Triple acid Digestion process

using 10 mL Nitric Acid (HNO₃) (69% GR Merck) for 10 min, 1 mL Perchloric Acid (HClO₄) (70% GR - Merck) for 5 min and 7 mL Hydrogen Peroxide (H₂O₂) (30% GR Merck) for 10 min at 250 Volt magnetron power settings sequentially. These are the most effective acids used in the digestion of feathers and determination of minerals in the detectable form. A blank without sample was prepared as mentioned above. The digested solutions were filtered using Whatman No.1 filter paper, made up to 25mL of 10 per cent acid water water and kept in 4° Celsius until analysis for later use (Plate 4.6).

4.4 Instrumentation

Digested samples were analyzed using a double beam Atomic Absorption Spectrophotometer (Perkin Elmer, Model AA 800). Atomic Absorption Spectrophotometer (AAS) is the most extensively used equipment for the determination of metals in a wide variety of matrices. In the present study too, this has been used. The main component of an Atomic Absorption Spectrophotometer is the light source, providing monochromatic light for the absorption process. The absorption of light is proportional to the concentration of the metal present in the samples according to Beer's Law.

Atomic Absorption Spectroscopy calibration metal standards for all measured metals manufactured by SISCO Research Laboratories Private Limited, India were used for calibration. To ensure quality check over the calibration, blank and standards were measured thrice after every ten samples and their relative standard deviation was calculated. While the detection limit for cadmium, lead, chromium, nickel, copper and zinc was 0.001 µg/g, 0.03 µg/g, 0.02 µg/g, 0.04 µg/g, 0.002 µg/g and 0.02 µg/g respectively (Plate 4.7), the quantification limits of metals were three times the detection limit. A spike, a blank, a standard, and a sample were run in triplicate in each analytical run. Spiked recoveries ranged from 84 to 98 per cent. Results obtained from the AAS were in ppb (ng/g) and were converted to ppm (µg/g) in order to evaluate the method efficiencies. The results were not corrected for per cent recovery and expressed in µg/g ± SE of wet weight. In order to facilitate comparison with the reported levels on a dry weight basis, a conversion factor of three was used (Clark and Scheuhammer 2003) wherever required.

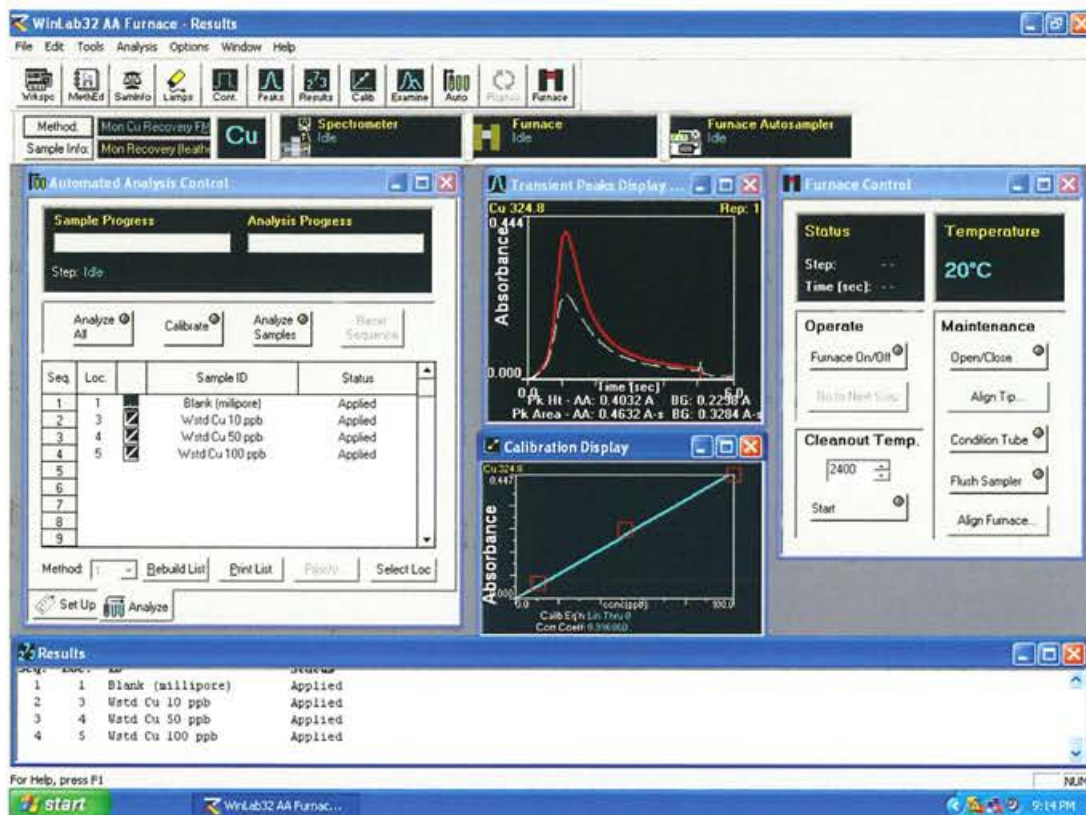


Plate 4.7 Standard graph for copper standard solution

4.5 Statistical analyses

Statistical analyses were performed using SPSS software version 17. The distribution of the data was checked using Shapiro - Wilk W - test. Since the data was normally distributed, two -way ANOVA (Parametric) was performed to understand the variations in the accumulation pattern of metals in organs and species. Each sample was analyzed in triplicate and the results were reported as Mean \pm Standard Error (SE) on a wet weight basis.

One - way ANOVA was performed to understand the variations in the accumulation pattern of metals among feathers and species. Each sample was analyzed in triplicate and the results were reported as Mean \pm Standard Error on a dry weight basis.

Correlations in metal levels between organs and feathers were assessed using Spearman's correlations (ρ). Internal organ kidney and breast feathers accumulated the maximum concentrations of metals, and hence correlation was checked only between kidney and breast feather.

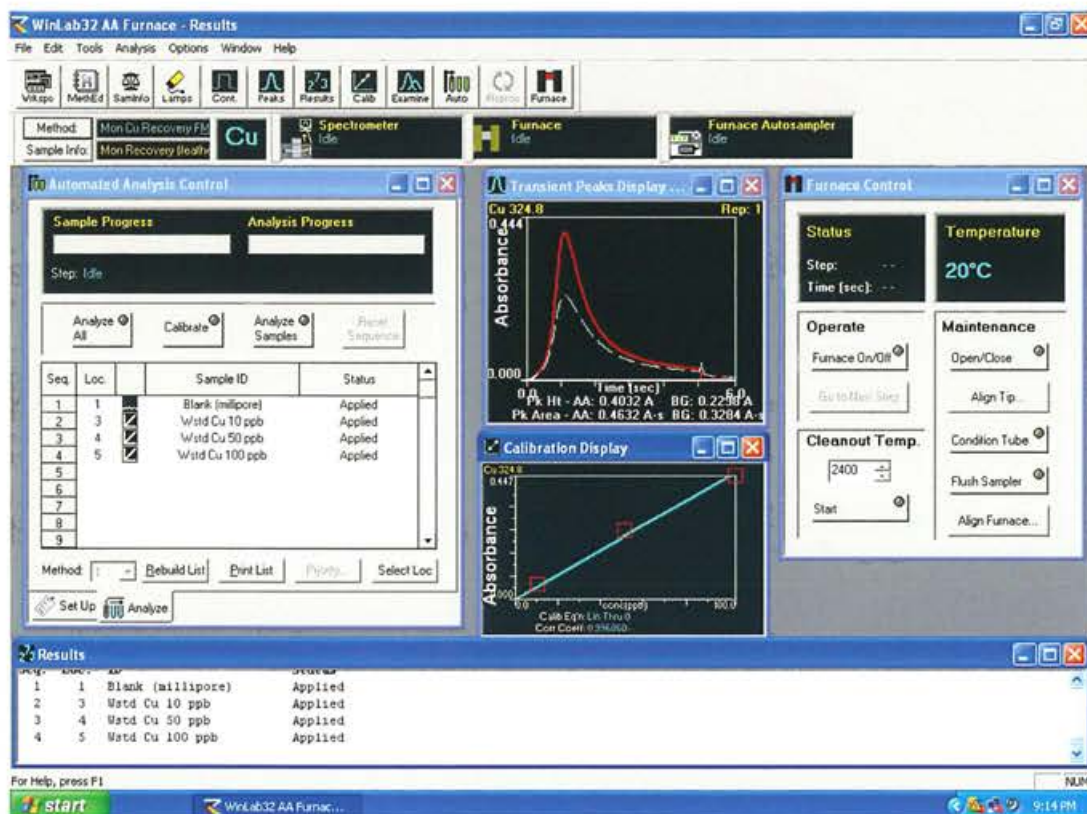


Plate 4.7 Standard graph for copper standard solution

4.5 Statistical analyses

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One - way ANOVA was performed to understand the variations in the accumulation pattern of metals among feathers and species. Each sample was analyzed in triplicate and the results were reported as Mean \pm Standard Error on a dry weight basis.

Correlations in metal levels between organs and feathers were assessed using Spearman's correlations (ρ). Internal organ kidney and breast feathers accumulated the maximum concentrations of metals, and hence correlation was checked only between kidney and breast feather.

Result and Discussion

5. RESULTS AND DISCUSSION

Tissues such as, muscle, liver and kidney along with gizzard of the birds were analyzed for select metals, namely cadmium, lead, chromium, nickel, copper and zinc. Mean and standard error were calculated only for species which had more than three individuals. The data have been compiled to look at the variation in the concentrations of individual metals, total metal load among all the species and with respect to organs, states, feeding habits and feathers.

5.1.a Levels of metals in tissues of various species of birds from select states in India during 2012 - 2014

Concentration of total metals recorded varied significantly among the species (irrespective of organs) (One-way ANOVA, $df= 30$; $F= 71.223$; $p < 0.05$). Among the 31 species analyzed, the maximum level of total metal was recorded in Spot-billed Pelican ($243.63 \pm 10.77 \mu\text{g/g}$) followed by Indian Black Ibis ($232.40 \pm 5.69 \mu\text{g/g}$), Himalayan Vulture ($228.72 \pm 2.80 \mu\text{g/g}$) and Great White Pelican ($228.70 \pm 2.24 \mu\text{g/g}$). While the level of total metals ranged from 203.96 to 226.30 $\mu\text{g/g}$ among 14 species of birds analyzed, in the remaining twelve species of birds, namely Rock Pigeon, Eurasian Collared Dove, Rose-ringed Parakeet, Black Drongo, Cattle Egret, Indian Roller, Indian Pitta, Bar-headed Goose, Red-wattled Lapwing, Common Coot, Asian Koel and Common Kingfisher, it ranged from 130.21 to 198.32 $\mu\text{g/g}$. Among all the species of birds, the minimum level was observed in Emerald Dove ($97.28 \pm 8.19 \mu\text{g/g}$) (Figure 1). The levels of metals recorded with respect to organs in all the species of birds were also computed, and the results are presented in (Figure 5.1).



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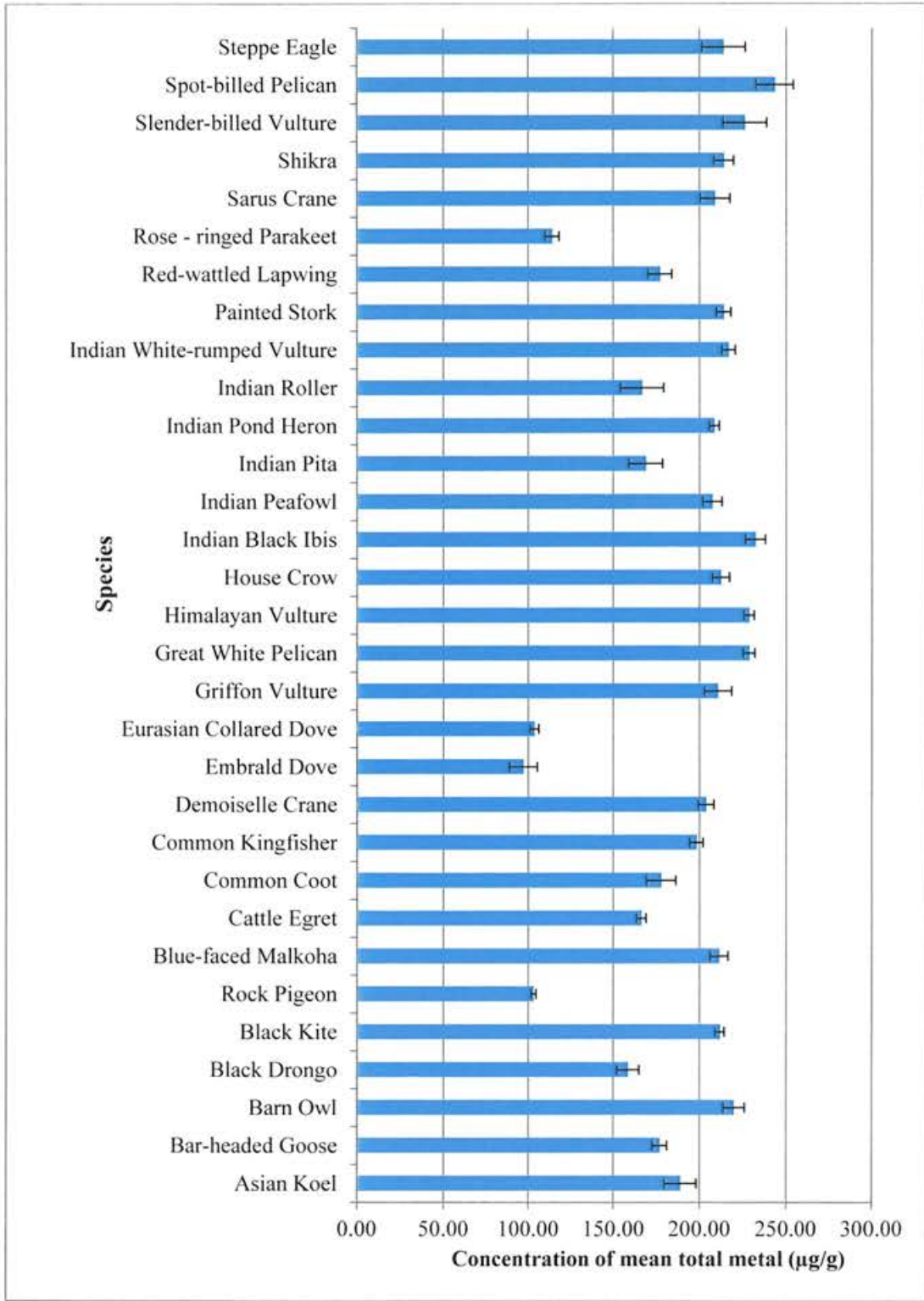


Figure 5.1 Variation in levels of total metal recorded in various species of birds collected dead between 2012 and 2014

5.1.1 Levels of cadmium concentration (mean \pm SE, $\mu\text{g/g}$) in various species of birds collected dead between 2012 and 2014

Among the 31 species of birds studied, the maximum concentration of cadmium was recorded in the kidney tissue of Spot-billed Pelican ($3.54 \pm 1.68 \mu\text{g/g}$) followed by Indian White-rumped Vulture ($3.32 \pm 0.23 \mu\text{g/g}$) and Indian-spotted Eagle ($3.21 \mu\text{g/g}$). The levels of cadmium in the kidney of Asian Koel, Red-wattled Lapwing, Shikra, Barn Owl Common Kingfisher and liver of Indian Spotted Eagle were 2.31, 2.60, 2.52, 2.01, 2.29 and $2.61 \mu\text{g/g}$ respectively. Below Detection Level (BDL) was observed in the muscle tissue of Emerald Dove, Eurasian Collared Dove, Common Coot, Indian Pond Heron, Himalayan Vulture and in liver tissue of Slender-billed Vulture. Statistical analysis showed that there were significant variations in the cadmium concentrations among the species (One-way ANOVA $F=5.587$; $df=30$; $p<0.05$) in table 5.1.

Levels of cadmium recorded in the birds studied were comparable with the levels reported in India and elsewhere (Frank and Borg 1979; Wiemeyer et al. 1986; Schilderman et al. 1997; Zaccaroni et al. 2003; Kalisinska and Salicki 2010; Abduljaleel et al. 2014). Janaydeh et al. (2018) reported the levels of cadmium in kidney ($1.78 \mu\text{g/g}$) and muscle tissues ($0.66 \mu\text{g/g}$) of House Crow in Malasisya. Rajamani and Muralidharan (2015) described the levels of cadmium in male and female Indian White-rumped Vulture in India. Levels of cadmium varied from 0.43 to $0.51 \mu\text{g/g}$ in kidney, $0.31-0.51 \mu\text{g/g}$ in liver and $0.43-2.35 \mu\text{g/g}$ in muscle. In the present study, muscle tissues of White-rumped Vulture had similar concentration, while liver and kidney had higher levels of cadmium. Levels of cadmium ($0.031 \mu\text{g/g}$) in Great Egret were reported by Burger (2013) in New Jersey, and the levels of cadmium recorded in Egrets in the present study were higher than the levels reported above. Begum and Sehrin (2013) documented the concentration of cadmium in Rock Pigeon sampled from Bangladesh, and the levels of cadmium in Rock Pigeon in the present study were higher than the levels reported in Bangladesh.

Table 5.1 Levels of cadmium concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014.

S. No.	Name of the species	Kidney	Liver	Muscle
1.	Bar-headed Goose	1.82 \pm 0.02	0.03 \pm 0.009	NA
2.	Indian Peafowl	1.97 \pm 0.10	0.93 \pm 0.26	0.13 \pm 0.02
3.	Emerald Dove	NA	0.80	BDL
4.	Eurasian Collared Dove	1.30 \pm 0.14	1.48 \pm 0.09	BDL
5.	Rock Pigeon	1.55 \pm 0.25	1.17 \pm 0.05	0.28 \pm 0.07
6.	Asian Koel	2.31	0.67 \pm 0.34	0.10 \pm 0.02
7.	Blue-faced Malkoha	1.63 \pm 0.17	1.29 \pm 0.15	0.48 \pm 0.23
8.	Common Coot	1.62 \pm 0.06	0.42 \pm 0.08	BDL
9.	Demoiselle Crane	1.53 \pm 0.002	0.18 \pm 0.03	0.04
10.	Sarus Crane	1.68	1.58 \pm 0.02	0.13 \pm 0.006
11.	Painted Stork	1.60 \pm 0.06	0.68 \pm 0.35	0.64 \pm 0.12
12.	Great White Pelican	1.54	0.09	NA
13.	Spot-billed Pelican	3.54 \pm 1.00	1.51 \pm 0.22	1.18 \pm 0.04
14.	Cattle Egret	1.20 \pm 0.06	1.16 \pm 0.04	0.12 \pm 0.03
15.	Indian Pond Heron	1.68 \pm 0.10	0.07 \pm 0.02	BDL
16.	Indian Black Ibis	1.74 \pm 0.04	0.78 \pm 0.26	0.27 \pm 0.15
17.	Red-wattled Lapwing	2.60 \pm 0.17	1.15 \pm 0.16	1.44 \pm 0.44
18.	Himalayan Vulture	2.02 \pm 0.16	0.05 \pm 0.01	BDL
19.	Indian White-rumped Vulture	3.32 \pm 0.23	1.62 \pm 0.22	0.42 \pm 0.12
20.	Slender-billed Vulture	1.98	BDL	NA
21.	Griffon Vulture	1.67 \pm 0.13	0.66 \pm 0.56	0.06 \pm 0.01
22.	Indian Spotted Eagle	3.21	2.61	1.09
23.	Shikra	2.52 \pm 0.09	1.52 \pm 0.19	0.36 \pm 0.18
24.	Black Kite	1.56 \pm 0.06	0.21 \pm 0.06	BDL
25.	Barn Owl	2.01 \pm 0.23	0.80 \pm 0.03	0.10 \pm 0.04
26.	Indian Roller	1.11	0.02	BDL
27.	Common Kingfisher	2.29	1.85	0.27
28.	Rose-ringed Parakeet	1.24 \pm 0.04	1.58 \pm 0.12	0.36 \pm 0.12
29.	Indian Pitta	1.54	1.15	0.85
30.	Black Drongo	1.38	BDL	0.12 \pm 0.05
31.	House Crow	1.71 \pm 0.09	0.41 \pm 0.20	0.12 \pm 0.03

Tsipoura et al. (2011) reported levels of cadmium in muscle tissues (4.17 $\mu\text{g/g}$) and liver tissues (122 $\mu\text{g/g}$) of Canada Geese in Harrier Meadow, USA. Levels of cadmium recorded in the Bar-headed Goose in the present study were lower than the levels reported in the Geese refereed above. The levels of cadmium in muscle (7.73 $\mu\text{g/g}$) and liver (311 $\mu\text{g/g}$) tissues of a few water birds were reported in New Jersey (Tsipoura et al. 2011).The levels of cadmium reported in waterbirds of the present study were comparable with the above findings. Torres et al. (2010) reported the levels of cadmium in muscle (0.01 $\mu\text{g/g}$), kidney (0.68 $\mu\text{g/g}$) and liver tissues (0.11 $\mu\text{g/g}$) of Rock Pigeon collected from Spain. The levels of cadmium analyzed in doves in the present study were higher than the levels reported in the previous literature. Burger and Gochefeld (1990) reported the levels of cadmium in muscle (8.7 $\mu\text{g/g}$), liver (374 $\mu\text{g/g}$) and kidney tissues (1.86 $\mu\text{g/g}$) in Herring Gull, a predominant fish-eater from United States of America. The levels of cadmium observed in the fish-eating Painted Stork in the present study were lower than the levels reported in USA. However, the levels of cadmium recorded in the Spot-billed Pelican in the current study showed higher levels than that of the previous studies. Levels of cadmium in liver (0.68 $\mu\text{g/g}$), kidney (1.32 $\mu\text{g/g}$) and muscle (0.001 $\mu\text{g/g}$) of Great Tit have been reported from China (Deng et al. 2007). They also reported the cadmium levels in liver (0.56 $\mu\text{g/g}$), kidney (2.59 $\mu\text{g/g}$) and muscle (0.01 $\mu\text{g/g}$) tissues of Green Finch in China. Overall, the levels of cadmium recorded in the present study are comparable well with the previous literatures and not expected to harm the birds.

Levels of cadmium in liver (0.36 $\mu\text{g/g}$) and kidney (0.19 $\mu\text{g/g}$) of Pigeons in Korea have been reported by Nam and Lee (2006). Further, Beyer et al. (2004) documented the levels cadmium in liver (4.30 $\mu\text{g/g}$) and kidney (33.00 $\mu\text{g/g}$) tissues of Mourning Dove from USA. The levels of cadmium recorded in dove and pigeons in the present study were higher than the levels reported in USA. Migula and Augustyniak (2000) compiled the levels of Cd in muscle (0.86 $\mu\text{g/g}$), liver (2.62 $\mu\text{g/g}$) and kidney tissues (3.82 $\mu\text{g/g}$) of a few gull species in Poland, and the levels were comparable with fish- eating birds included in the present study.

Hutton (1981) reported the levels of cadmium in kidney (27 $\mu\text{g/g}$) and liver (10.5 $\mu\text{g/g}$) of Oystercatcher; and kidney (13 $\mu\text{g/g}$) and liver (2.05 $\mu\text{g/g}$) of Herring Gull. This study also reported the levels of cadmium in kidney (81.4 $\mu\text{g/g}$) and liver (7.52 $\mu\text{g/g}$) of

Great Skua from London. Frank and Borg (1979) reported the levels of cadmium in liver of several species of birds in Sweden, and the levels ranged from 1 to 5 $\mu\text{g/g}$. Sileo et al. (2003) reported the levels of cadmium in liver (0.42 $\mu\text{g/g}$) and kidney (3.14 $\mu\text{g/g}$) of Mallard Duck from USA. They also reported the levels of cadmium in liver (3.70 to 12 $\mu\text{g/g}$) and kidney of goose species (12 to 126 $\mu\text{g/g}$) in USA. Levels of cadmium in muscle (0.006 $\mu\text{g/g}$), liver (0.721 $\mu\text{g/g}$) and kidney (4.308 $\mu\text{g/g}$) in shot Pheasants were also reported in Poland (Dzuga et al. 2012). Kalisinska and Salicki (2010) highlighted the levels of cadmium in muscle (0.34 $\mu\text{g/g}$) of Greater Scaup from Poland. However, the levels of cadmium recorded in a few species of waterbirds analyzed in the present study were lower than the levels reported earlier.

Kitowski et al. (2016) reported the levels of cadmium in liver of White-tailed Sea-eagle *Haliaeetus albicilla* (0.29 $\mu\text{g/g}$), Marsh Harrier (0.282 $\mu\text{g/g}$), Goshawk (0.519 $\mu\text{g/g}$), Sparrow Hawk (0.55 $\mu\text{g/g}$), Common Buzzard (0.78 $\mu\text{g/g}$) and Kestrel (0.32 $\mu\text{g/g}$) in Poland. Zaccaroni et al. (2003) reported the levels of cadmium in male and female Little Owls in Itai and the levels in liver ranged from 102 to 115 ng/g . The levels of cadmium recorded in a few raptorial birds in the present study were higher than the levels reported in birds sampled in Poland and Italy but not upto the levels which can effects birds.

From the available literature (Cain et al. 1983; Furness 1996; Battaglia et al. 2005; Toman et al. 2005; Lucia et al. 2009), it can be inferred that the kidney damage, suppression of egg production, and testicular damage occurred in many species of waterbirds when the cadmium levels approached 20 $\mu\text{g/g}$ in kidney. Cadmium is also reported as a nephrotoxin, most often investigated in the kidney than liver tissues of sea birds (Kalisinska and Salicki 2010). Cadmium concentration in the kidney above 30 $\mu\text{g/g}$ reported to have shows renal tubular necrosis in Mallard Ducks (White and Finley 1978). The levels of cadmium reported in kidney of birds in the present study were lower than the above mentioned levels which had caused kidney damage, suppression of egg production, testicular damage and tubular necrosis.

Burgat (1990) documented that the levels of cadmium above 3 $\mu\text{g/g}$ in liver might indicate an increased environmental exposure, and all the species of birds included in the present study had less than the threshold value. Cadmium can cause adverse behavioral

effects at even lower concentrations (Eisler 1985a; Beyer et al 2004), including slow growth rates (Spahn and Sherry 1999) in birds. Hence, as of now, the levels of cadmium do not appear to be harmful. However, none of the birds included in the present study exceeded the above - cited levels.

5.1.2 Levels of lead concentrations (mean \pm SE, $\mu\text{g/g}$) in various species of birds collected dead between 2012 and 2014

The maximum concentration of lead was recorded in kidney of Indian Pitta (2.39 $\mu\text{g/g}$) followed by the kidney of Slender-billed Vulture (1.90 $\mu\text{g/g}$), Indian Peafowl (1.84 \pm 0.02 $\mu\text{g/g}$), liver of Indian-spotted Eagle (1.73 $\mu\text{g/g}$) and kidney of Himalayan Vulture (1.70 \pm 0.07 $\mu\text{g/g}$). The levels of lead in kidney of Demoiselle Crane, Sarus Crane, Painted Stork, Indian White-rumped Vulture, Griffon Vulture, liver and kidney of Spot-billed Pelican were 1.58, 1.48, 1.43, 1.59, 1.46 and 1.64 $\mu\text{g/g}$ respectively. Below Detection Level (BDL) was observed in the muscles of Emerald Dove, Demoiselle Crane, Sarus Crane, Red-wattled Lapwing and Black Kite (Table 5.2). Significant variations in lead levels were observed among the species of birds studied (one-way ANOVA $F=4.326$; $df=30$; $p<0.05$).

Levels of lead in kidney (21.47 $\mu\text{g/g}$) and muscle (5.06 $\mu\text{g/g}$) in the House Crow were reported from Malaysia (Janaydeh et al. 2016). The levels of lead recorded in the House Crow in the present study were lower than the levels reported in the above - referred study. Giammarino et al. (2014) documented the levels of lead in liver (0.09 $\mu\text{g/g}$) and kidney (0.07 $\mu\text{g/g}$) in Hooded Crow from Italy. Komosa et al. (2012) reported the levels of lead in liver of Rook (2.35 $\mu\text{g/g}$) from Poland. The levels of lead recorded in the House Crow in the present study were higher than the levels reported in the above - mentioned studies. Rajamani and Muralidharan (2015) reported the levels of lead in male and female Indian White-rumped Vulture to have ranged from 2.42 to 2.45 $\mu\text{g/g}$ in kidney, 1.92 to 2.78 $\mu\text{g/g}$ in liver and 0.41 to 1.72 $\mu\text{g/g}$ in muscle. The levels of lead observed in Indian White-rumped Vulture included in the present study were lower than the previous research findings. Accumulation of lead at subclinical levels could make vultures susceptible to diseases and environmental stresses (Pérez-López et al. 2008) such as food scarcity and habitat destruction. Continued monitoring of the impact of potential contaminants including metals on vulture was necessary for protecting the species in the Indian Sub-continent (Rajamani and Subramanian 2015).

Table 5.2 Levels of lead concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014

S. No.	Name of the species	Kidney	Liver	Muscle
1.	Bar-headed Goose	1.39 \pm 0.11	BDL	NA
2.	Indian Peafowl	1.84 \pm 0.02	1.33 \pm 0.07	0.24 \pm 0.07
3.	Emerald Dove	NA	0.98	BDL
4.	Eurasian Collared Dove	1.15 \pm 0.02	0.76 \pm 0.13	0.17 \pm 0.04
5.	Rock Pigeon	1.27 \pm 0.04	0.40 \pm 0.08	0.23 \pm 0.03
6.	Asian Koel	1.42	0.74 \pm 0.21	0.09 \pm 0.05
7.	Blue-faced Malkoha	1.38 \pm 0.03	0.51 \pm 0.16	0.34 \pm 0.11
8.	Common Coot	1.32 \pm 0.06	0.35 \pm 0.12	0.17 \pm 0.05
9.	Demoiselle Crane	1.58 \pm 0.14	0.65 \pm 0.38	BDL
10.	Sarus Crane	1.48	0.08 \pm 0.008	BDL
11.	Painted Stork	1.43 \pm 0.10	0.90 \pm 0.37	0.29 \pm 0.20
12.	Great White Pelican	1.60	0.58	NA
13.	Spot-billed Pelican	1.59 \pm 0.24	1.64 \pm 0.06	0.64 \pm 0.28
14.	Cattle Egret	1.16 \pm 0.01	0.64 \pm 0.11	0.29 \pm 0.67
15.	Indian Pond Heron	1.36 \pm 0.09	0.80 \pm 0.11	0.48 \pm 0.09
16.	Indian Black Ibis	1.31 \pm 0.03	1.13 \pm 0.21	0.83 \pm 0.14
17.	Red-wattled Lapwing	1.16 \pm 0.04	1.16 \pm 0.14	BDL
18.	Himalayan Vulture	1.70 \pm 0.07	0.50 \pm 0.11	0.15
19.	White-rumped Vulture	1.59 \pm 0.15	1.34 \pm 0.10	0.23 \pm 0.10
20.	Slender-billed Vulture	1.90	0.91	NA
21.	Griffon Vulture	1.46 \pm 0.05	0.58 \pm 0.33	0.46 \pm 0.162
22.	Indian Spotted Eagle	1.66	1.73	0.84
23.	Shikra	1.28 \pm 0.09	1.05 \pm 0.45	0.25 \pm 0.08
24.	Black Kite	1.24 \pm 0.03	0.82 \pm 0.12	BDL
25.	Barn Owl	1.41 \pm 0.09	1.46 \pm 0.22	0.40 \pm 0.33
26.	Indian Roller	1.24	0.65	0.50
27.	Common Kingfisher	1.29	1.15	0.76
28.	Rose-ringed Parakeet	1.20 \pm 0.04	0.48 \pm 0.13	0.41 \pm 0.12
29.	Indian Pitta	2.39	1.41	BDL
30.	Black Drongo	1.08	0.34	0.58 \pm 0.19
31.	House Crow	1.47 \pm 0.04	1.34 \pm 0.07	0.40 \pm 0.12

Begum and Sherin (2013) reported the levels of lead in muscle (0.39 $\mu\text{g/g}$), liver (2.85 $\mu\text{g/g}$) and kidney (1.17 $\mu\text{g/g}$) in Rock Pigeon sampled from Bangladesh. Beyar et al. (2004) reported the levels of lead in liver (1.2 $\mu\text{g/g}$) and kidney (7.2 $\mu\text{g/g}$) of a few species of dove namely Rock Pigeon, Eurasian Collared Dove and Emerald Dove sampled from USA. Nam and Lee (2006) reported the levels of lead in kidney (5.18 $\mu\text{g/g}$) and liver (5.18 $\mu\text{g/g}$) of Rock Pigeon collected from Korea. Torres et al. (2010) reported lead in muscle (0.11 $\mu\text{g/g}$), kidney (0.29 $\mu\text{g/g}$) and liver (0.29 $\mu\text{g/g}$) in Rock Pigeon sampled from Spain. The levels of lead in dove species, namely Emerald Dove, Eurasian Collared Dove and Rock Pigeon analyzed in the present study were lower than the levels reported in the studies referred - above.

Tsipoura et al. (2011) reported the level of lead in muscle (3.31 $\mu\text{g/g}$) and liver (208 $\mu\text{g/g}$) in Canada Geese sampled from USA. The levels of lead in muscle (3.31 $\mu\text{g/g}$) and liver (238 $\mu\text{g/g}$) of a few waterbird species were reported from New Jersey (Tsipoura et al. 2011). Szymczyk and Zalewski (2003) documented the levels of lead in liver (0.22 $\mu\text{g/g}$) of Mallard Duck from Poland. Migula and Augustyniak (2000) compiled the levels of lead in muscle (12.86 $\mu\text{g/g}$), liver (12.35 $\mu\text{g/g}$) and kidney (15.77 $\mu\text{g/g}$) in a few Gull species of Poland. Hutton (1981) reported the levels of lead in kidney (32.14 $\mu\text{g/g}$) and liver (21.6 $\mu\text{g/g}$) of a few species of sea birds in United Kingdom. Lucia et al. (2010a) reported the levels of lead in muscle (4.3 $\mu\text{g/g}$), liver (15.4 $\mu\text{g/g}$) and kidney (152.3 $\mu\text{g/g}$) of Greylag Goose from France. The levels of lead in muscle (8.22 $\mu\text{g/g}$), liver (51.8 $\mu\text{g/g}$) and kidney (101 $\mu\text{g/g}$) in Herring Gull were reported in USA (Burger and Gochfeld 1990). Sileo et al. (2003) reported the levels of lead in liver (3.8 $\mu\text{g/g}$) and kidney (51 $\mu\text{g/g}$) in Mallard in USA. They also reported lead in liver (2.1 to 2.9 $\mu\text{g/g}$) of Canada Goose in Oklahoma and Kansas, USA and the levels ranged from 4.5 to 51 $\mu\text{g/g}$ in kidney. The levels of lead observed in waterfowls in the present study were lower than the levels reported in the studies listed above.

Jayakumar and Muralidharan (2011) documented lead in muscle (20.77 $\mu\text{g/g}$), liver (15.44 $\mu\text{g/g}$) and kidney (9.4 $\mu\text{g/g}$) of Indian Pond Heron from Nilgiri, Tamil Nadu, India. The levels of lead observed in Indian Pond Heron in the present study were 10 times lower than the level mentioned above. Van wyk et al. (2001) reported lead in muscle (7.79 $\mu\text{g/g}$), liver (14.81 $\mu\text{g/g}$) and kidney (12.3 $\mu\text{g/g}$) in African White-rumped Vulture in South Africa. The levels of lead in Indian White-rumped Vulture included in

the present study were lower than the levels reported in African White-rumped Vulture. Further, Naccari et al. (2009) reported the levels of lead in muscle (0.87 µg/g), liver (14.53 µg/g) and kidney (0.54 µg/g) of Common Buzzard in Italy. The levels of lead recorded in the muscle and kidney of raptorial birds in the present study were lower than the levels reported by Naccari et al. (2009).

Kitowski et al. (2016) highlighted the levels of lead in the liver of White – tailed Sea-eagles (0.585 µg/g), Marsh Harrier (0.684 µg/g), Goshawk (1.152 µg/g), Sparrow Hawk (0.65 µg/g), Common Buzzard (1.217 µg/g) and Kestrel (0.728 µg/g) in Poland. Similarly, Zaccaroni et al. (2003) reported lead in liver of male and female Little Owl (a nocturnal raptor) from 0.188 to 0.332 µg/g. Concentration of lead in liver and kidney tissues of African White-rumped Vulture, (liver – 4.53 µg/g; kidney – 3.17 µg/g) and Cinereous Vulture (liver – 3.45 µg/g; kidney – 3.45 µg/g) were reported in South Africa (Van Wyk et al. 2001; Nam and Lee 2009). Kavun (2004) reported the mean levels of lead in liver (21.87 µg/g) and kidney (21.33 µg/g) of the globally threatened Cinereous Vulture, wintering in Southern Primorye, Russia. The levels of lead in liver of a few raptorial birds, namely Himalayan Vulture, Indian White-rumped Vulture, Slender-billed Vulture, Spotted Eagle, Shikra, Black Kite and Barn Owl analyzed in the present study were higher than the levels reported in the studies explained above.

Deng et al. (2007) compiled the levels of lead in liver (0.64 µg/g), kidney (1.21 µg/g) and muscle (0.01 µg/g) of Great Tit from China. They also reported lead in liver (0.45 µg/g), kidney (6.68 µg/g) and muscle (0.11 µg/g) in Green Finch from China. Kalisinska and Salicki (2010) reported the levels of lead in muscle (2.29 µg/g) of Greater Scaup from Poland. The levels of lead in muscle (0.007 µg/g), liver (0.059 µg/g) and kidney (0.096 µg/g) of shot Pheasants have been reported in Poland (Dzungan et al. 2012). The levels of lead observed in birds included in this study were similar to those levels available in the above - referred studies.

It can be noted that if the levels of lead are above 2.0 µg/g in livers and above 2.0 µg/g in kidneys of wild birds, it can cause toxic and fatal effects respectively (Franson 1996; Clark and Scheuhammer 2003). The levels of lead observed in liver of Indian-spotted Eagle (1.73 µg/g) and Spot-billed Pelican (1.64 ± 0.06 µg/g) in the present study were near to the levels which can be fatal. Unfortunately, these two species are

already in Vulnerable and Near Threatened categories (Birdlife International 2019). Similarly, a higher concentration of lead was recorded in kidney of Indian Pitta (2.39 µg/g), Critically Endangered Slender-billed Vulture (1.90 µg/g), National bird of India- the Indian Peafowl (1.84 ± 0.02 µg/g) and Near Threatened Himalayan Vulture (1.70 ± 0.07 µg/g). This is a matter of serious concern.

Birds are ideal models for lead toxicity (Burger et al. 2015). Hoffman et al. (2000) observed lead to produce calcification in marrow cells of Geese *Branta canadensis* in USA. Similarly, Gochfeld (2000) observed a few abnormalities with increasing lead concentration in birds. Improper balance, depth perception, behavioral changes, such as begging and feeding, treadmill learning, and individual recognition were some of the abnormalities. The levels of lead in tissues of the birds included in the present study were lower than the levels reported to cause various ill effects mentioned above. Further, it is reported that the Anseriformes birds were affected with sub-clinical poisoning of lead when the levels exceeded 6 µg/g in the both liver and kidney (Burger and Gochfeld 2000c).

The clinical and sub-clinical poisonings were also noted in birds with increased concentration of lead in both kidney (6-15 µg/g) and liver (6-10 µg/g) (Beyer et al. 2000). Kim and Koo (2010) reported the threshold limits (<6 µg/g) of lead in birds, which could be indicative of chronic lead exposure which subsequently causes clinical signs, namely diarrhea and anorexia in birds (Van der Horst 1993). However, the levels of lead reported in liver and kidney of birds analyzed in the present study were well below the levels mentioned above.

5.1.3. Levels of chromium concentrations (mean ± SE, µg/g) in various species of birds collected dead between 2012 and 2014

Statistical tests showed that there were significant variations in chromium residues among the bird species (One-way ANOVA $F=8.789$; $df=30$; $p<0.05$) included in the current study. Among the species analyzed, the Spot-billed Pelican had the highest concentration of chromium (4.60 ± 1.54 µg/g) in liver followed by liver of Common Kingfisher (4.03 µg/g), kidney of Indian Spotted Eagle (3.98 µg/g), livers of Asian Koel (3.90 ± 1.46 µg/g) and Painted Stork (3.09 ± 0.65 µg/g). The lowest level was observed in muscle of Rose-ringed Parakeet (0.21 ± 0.04 µg/g). Concentration of Cr was below detection level (BDL) in Himalayan Vulture, liver of Slender-billed Vulture, muscle of Indian Spotted Eagle, Indian Roller and kidney of House Crow (Table 5.3).

Table 5.3 Levels of chromium concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014.

S. No.	Name of the species	Kidney	Liver	Muscle
1.	Bar-headed Goose	0.96 \pm 0.87	NA	1.04 \pm 0.005
2.	Indian Peafowl	1.45 \pm 0.12	0.44 \pm 0.18	0.15 \pm 0.04
3.	Emerald Dove	NA	2.12	0.31
4.	Eurasian Collared Dove	1.77 \pm 0.81	1.70 \pm 0.35	0.62 \pm 0.25
5.	Rock Pigeon	2.45 \pm 0.79	1.47 \pm 0.24	0.91 \pm 0.24
6.	Asian Koel	0.15	3.9 \pm 1.46	1.16 \pm 0.31
7.	Blue-faced Malkoha	0.79 \pm 0.24	1.15 \pm 0.42	0.39 \pm 0.08
8.	Common Coot	0.53 \pm 0.14	0.46 \pm 0.12	0.31 \pm 0.11
9.	Demoiselle Crane	1.66 \pm 0.03	1.40 \pm 0.14	0.30
10.	Sarus Crane	1.65	2.04 \pm 0.12	0.71 \pm 0.15
11.	Painted Stork	2.42 \pm 0.65	3.09 \pm 0.65	2.02 \pm 0.67
12.	Great White Pelican	0.21	0.21	NA
13.	Spot-billed Pelican	2.88 \pm 0.33	4.60 \pm 1.54	1.52 \pm 0.22
14.	Cattle Egret	1.94 \pm 0.15	1.93 \pm 0.18	0.35 \pm 0.05
15.	Indian Pond Heron	0.41 \pm 0.09	0.43 \pm 0.15	0.21 \pm 0.04
16.	Indian Black Ibis	1.97 \pm 0.41	2.66 \pm 0.28	0.65 \pm 0.19
17.	Red-wattled Lapwing	2.45 \pm 0.23	3.18 \pm 0.33	2.11 \pm 0.31
18.	Himalayan Vulture	0.92 \pm 0.59	0.48 \pm 0.16	BDL
19.	White-rumped Vulture	2.49 \pm 0.33	2.13 \pm 0.24	2.05 \pm 0.49
20.	Slender-billed Vulture	2.49	BDL	NA
21.	Griffon Vulture	2.33 \pm 0.38	2.04 \pm 0.10	0.30 \pm 0.11
22.	Indian Spotted Eagle	3.98	3.21	BDL
23.	Shikra	2.39 \pm 1.35	3.23 \pm 0.29	2.00 \pm 0.39
24.	Black Kite	0.44 \pm 0.15	0.80 \pm 0.51	0.57 \pm 0.23
25.	Barn Owl	1.58 \pm 1.11	1.65 \pm 0.05	0.17 \pm 0.08
26.	Indian Roller	1.26	0.21	BDL
27.	Common Kingfisher	1.85	4.03	0.47
28.	Rose-ringed Parakeet	0.50 \pm 0.09	0.36 \pm 0.03	0.21 \pm 0.04
29.	Indian Pitta	2.63	1.94	0.70
30.	Black Drongo	1.76	1.62	1.52 \pm 1.07
31.	House Crow	BDL	1.70 \pm 0.22	0.29 \pm 0.10

Egwumah et al. (2017) reported chromium in the liver tissues of Black-headed Oriole from various sites of Nigeria that ranged from 5.29 to 6.26 $\mu\text{g/g}$. Begum and Sherin (2013) reported the levels of chromium in muscle (0.91 $\mu\text{g/g}$) and kidney (0.93 $\mu\text{g/g}$) in Rock Pigeon sampled from Bangladesh. The levels of chromium in kidney of all dove species in the present study were higher than the levels mentioned in the studies referred above. Tshipoura et al. (2011) reported the level of chromium in muscle (48.5 $\mu\text{g/g}$) and liver (201 $\mu\text{g/g}$) in Canada Geese sampled from USA. Levels of chromium in muscle (28.1 $\mu\text{g/g}$) and liver (110 $\mu\text{g/g}$) of a few water birds were reported from New Jersey (Tshipoura et al. 2011). The levels of chromium detected in waterfowls included in the current study were far less than the previous studies. Van wyk et al. (2001) reported chromium in muscle (13.56 $\mu\text{g/g}$), liver (19.57 $\mu\text{g/g}$) and kidney (18.39 $\mu\text{g/g}$) tissues of African White-rumped Vulture in South Africa. In the present study Indian White-rumped Vulture had lower level of chromium than those levels reported by Van wyk et al. (2001).

Kitowski et al. (2016) reported chromium in the liver tissues of White-tailed Sea-Eagles (0.81 $\mu\text{g/g}$), Marsh Harrier (0.752 $\mu\text{g/g}$), Goshawk (0.549 $\mu\text{g/g}$) Sparrow Hawk (1.22 $\mu\text{g/g}$), Common Buzzard (0.466 $\mu\text{g/g}$) and Kestrel (0.64 $\mu\text{g/g}$) in Poland. Zaccaroni et al. (2003) reported the levels of chromium in male and female Little Owls in Italy and the levels in liver ranged from 2.88 to 2.97 $\mu\text{g/g}$. Among raptors studied, the chromium levels in liver of Himalayan Vulture was lower and in other raptors, namely Slender-billed Vulture, Eurasian Vulture, Indian Spotted Eagle, Shikra, Black Kite. However, Barn Owl showed higher levels than the previous studies. Deng et al. (2007) compiled the levels of chromium in liver (1.86 $\mu\text{g/g}$), kidney (6.26 $\mu\text{g/g}$) and muscle (1.14 $\mu\text{g/g}$) of Great Tit from China. They also reported the levels of chromium in liver (0.96 $\mu\text{g/g}$), kidney (2.61 $\mu\text{g/g}$) and muscle (1.07 $\mu\text{g/g}$) in Green Finch from China. The chromium levels detected in birds of the present study were comparable with earlier findings. Further, it may be noted that when the levels of chromium exceeded 40 $\mu\text{g/g}$ in tissues, it should be considered as evidence of chromium contamination in birds (Eisler 1986). However, the levels of chromium did not exceed in any of the species of birds currently studied.

Mora and Anderson (1995) reported chromium concentrations in liver (6.5 to 17 $\mu\text{g/g}$) to be below the threshold level for biological effects. It is further noted that the chromium causes stunted growth, head and eye abnormalities among wildlife (Custer et al. 2003).

However, the levels of chromium recorded in various tissues of birds of the present study were lower than the levels which could cause stunted growth and abnormalities in birds.

Based on the levels documented in the current study, the concentrations of chromium in all the organs were below the threshold level (50 µg/g) as reported by (Abduljaleel et al. 2012). However, if the exposure to different forms of chromium increases, it can lead to many ill effects including cancer (Kler et al. 2014). Elevated chromium in Mallard Duck has caused reproductive impairment (Kerte'sz and Fa'ncsi 2003). Kler et al. (2014) reported that the chromium concentration produced adverse effects in embryo development, viability and hatching in Mallard Ducks. Common Terns, nesting in Rhode Island also did not show any reduced reproductive success, reduced growth rate, even though they were exposed to chromium in livers upto 18.3 µg/g (Custer et al. 1986).

A case which contradicts the statements made above has been documented on the potential population-level effects of chromium in the food chain of an aquatic bird – the Lesser Scaup in Central America. Bioaccumulation of chromium was identified in the migratory Lesser Scaup population in the course of searching for the cause of their dramatic population decline. However, elevated chromium levels detected in the scaups' livers were lower than any levels known to have caused effects in lab studies. Further little data exists regarding chromium threshold effect and effects on reproductive capacity of birds in field situations (Custer et al. 2003).

5.1.4. Levels of nickel concentrations (mean ± SE, µg/g) in various species of birds collected dead between 2012 and 2014

Of all the species of birds studied, the levels of nickel in Indian White-rumped Vulture were found to be the highest in kidney tissues ($10.85 \pm 0.78 \mu\text{g/g}$). The second maximum concentration of Nickel was followed by Griffon Vulture ($10.82 \pm 9.80 \mu\text{g/g}$). Level in liver tissues of Indian White-rumped Vulture ($9.37 \pm 1.38 \mu\text{g/g}$) was comparable. The levels of nickel in liver and kidney of Indian Pitta, liver of Common Kingfisher and liver of Painted Stork were 8.82, 7.69, 7.68 and 7.12 µg/g respectively, and the levels varied significantly among the species (One-way ANOVA $F=4.562$; $df=30$; $p<0.05$). Liver tissues of Bar-headed Goose and Common Coot, muscle of Emerald Dove, Rock Pigeon, Asian Koel, Himalayan Vulture, Shikra and Indian Roller had levels below detection limit (Table 5.4).

Deng et al. (2007) compiled the levels of nickel in liver (0.6 µg/g), kidney (1.48 µg/g) and muscle tissues (0.41µg/g) of Great Tit from China. They also reported the levels of nickel in liver (0.28 µg/g), kidney (0.91 µg/g) and muscle tissues (1.76 µg/g) in Green Finch from China. The levels of nickel in birds of the present study were comparable to these levels. Wren et al. (1983) reported nickel in muscle tissues (1.5µg/g) in American Coot from Ontario. The levels of nickel reported in Common Coot in the present study was higher than the levels mentioned above. Ranta et al. (1978) reported the levels of nickel in liver (1.3 to 8.4 µg/g) of Mallard Duck from Ontario. Custer and Mulhern (1983) reported nickel in liver (0.1 to 9.2 µg/g) of Black-crowned Night Heron in USA. Burger and Gochefeld (1985) reported nickel in liver (1.5 µg/g) of Black Duck and Mallard Duck (0.9 µg/g) from New Jersey. The levels of nickel in waterbirds included in the present study were comparable with above referred earlier studies. Honda et al. (1985) reported the levels of nickel in muscle tissues of Egret species (0.03-0.07 µg/g) from Korea. Gochfeld and Burger (1987) reported the levels of nickel in liver (0.13 µg/g) of Mallard Duck from New Jersey. The levels of nickel recorded in Cattle Egret and Bar-headed Goose included in the present study were higher than the levels reported above.

Falandysz (1986) reported nickel in muscle (0.03 to 0.17 µg/g), liver (0.15 to 0.29 µg/g) and kidney tissues (0.11 to 0.15 µg/g) of White-tailed Sea-Eagle collected from Poland. Further, Kitowski et al. (2016) reported the levels of nickel in liver of White –tailed Sea-Eagles (0.234 µg/g) in Poland. The levels of nickel in kidney, liver and muscle tissues of Indian Spotted Eagle were 10 folds higher than the levels reported in White-tailed Sea-Eagle in Poland. The levels of nickel in liver of Marsh Harrier (0.3 µg/g), Goshawk (0.295 µg/g) Sparrow Hawk (0.248 µg/g), Common Buzzard (0.247 µg/g) and Kestrel (0.347 µg/g) in Poland have been reported (Kitowski et al. 2016). The levels of nickel in the raptorial birds included in the present study were higher than the levels mentioned in above-referred studies.

Van wyk et al. (2001) reported nickel in muscle (10.75 µg/g), liver (13.92 µg/g) and kidney tissues (8.1µg/g) of African White-backed Vulture in South Africa. The levels of nickel observed in muscle and liver of Indian White–backed Vulture were lower and the levels in kidney were higher than the African White-rumped Vulture from South Africa. The levels of nickel observed in the present study can be compared well with the Turkey Vulture (0.1 µg/g) from California (Wiemeyer et al. 1986). But there is no information on the ill-effects of such levels on the species of birds referred.

Table 5.4 Levels of nickel concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014.

S. No.	Name of the species	Kidney	Liver	Muscle
1.	Bar-headed Goose	2.06 \pm 0.02	BDL	NA
2.	Indian Peafowl	1.74 \pm 0.07	0.85 \pm 0.54	0.38 \pm 0.12
3.	Emerald Dove	NA	0.40	BDL
4.	Eurasian Collared Dove	2.63 \pm 0.13	4.74 \pm 3.32	0.42 \pm 0.16
5.	Rock Pigeon	1.54 \pm 0.36	1.52 \pm 0.27	BDL
6.	Asian Koel	0.15	0.13 \pm 0.03	BDL
7.	Blue-faced Malkoha	6.24 \pm 4.12	4.63 \pm 3.30	0.72 \pm 0.47
8.	Common Coot	0.53 \pm 0.14	BDL	0.32 \pm 0.11
9.	Demoiselle Crane	1.35 \pm 0.18	4.24 \pm 1.44	1.50
10.	Sarus Crane	2.64	2.08 \pm 0.74	0.52 \pm 0.25
11.	Painted Stork	3.33 \pm 0.64	7.12 \pm 2.38	0.67 \pm 0.29
12.	Great White Pelican	1.19	1.15	NA
13.	Spot-billed Pelican	2.98 \pm 0.69	2.87 \pm 0.45	5.71 \pm 2.65
14.	Cattle Egret	2.79 \pm 0.18	1.49 \pm 0.41	0.54 \pm 0.10
15.	Indian Pond Heron	1.53 \pm 0.21	1.21 \pm 0.26	0.67 \pm 0.06
16.	Indian Black Ibis	5.90 \pm 1.42	5.52 \pm 2.71	1.23 \pm 0.17
17.	Red-wattled Lapwing	2.87 \pm 1.10	4.53 \pm 0.55	1.76 \pm 0.126
18.	Himalayan Vulture	0.62 \pm 0.14	0.78 \pm 0.17	BDL
19.	Indian White-rumped Vulture	10.85 \pm 0.78	9.37 \pm 1.38	3.18 \pm 1.35
20.	Slender-billed Vulture	6.31	4.96	NA
21.	Griffon Vulture	10.82 \pm 9.80	1.43 \pm 0.53	1.29 \pm 0.15
22.	Indian Spotted Eagle	1.42	1.36	1.24
23.	Shikra	6.08 \pm 0.48	2.69 \pm 0.55	BDL
24.	Black Kite	4.27 \pm 0.16	0.39 \pm 0.14	0.85 \pm 0.36
25.	Barn Owl	5.53 \pm 0.55	4.73 \pm 0.84	0.66 \pm 0.57
26.	Indian Roller	1.48	1.30	BDL
27.	Common Kingfisher	3.25	7.68	1.47
28.	Rose-ringed Parakeet	1.87 \pm 0.25	4.05 \pm 1.50	2.73 \pm 0.93
29.	Indian Pitta	7.69	8.82	1.90
30.	Black Drongo	1.76	0.13	1.52 \pm 1.04
31.	House Crow	4.03 \pm 1.13	5.18 \pm 4.34	3.83 \pm 2.21

In general, the information on the toxic influence/effect of nickel on birds is more limited than that of mammals. According to Outridge and Scheuhammer (1993), the higher doses of nickel ranging from 300 to 800 $\mu\text{g/g}$ in the diet of Great Tit chicks produced variable effects including death, whereas, the higher dose of nickel in adult birds showed no evidence of systemic or reproductive toxicity. Hence, tissue concentrations of nickel are not considered as a reliable indicator of potential toxicity. However, it could be used as an indicator of environmental conditions (Cempel and Nikel 2006). Levels of nickel in kidney tissues of 25 Mallard ducklings which were reported to have died in USA had 1200 $\mu\text{g/g}$. Further, 11 ducklings which survived had undergone necropsy and the nickel concentrations in liver tissues were less than 1 $\mu\text{g/g}$. The levels of nickel reported in liver of the present study were lower than the levels reported to have caused mortality in birds (Cain and Paffort 1981). Generally, the levels of nickel in kidneys were higher than liver. It also to be noted that if the levels of nickel exceeded 30 $\mu\text{g/g}$ in the kidneys and 9 $\mu\text{g/g}$ in the liver could cause adverse effects on birds (Outridge and Scheuhammer 1993). The levels of nickel recorded in kidney and liver tissues in the bird species included in the present study were lower than the levels reported to have caused adverse effects in birds elsewhere in the world.

5.1.5. Levels of copper concentration (mean \pm SE, $\mu\text{g/g}$) in various species of birds collected dead between 2012 and 2014

The highest concentration of copper was recorded in liver tissues of Spot-billed Pelican ($53.14 \pm 14.13 \mu\text{g/g}$) followed by kidney tissues of Eurasian Vulture ($52.70 \pm 8.34 \mu\text{g/g}$), liver tissues of Red-napped Ibis ($51.64 \pm 9.20 \mu\text{g/g}$) and kidney of Indian Spotted Eagle ($51.11 \mu\text{g/g}$). The levels of copper in muscle of Spot-billed Pelican, kidney tissues of Red-napped Ibis, Himalayan Vulture, Sarus Crane, Slender-billed Vulture, Roller and kidney of Great White Pelican were 42.43, 42.93, 43.47, 44.33, 47.18, 47.62 and 48.92 $\mu\text{g/g}$ respectively. The minimum levels of copper were recorded in muscle tissues of Black Drongo (10.52 $\mu\text{g/g}$) and Asian Koel (11.11 $\mu\text{g/g}$) (Table 5.5). A significant variation was observed in the levels of cadmium detected among the species (One-way ANOVA $F=5.393$; $df=30$; $p<0.05$).

Table 5.5 Levels of copper concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014.

S. No.	Name of the species	Muscle	Liver	Kidney
1.	Bar-headed Goose	NA	14.03 \pm 1.50	12.00 \pm 0.21
2.	Indian Peafowl	12.10 \pm 0.58	33.88 \pm 3.65	29.32 \pm 9.90
3.	Emerald Dove	11.94	33.43	NA
4.	Eurasian Collared Dove	16.27 \pm 3.09	27.19 \pm 3.50	28.12 \pm 9.25
5.	Rock Pigeon	12.73 \pm 0.51	19.49 \pm 1.73	31.65 \pm 5.86
6.	Asian Koel	11.11 \pm 6.82	32.80 \pm 1.45	42.08
7.	Blue-faced Malkoha	15.64 \pm 2.38	27.17 \pm 4.18	25.29 \pm 3.74
8.	Common Coot	11.85 \pm 0.50	31.35 \pm 1.69	34.04 \pm 5.31
9.	Demoiselle Crane	11.88	13.30 \pm 1.10	15.35 \pm 1.84
10.	Sarus Crane	20.62 \pm 2.53	28.98 \pm 5.20	44.33
11.	Painted Stork	17.63 \pm 0.87	22.59 \pm 7.66	24.40 \pm 2.25
12.	Great White Pelican	NA	38.48	42.43
13.	Spot-billed Pelican	47.62 \pm 18.59	53.14 \pm 14.13	40.71 \pm 20.07
14.	Cattle Egret	16.73 \pm 3.33	32.22 \pm 1.17	39.10 \pm 1.46
15.	Indian Pond Heron	12.14 \pm 0.37	35.42 \pm 1.04	42.34 \pm 1.30
16.	Indian Black Ibis	31.91 \pm 9.30	51.64 \pm 9.20	48.92 \pm 9.77
17.	Red-wattled Lapwing	14.25 \pm 1.32	42.23 \pm 5.51	30.04 \pm 10.32
18.	Himalayan Vulture	19.89	38.42 \pm 0.56	47.18 \pm 1.87
19.	White-rumped Vulture	17.54 \pm 2.42	32.15 \pm 4.43	19.15 \pm 5.39
20.	Slender-billed Vulture	NA	25.04	42.95
21.	Griffon Vulture	12.75 \pm 2.05	25.78 \pm 4.45	52.70 \pm 8.34
22.	Indian Spotted Eagle	13.39	28.75	51.11
23.	Shikra	17.45 \pm 0.96	26.24 \pm 2.93	43.47 \pm 5.90
24.	Black Kite	12.25 \pm 0.65	34.00 \pm 0.96	41.68 \pm 0.54
25.	Barn Owl	13.20 \pm 2.67	32.65 \pm 3.80	41.03 \pm 3.56
26.	Indian Roller	13.61	33.15	42.93
27.	Common Kingfisher	29.00	26.50	24.94
28.	Rose-ringed Parakeet	20.36 \pm 4.22	22.35 \pm 3.62	25.49 \pm 5.07
29.	Indian Pitta	13.75	26.82	30.19
30.	Black Drongo	10.52 \pm 0.47	27.84	35.48
31.	House Crow	16.07 \pm 1.78	40.19 \pm 4.86	30.39 \pm 4.43

Janaydeh et al. (2016) reported copper in muscle (13.1 µg/g), liver (23.93 µg/g) and kidney tissues (16.61 µg/g) of House Crow in Malaysia. Komosa et al. (2012) reported the levels of copper in liver tissues of Rook (11.8 µg/g) sampled from Poland. Horai et al. (2007) documented the levels of copper in muscle (13.2 µg/g), liver (42.5 µg/g) and kidney (14.09 µg/g) tissues of Jungle Crow from Japan. They also reported the levels of copper in muscle (13.3 µg/g), liver (29.6 µg/g) and kidney tissues (16 µg/g) in Carrion Crow sampled from Japan. The levels of copper in House Crow in the present study were higher than the levels reported in the publications mentioned above. Begum and Sherin (2013) reported the levels of copper in muscle (5.48 µg/g) and kidney (12.33 µg/g) tissues of Rock Pigeon collected from Bangladesh. They also reported the levels of copper in muscle (26.09 µg/g), liver (43.6 µg/g) and kidney (34.77 µg/g) in Rock Pigeon from Bangladesh. Torres et al. (2010) reported the levels of copper in muscle (4.02 µg/g), kidney (3.41 µg/g) and liver (2.84 µg/g) tissues of Rock Pigeon in Spain. The levels recorded in the Rock Pigeon and Eurasian Collared in the present study were far less than the levels reported in previous studies stated above.

Jayakumar and Muralidharan (2011) documented the levels of copper in muscle (4.53 µg/g), liver (9.32 µg/g) and kidney (9.77 µg/g) tissues of Indian Pond Heron sampled from Nilgiris in Tamil Nadu, India. The levels of copper reported in same species in the present study were lower than the levels mentioned above. Lucia et al. (2010a) reported the levels of copper in muscle (43 µg/g), liver (92.5 µg/g) and kidney tissues (39.9 µg/g) of Graylag Goose in France and the copper level recorded in Bar-headed Goose in the present study were lower than the previous reports. The levels of copper in the liver tissues of birds in the present study were higher than the levels reported by Szymczyk and Zalewski (2003) in liver (7.08 µg/g) of Mallard Duck in Poland.

Migula and Augustyniak (2000) compiled the levels of copper in muscle (13.9 µg/g), liver (27.2 µg/g) and kidney tissues (18.8 µg/g) of a few Gull species in Poland. Sileo et al. (2004) reported the levels of copper in liver (21.0 µg/g) and kidney (150.0 µg/g) tissues of Mallard Duck from USA. They also reported copper in liver and kidney tissues of Goose in the US that ranged from 16 to 50 µg/g and 21 to 104 µg/g respectively. The levels of copper in the waterbirds of present study were lower than the previous studies. Van wyk et al. (2001) reported the levels of copper in muscle (18.23 µg/g),

liver (31.85 $\mu\text{g/g}$) and kidney (14.9 $\mu\text{g/g}$) in African White-backed Vulture sampled from South Africa. Further, Indian White-rumped Vulture documented by Rajamani and Muralidharan (2015) reported the levels of copper to have ranged from 6.53 to 28.92 $\mu\text{g/g}$ in kidney, 5.42 to 5.76 $\mu\text{g/g}$ in liver and 5.17 to 5.23 $\mu\text{g/g}$ in muscle of male and female Indian White-rumped Vulture. The levels of copper in the Indian White-rumped Vulture, Himalayan Vulture and Slender-billed Vulture in the present study were less than the above-referred values.

The raptors in the present study had lower levels of copper in muscle and liver tissues and, higher levels in kidney when compared to the study by Naccari et al. (2009) who reported copper in muscle (33.49 $\mu\text{g/g}$), liver (39.51 $\mu\text{g/g}$) and kidney (9.61 $\mu\text{g/g}$) in Common Buzzard from Italy. Deng et al. (2007) compiled the levels of copper in liver (37.47 $\mu\text{g/g}$), kidney (16.11 $\mu\text{g/g}$) and muscle tissues (13.66 $\mu\text{g/g}$) of Great Tit from China. They also reported the levels of copper in liver (27.01 $\mu\text{g/g}$), kidney (20.62 $\mu\text{g/g}$) and muscle (18.99 $\mu\text{g/g}$) tissues of Green Finch from China. The levels of copper recorded in the present study were higher than the levels reported in liver (9.32 $\mu\text{g/g}$) and muscle (4.53 $\mu\text{g/g}$) of Indian Pond Heron (Jayakumar and Muralidharan 2011).

Copper is an essential trace element that is particularly toxic to organisms and organs at high doses. High concentration of copper can also cause public health hazards (Brito et al. 2005). Copper concentrations in birds are usually elevated in areas of human activity due to intensive copper use (Eisler 2000). Variation in copper concentrations is not attributed to contamination or higher natural background levels of copper in the habitat of birds but, rather it could be due to species-specific metabolic bioaccumulation (Honda et al. 1986; Kim et al. 1996). High copper contamination has been reported in other wild birds without any signs of toxicity (Clausen and Wolstrup 1978; Kim et al. 1996). Acute copper poisoning was described in liver of Canada Goose *Branta canadensis* ranging from 187 to 323 $\mu\text{g/g}$ (Henderson and Winter field 1975). According to Pyle et al. (2005), copper concentrations in the liver are usually regulated by a homeostatic control below 150 $\mu\text{g/g}$ and can exceed this threshold, only if the control mechanisms are overloaded. However, the levels reported in the present study were lower than the levels that could cause problem among birds.

5.1.6. Levels of zinc concentrations (mean \pm SE, $\mu\text{g/g}$) in various species of birds collected dead between 2012 and 2014

Among the 31 species of birds included in the study, concentration of zinc was found to be the highest in kidney of Sarus Crane (193.67 $\mu\text{g/g}$), followed by Painted Stork (193.65 \pm 0.13 $\mu\text{g/g}$), Indian Peafowl (191.90 \pm 1.19 $\mu\text{g/g}$) and Himalayan Vulture (191.74 \pm 2.02 $\mu\text{g/g}$). The levels recorded in kidney of Slender-billed Vulture, Barn Owl, Spot-billed Pelican, Demoiselle Crane and liver of Demoiselle Crane, Indian Black Ibis, Painted Stork, and Indian Spotted Eagle were in the range of 190.11, 189.15, 188.08, 187.17, 188.19, 187.12, 188.14 and 187.50 $\mu\text{g/g}$ respectively. There were significant variations in zinc concentrations among species (One - way ANOVA $F=136.101$; $df=30$; $p<0.05$) included in the current study. The minimum levels of zinc were observed in muscle of Emerald Dove (69.23 $\mu\text{g/g}$) and Eurasian Collared Dove (73.72 $\mu\text{g/g}$) (Table 5.6).

Janaydeh et al. (2016) reported the levels of zinc in muscle (44.24 $\mu\text{g/g}$), liver (156.44 $\mu\text{g/g}$) and kidney (61.94 $\mu\text{g/g}$) of House Crow from Malaysia. Komosa et al. (2012) reported the levels of zinc in liver of Rook (124.92 $\mu\text{g/g}$) in Poland. Similarly, Horai et al. (2007) documented the levels of Zn in muscle (58.9 $\mu\text{g/g}$), liver (79.1 $\mu\text{g/g}$) and kidney tissues of (142 $\mu\text{g/g}$) Jungle Crow in Japan. They also reported the levels of zinc in muscle (54 $\mu\text{g/g}$), liver (122 $\mu\text{g/g}$) and kidney tissues (75.3 $\mu\text{g/g}$) in Carrion Crow sampled from Japan. Rajamani and Muralidharan (2015) reported the levels of zinc in male and female Indian White - rumped Vulture sampled from India and the levels ranged from 4.61 to 30.96 $\mu\text{g/g}$ in kidney, 35.91 to 42.52 $\mu\text{g/g}$ in liver and 21 to 21.95 $\mu\text{g/g}$ in muscle. The levels of zinc in Indian White-rumped Vulture included in the present study were 10 folds higher than the previous study. However, the levels still appear to be nontoxic.

Begum and Sherin (2013) reported the levels of zinc in muscle (145.69 $\mu\text{g/g}$), liver (239.75 $\mu\text{g/g}$) and kidney tissues (41.4 $\mu\text{g/g}$) of Rock Pigeon collected from Bangladesh. The levels of zinc in kidney (117.2 $\mu\text{g/g}$) of Rock Pigeon were also reported from Korea (Nam and Lee 2006). Beyer et al. (2004) documented the zinc level in liver (100 $\mu\text{g/g}$) and kidney (140 $\mu\text{g/g}$) of Mourning Dove collected from United States of America. Torres et al. (2010) reported the levels of zinc in muscle (12.22 $\mu\text{g/g}$), kidney (40.99 $\mu\text{g/g}$) and liver (25.02 $\mu\text{g/g}$) of Rock Pigeon from Spain. The levels of zinc recorded in the Rock Pigeon, Eurasian Collared Dove and Emerald Dove in present study were far less than the levels reported in previous studies.

Table 5.6 Levels of zinc concentrations (mean \pm SE, $\mu\text{g/g}$) in various organs and species of birds collected from 2012-2014

S. No.	Name of the species	Muscle	Liver	Kidney
1.	Bar-headed Goose	NA	152.37 \pm 1.05	166.87 \pm 3.66
2.	Indian Peafowl	171.20 \pm 4.71	183.46 \pm 1.16	191.90 \pm 1.19
3.	Emerald Dove	69.23	78.86	NA
4.	Eurasian Collared Dove	73.72 \pm 1.56	86.64 \pm 5.62	77.61 \pm 0.45
5.	Rock Pigeon	78.40 \pm 1.40	85.42 \pm 1.97	78.83 \pm 0.37
6.	Asian Koel	154.39 \pm 6.82	179.79 \pm 3.25	77.05
7.	Blue-faced Malkoha	172.32 \pm 0.82	176.19 \pm 1.22	185.89 \pm 4.71
8.	Common Coot	161.67 \pm 2.70	177.25 \pm 3.72	184.75 \pm 3.05
9.	Demoiselle Crane	163.82	188.19 \pm 0.63	187.17 \pm 0.96
10.	Sarus Crane	154.25 \pm 1.67	184.62 \pm 1.93	193.67
11.	Painted Stork	171.24 \pm 0.60	188.14 \pm 2.60	193.65 \pm 0.13
12.	Great White Pelican	NA	184.95	184.86
13.	Spot-billed Pelican	177.90 \pm 4.85	186.28 \pm 4.86	188.08 \pm 2.74
14.	Cattle Egret	126.05 \pm 0.70	147.18 \pm 1.93	143.58 \pm 0.94
15.	Indian Pond Heron	175.75 \pm 2.01	178.76 \pm 3.56	179.14 \pm 3.00
16.	Indian Black Ibis	175.94 \pm 1.86	186.78 \pm 2.24	187.12 \pm 2.96
17.	Red-wattled Lapwing	126.90 \pm 1.19	152.86 \pm 3.99	152.58 \pm 3.73
18.	Himalayan Vulture	185.43	181.43 \pm 1.21	191.74 \pm 2.02
19.	Indian White-rumped Vulture	170.89 \pm 1.03	182.12 \pm 1.77	183.29 \pm 3.73
20.	Slender-billed Vulture	NA	182.84	190.11
21.	Griffon Vulture	172.84 \pm 0.54	183.13 \pm 4.23	177.25 \pm 2.98
22.	Indian Spotted Eagle	175.78	187.50	182.22
23.	Shikra	176.40 \pm 0.57	185.82 \pm 1.02	186.94 \pm 1.81
24.	Black Kite	175.47 \pm 1.10	181.66 \pm 1.76	181.40 \pm 2.31
25.	Barn Owl	183.28 \pm 1.26	185.25 \pm 1.84	189.15 \pm 2.10
26.	Indian Roller	127.21	146.30	145.86
27.	Common Kingfisher	168.50	157.99	159.8
28.	Rose-ringed Parakeet	81.89 \pm 4.50	85.71 \pm 4.50	91.81 \pm 5.40
29.	Indian Pitta	126.50	143.64	140.39
30.	Black Drongo	123.74 \pm 1.82	141.68	140.03
31.	House Crow	168.07 \pm 4.40	179.11 \pm 2.11	185.46 \pm 3.87

Jayakumar and Muralidharan (2011) documented the levels of zinc in muscle (21.31 $\mu\text{g/g}$), liver (4.74 $\mu\text{g/g}$) and kidney tissues (14.03 $\mu\text{g/g}$) of Indian Pond Heron and, the levels recorded in the present study were lower than these values. Lucia et al. (2010a) reported the levels of zinc in muscle (93.8 $\mu\text{g/g}$), liver (355.8 $\mu\text{g/g}$) and kidney (189.2 $\mu\text{g/g}$) of Graylag Goose from France. Further, Hoffman et al. (2000) reported the levels of zinc in liver (110 to 180 $\mu\text{g/g}$) and kidney (50 to 59 $\mu\text{g/g}$) of Canada Geese in USA. The Bar-headed Goose the only Goose species included in the study showed higher levels than the previous studies. Migula et al. (2000) compiled the levels of zinc in muscle (198.2 $\mu\text{g/g}$), liver (235.2 $\mu\text{g/g}$) and kidney tissues (208 $\mu\text{g/g}$) of a few species of gulls in Poland. Sileo et al. (2004) reported the levels of zinc in liver (280 $\mu\text{g/g}$) and kidney (220 $\mu\text{g/g}$) in Mallard Duck from USA. The levels of zinc in all the species of waterbirds in the present study were lower than the earlier reported values except Mallard Duck (24.288 $\mu\text{g/g}$) from Poland (Szymczyk and Zalewski 2003).

Concentrations of zinc noted from Indian White-rumped Vulture in the present study were much higher than the levels reported in muscle (19.65 $\mu\text{g/g}$), liver (32.21 $\mu\text{g/g}$) and kidney tissues (150.03 $\mu\text{g/g}$) of African White-rumped Vulture from South Africa (Van wyk et al. 2001). Naccari et al. (2009) reported the levels of zinc in muscle (52.76 $\mu\text{g/g}$), liver (137.51 $\mu\text{g/g}$) and kidney (66.54 $\mu\text{g/g}$) of Common Buzzard in Italy. Raptorial bird species analyzed from the present study had higher levels of zinc in liver and kidney than the previous studies (Van wyk et al. 2001; Naccari et al. 2009). Deng et al. (2007) compiled the levels of zinc in liver (117.15 $\mu\text{g/g}$), kidney (85.5 $\mu\text{g/g}$) and muscle (32.15 $\mu\text{g/g}$) of Great Tit in China. They also reported the levels of zinc in liver (105.89 $\mu\text{g/g}$), kidney (88.05 $\mu\text{g/g}$) and muscle (31.37 $\mu\text{g/g}$) in Green Finch. Abduljaleel (2014) reported the levels of zinc in liver (63.17 $\mu\text{g/g}$) of Quail in Iraq. The levels of zinc in muscle (4.57 $\mu\text{g/g}$), liver (20.01 $\mu\text{g/g}$) and kidney tissues (16.02 $\mu\text{g/g}$) of shot Pheasants have been documented in Poland (Dzungan et al. 2012). Zinc levels recorded in the present study were comparable and find literature supports. In birds, the threshold level of zinc toxicity was reported to be 1200 $\mu\text{g/g}$ (Gasaway and Buss 1972). Also, poisoning signs of Zn concentrations (840.0 and 1410.0 $\mu\text{g/g}$) were reported in liver of Mallard ducks in US (Levengood et al. 1999; Sileo et al. 2003). Taggart et al. (2009) reported the zinc concentrations in liver tissues to be poisonous at 525 $\mu\text{g/g}$.

The levels of Zn recorded in liver tissues of birds in the present study were lower than the threshold levels and not indicative of Zn poisoning.

Zinc, copper and nickel are the essential metals analyzed in the current study, and the levels were within normal ranges maintained by regular homeostatic mechanisms of birds. During the flight, birds utilize energy from their muscles and the extent of accumulation of metals in liver and kidney tissues were usually high due to the presence of a protein called 'metallothionein' (Wu et al. 2000). Indeed, this protein attaches to metals in the body mostly in the liver and kidney, captivates them and prevents their entrance in to the blood circulation, in order to finally eliminate them under favorable conditions from the body. Zinc is an essential metal required to support biological activities, but when their environmental concentrations rise, they can generate serious toxicological problems (Pe´rez-Lo´pez et al. 2008).

Spot-billed Pelican, a Near Threatened species had the maximum concentration of both cadmium and zinc. Cadmium occurs as a minor component in most zinc ores, and therefore are a byproducts of Zn production. In birds, metals are not equally distributed in all the tissues, which compare well with the findings of Rahman et al. (2012). Ohlendorf et al. (1985) and Sinkakarimi et al. (2018) studied the tissues which contained the highest concentration of metals such as lead, cadmium. Several studies in the past have shown the synergistic toxic effects of metals and pesticides (Ohlendorf et al. 1985; Sinkakarimi et al. 2018). Such evaluations have revealed the synergistic interactions of various metals and pesticides in wildlife. The synergistic effects of Diazinon and cadmium were different from estimated in comparison to the effects of these compounds separately (Feron and Groten 2002).

Doneley (1992) reported moderate to severe nephrosis in caged and aviary birds containing hepatic Zinc levels of 320 and 534 µg/g respectively. Concentrations in liver were 473 to 1990 µg/g, the toxic threshold at which clinical signs of zinc poisoning were observed in mallards (Levengood et al. 1999). Taggart et al. (2009) reported that zinc concentrations are poisonous when the level reached 525.0 µg/g in liver. Also, poisoning signs were reported in mallards with 840 µg/g of zinc in liver (Sileo et al. 2003) and 1410 µg/g (Levengood et al. 1999). Sileo et al. (2003) diagnosed Zn poisoning in wild

waterfowls with liver concentration which ranged from 280.00 to 2900.00 $\mu\text{g/g}$. Levels in all the species of birds analyzed in the present study were found to be lower than the levels that could cause poisoning among birds.

5.1.b Levels of metals in different tissues of birds

To understand as to which tissue accumulates metals the maximum, data were compiled species wise. The levels of total metals varied significantly among the organs (One-way ANOVA $F=71.223$; $df=2$; $p<0.05$). Among the three organs analyzed, kidney had the highest concentration of total metals followed by liver, while muscle showed the least accumulation (Figure 5.2). Similarly, the levels of zinc and copper were found to be the maximum in kidney than that of liver and muscle, while the levels of nickel, lead, cadmium and chromium recorded were relatively similar in all the organs (Figure 5.3). Kidney has the highest level of most metals analyzed and remains a good indicator for metals in internal tissues of birds (Donia 2015; Yohannes et al. 2017)

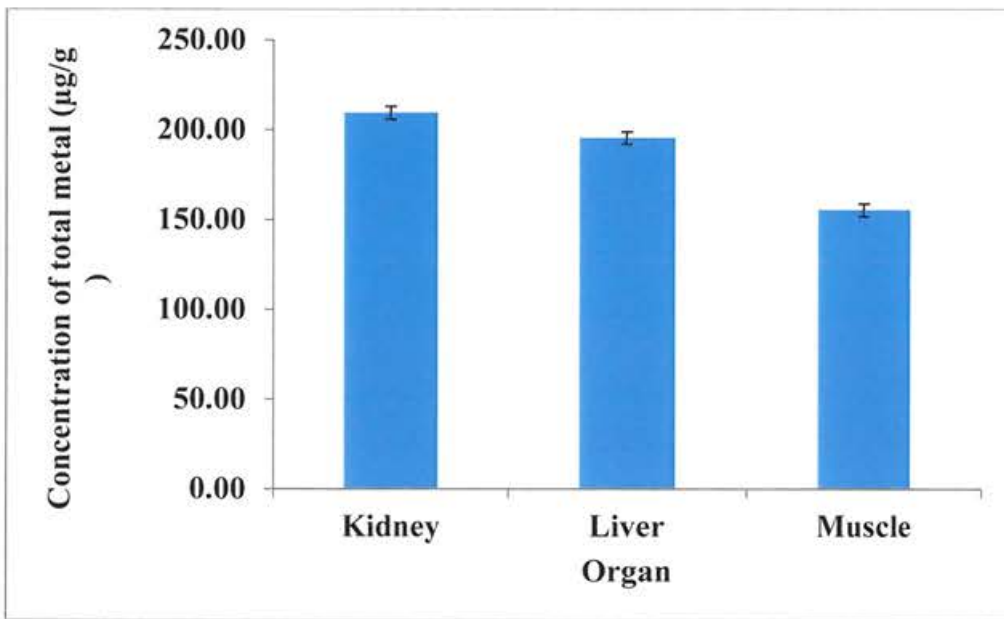


Figure 5.2 Variation in total metal concentration among different organs of birds collected between 2012 and 2014

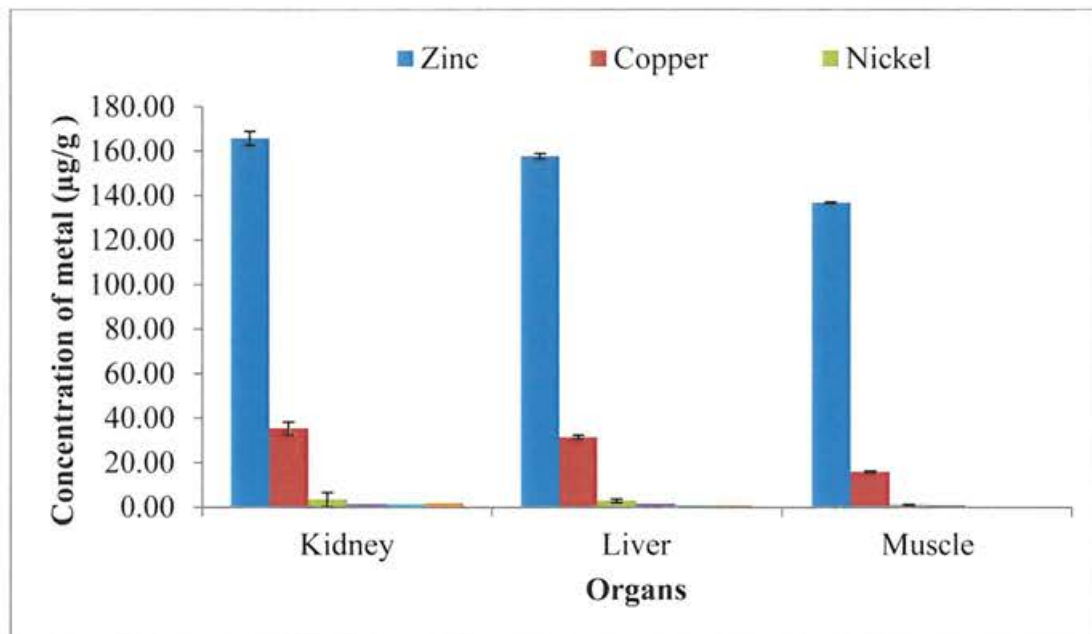


Figure 5.3 Variation in levels of individual metals among different tissues of birds collected between 2012 and 2014

Yohannes et al. (2017) reported the lead accumulation to vary among different tissues, which is comparable to be with the findings of the present study. Binkowski and Meissner (2013) reported the highest concentration of cadmium to be in kidney tissues which indicates efficiency of the detoxification process. It is to be noted that the kidney is the main internal organ targeted for lead detoxification and bioaccumulation (Franson 1996). Kidney, followed by liver, was the main organ for cadmium accumulation in all the species of birds studied earlier (Gomez et al. 2004; Lucia et al. 2010b; Yohannes et al. 2017).

Maximum levels of cadmium accumulation in kidneys of aquatic birds have been reported by many researchers (Nam et al. 2005; Kojadinovic et al. 2007; Lucia et al. 2010a; Yohannes et al. 2017). This demonstrates the role of kidney in the detoxification process and storage of nonessential elements as reported elsewhere (Nam et al. 2005; Lucia et al. 2009 and 2010a). The muscle is generally considered to have a weak accumulating potential as reported elsewhere (Bervoets and Blust 2003; Erdoğrul and Erbilir 2007; Uysal et al. 2009; Uluozlu et al. 2009).

White et al. (1978) and Goyer et al. (1989) suggested that as cadmium concentration in kidney drastically falls after cadmium induced tubular dysfunction,

kidney does not always reflect the actual concentration of cadmium. On the contrary, with more stable level of cadmium in liver which is resistant towards the toxic effects of cadmium, liver is the best indicator of contamination among birds. Therefore, to make it fool proof, kidney and liver may have to be analyzed for finding the accumulation of cadmium in birds.

Toxic metals such as lead and cadmium are most often found in the liver and kidney tissues of birds due to temporary deposition and the role of these organs in detoxification (Kalisińska and Salicki 2010). The renewal of liver cells during intoxication leads to the release of these conjugates into the systemic circulation, where they are subsequently delivered to the kidneys (Thevenod 2003) for elimination. During chronic intoxication, the concentration of metal-binding proteins increases in the blood. Metals are also known to be potent inducers of metallothionein and glutathione in renal and liver tissues (Zalups 2000).

Assimilated Ni is excreted primarily in the urine (Underwood 1977; Merritt et al. 1989), whereas excretion in the bile is of less importance (Marzouk and Sunderman 1985). Lock et al. (1992) and Elliott (2005) also proved an element accumulates high in kidney of Flesh-footed Shear waters. The proportional increase in Zn concentrations in kidneys has been shown to be greater than that in livers of dosed birds (Gasaway and Buss 1972; Levengood et al. 1999; Day et al. 2003). The similar trend was also noted in birds of the present study.

5.1.c Levels of metals in gizzard of various species of birds

A total of 127 samples of gizzard comprising 24 species of birds were collected and analyzed for metal contaminations. The concentration of total metals recorded in gizzard varied significantly among the species (One-way ANOVA, $df= 30$; $F= 77.270$; $p<0.05$). Among the species analyzed, the maximum levels of total metal were recorded in Spot-billed Pelican ($250.10 \pm 31.97 \mu\text{g/g}$) followed by Blue-faced Malkoha ($220.17 \pm 10.87\mu\text{g/g}$), Indian Black Ibis ($219.64 \pm 10.97 \mu\text{g/g}$) and Painted Stork ($214.92 \pm 5.81 \mu\text{g/g}$). The concentration of total metal ranged from 201.75 to 212.58 $\mu\text{g/g}$ in eight species of birds analyzed. The levels of total metals in eleven species of birds, namely House Crow, Indian White-rumped Vulture, Eurasian Vulture, Common Coot, Indian Pitta, Red-wattled Lapwing, Cattle Egret, Black Drongo, Indian Roller, Eurasian Collared Dove and Rock

Pigeon ranged from 105.59 to 199.90 $\mu\text{g/g}$, while the minimum level was observed in Emerald Dove ($95.50 \pm 9.42 \mu\text{g/g}$) (Table 5.7).

Among the 24 species of birds studied, the maximum concentration of cadmium was recorded in gizzard of Common Kingfisher ($4.05 \mu\text{g/g}$) followed by Spot-billed Pelican ($3.92 \pm 2.07 \mu\text{g/g}$) and Sarus Crane ($3.04 \pm 0.13 \mu\text{g/g}$), while the least concentration was noted in Asian Koel (Table 5.8). The levels of cadmium detected varied significantly among the species (One-way ANOVA $F=4.979$; $df=23$; $p<0.05$). The maximum concentration of lead was recorded in Painted Stork ($1.43 \mu\text{g/g}$) followed by Spot-billed Pelican and Black Drongo. Significant variation in lead levels was observed among the species (One-way ANOVA $F=2.692$; $df=23$; $p<0.05$). Among the species analyzed, the Common Kingfisher had the highest concentration of chromium ($4.63 \mu\text{g/g}$) followed by Spot-billed Pelican and Indian Black Ibis, whereas the lowest was observed in Indian Roller. The maximum concentration of Nickel was recorded in Blue-faced Malkoha ($7.78 \mu\text{g/g}$) followed by Spot-billed Pelican and Indian White-rumped Vulture. The highest concentration of copper was recorded in Spot-billed Pelican ($54.05 \mu\text{g/g}$) followed by Indian Black Ibis and Blue-faced Malkoha. There was a significant variation in zinc residues among the species (One-way ANOVA $F=77.270$; $df=23$; $p<0.05$). A highest concentration of zinc was observed in Spot-billed Pelican ($181.43 \mu\text{g/g}$) followed by Barn Owl ($179.98 \mu\text{g/g}$), whereas the least was noted in Emerald Dove (Table 5.8).

The levels of cadmium recorded in birds in the present study were comparable with levels reported in India and elsewhere. Levengood et al. (1999) reported Cd in gizzard ($0.08 \mu\text{g/g}$) of Mallard in USA. The levels of Cd reported in the present study were comparably higher in waterfowls namely Common Kingfisher, Spot-billed Pelican and Sarus Crane.

The levels of lead in gizzard contents ($0.41 - 0.79 \mu\text{g/g}$) in Rock Pigeon were reported from Bangladesh (Begum and Sherin 2013). Similarly, Abduljaleel et al. (2012) reported the levels of Pb in gizzard ($0.75 \mu\text{g/g}$) of Quail from Malaysia. The levels of lead recorded in Rock Pigeon ($0.31 \mu\text{g/g}$) in the present study were lower than the levels reported previously.

Godwin et al. (2016) reported the levels of chromium in gizzard contents (9.96 $\mu\text{g/g}$) of Tree Swallow from Canada and is comparable with the levels of Cr in gizzard of Common Kingfisher (4.63 $\mu\text{g/g}$) and Spot-billed Pelican (4.17 $\mu\text{g/g}$) included in the present study.

Godwin et al. (2016) reported the levels of nickel in gizzard contents (2.03 $\mu\text{g/g}$) of Tree Swallow. The levels of nickel observed in the gizzard of Indian White-rumped Vulture and Indian Black Ibis were higher than the Tree Swallow sampled from Canada.

Abduljaleel et al. (2012) reported in gizzard contents (52.89 $\mu\text{g/g}$) of Quail from Malaysia which comparable with the levels of copper in gizzard contents of Spot-billed Pelican (54.05 $\mu\text{g/g}$) included in the present study.

The levels of zinc recorded in birds included in the present study are comparable with the levels reported in India and elsewhere. Begum and Sherin (2013) reported the levels of zinc in gizzard (124.79 $\mu\text{g/g}$) of Rock Pigeon in Bangladesh. Rock Pigeon in the present study had 84.48 ± 0.08 $\mu\text{g/g}$ of zinc and was lower than the previous study. Except Eurasian Collared Dove (85.19 ± 1.40 $\mu\text{g/g}$) and Emerald Dove (75.60 ± 2.75 $\mu\text{g/g}$), the levels of zinc in present study were higher than the levels reported by Abduljaeel et al. 2012 of Zn (118.93 $\mu\text{g/g}$) in Quail collected from Malaysia.

Considering the levels of metals in tissues and gizzard of different bird species from four states, essential metals, namely nickel, copper and zinc were found to be high and non-essential metals, namely chromium, lead and cadmium were found to be less. The levels observed in the current study shows the accumulation pattern to be indicative of consistent exposure and hence, regular monitoring is suggested.

Table 5.7 Variation of metal concentrations (mean \pm SE, $\mu\text{g/g}$) in gizzard of birds collected from 2012 to 2014.

S. No.	Species	N	Zinc	Copper	Nickel	Chromium	Lead	Cadmium	TM
1	Asian Koel	03	176.96 \pm 0.76	24.24 \pm 1.69	0.22 \pm 0.05	0.21 \pm 0.04	0.11 \pm 0.02	0.01 \pm 0.01	201.75 \pm 2.13
2	Barn Owl	02	179.98 \pm 3.40	19.68 \pm 2.53	1.68 \pm 0.62	0.26 \pm 0.15	0.06 \pm 0.0	0.12 \pm 0.07	201.77 \pm 5.52
3	Black Drongo	02	125.37 \pm 0.02	20.67 \pm 1.48	0.85 \pm 0.45	2.61 \pm 1.32	1.31 \pm 0.66	2.56 \pm 2.47	153.35 \pm 2.54
4	Black Kite	10	177.51 \pm 1.03	22.00 \pm 0.7	1.41 \pm 0.40	1.12 \pm 0.57	0.07 \pm 0.01	0.18 \pm 0.05	202.29 \pm 1.32
5	Rock Pigeon	13	84.48 \pm 0.08	19.57 \pm 1.73	0.71 \pm 0.18	0.45 \pm 0.16	0.31 \pm 0.06	0.06 \pm 0.01	105.59 \pm 2.01
6	Blue-faced Malkoha	06	176.83 \pm 1.04	33.80 \pm 7.15	7.78 \pm 6.23	1.29 \pm 0.48	0.25 \pm 0.14	0.23 \pm 0.16	220.17 \pm 10.87
7	Cattle Egret	18	126.73 \pm 0.79	25.70 \pm 2.48	1.03 \pm 0.24	1.23 \pm 0.16	0.54 \pm 0.19	0.98 \pm 0.27	156.21 \pm 2.93
8	Common Coot	07	156.48 \pm 13.86	22.48 \pm 0.65	0.24 \pm 0.06	0.24 \pm 0.06	0.12 \pm 0.03	0.23 \pm 0.06	179.79 \pm 13.61
9	Common Kingfisher	01	160.39	30.19	1.69	4.63	0.85	4.05	201.80
10	Emerald Dove	02	75.60 \pm 2.75	17.41 \pm 7.36	0.17 \pm 0.12	1.43 \pm 0.42	0.70 \pm 0.10	0.21 \pm 0.05	95.50 \pm 9.42
11	Eurasian Collared Dove	14	85.19 \pm 1.40	22.14 \pm 1.20	1.09 \pm 0.97	0.76 \pm 0.33	0.09 \pm 0.02	0.26 \pm 0.08	109.53 \pm 1.50
12	Griffon Vulture	03	172.64 \pm 1.23	21.32 \pm 0.62	1.05 \pm 0.08	0.26 \pm 0.10	0.52 \pm 0.04	0.08 \pm 0.03	195.87 \pm 1.89
13	House Crow	05	177.34 \pm 2.96	20.35 \pm 2.63	0.90 \pm 0.28	0.26 \pm 0.08	0.23 \pm 0.07	0.82 \pm 0.30	199.90 \pm 2.28
14	Indian Black Ibis	06	162.07 \pm 0.41	48.53 \pm 8.80	4.43 \pm 3.86	2.87 \pm 0.53	0.55 \pm 0.27	1.20 \pm 0.71	219.64 \pm 10.97

S. No.	Species	N	Zinc	Copper	Nickel	Chromium	Lead	Cadmium	TM
15	Indian Peafowl	03	174.04±1.61	25.22±1.81	1.16±0.42	0.68±0.34	0.71±0.30	0.14±0.01	201.96±3.17
16	Indian Pitta	01	126.25	32.63	1.50	1.09	0.75	1.40	163.62
17	Indian Pond Heron	07	177.24±2.98	23.26±0.69	0.83±0.22	0.36±0.16	0.43±0.11	0.09±0.03	202.21±2.92
18	Indian Roller	01	128.08	21.04	0.56	0.09	0.28	0.12	150.17
19	Indian White-rumped Vulture	02	171.62±0.44	17.17±3.86	4.59±4.14	1.49±1.37	0.31±0.08	1.96±1.83	197.14±3.12
20	Painted Stork	04	175.65±0.69	33.60±4.02	1.07±0.62	2.15±0.93	1.43±0.46	1.02±0.67	214.92±5.81
21	Red-wattled Lapwing	03	128.20±0.51	27.60±1.24	1.69±0.49	2.38±0.55	0.14±0.07	2.41±0.54	162.42±2.18
22	Sarus Crane	02	173.66±0.35	26.83±5.18	1.28±0.55	1.64±0.09	0.27±0.05	3.04±0.13	206.71±6.33
23	Shikra	05	174.28±1.58	29.64±9.00	3.12±1.35	2.33±0.54	0.26±0.10	2.95±0.78	212.58±10.88
24	Spot-billed Pelican	03	181.43±3.67	54.05±23.50	5.12±1.49	4.17±1.39	1.41±0.76	3.92±2.07	250.10±31.97
One-way ANOVA		F Value	77.270	2.811	0.892	4.068	2.692	4.979	33.727
		P Value	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05

TM- Total Metal Load

5.1.d Variation in metal contamination among birds with different feeding habits.

Among the 31 species studied, 30 were grouped into six feeding guilds such as granivore (04), herbivore (02), insectivore (05), piscivore (04), carnivore (10) and omnivore (06) excluding Rose-ringed Parakeet (frugivore) due to the lack of samples in the guild. The grouping was done based on their food habits as mentioned in Ali and Ripley (1987). Organs of each individual were pooled to find out variation among feeding guilds.

Among the bird species analyzed, carnivorous birds had the highest concentration of total metal load followed by omnivorous and piscivorous birds. The levels of total metal load detected in insectivorous and herbivorous birds were 167.60 and 114.23 $\mu\text{g/g}$ respectively (Figure 5.4). The lowest level of total metal was observed in granivorous birds. Similarly, the levels of zinc and copper were recorded maximum in carnivorous birds followed by omnivorous birds. The amount of cadmium, lead, chromium and nickel were detected more or less similar among all feeding guilds (Figure 5.5).

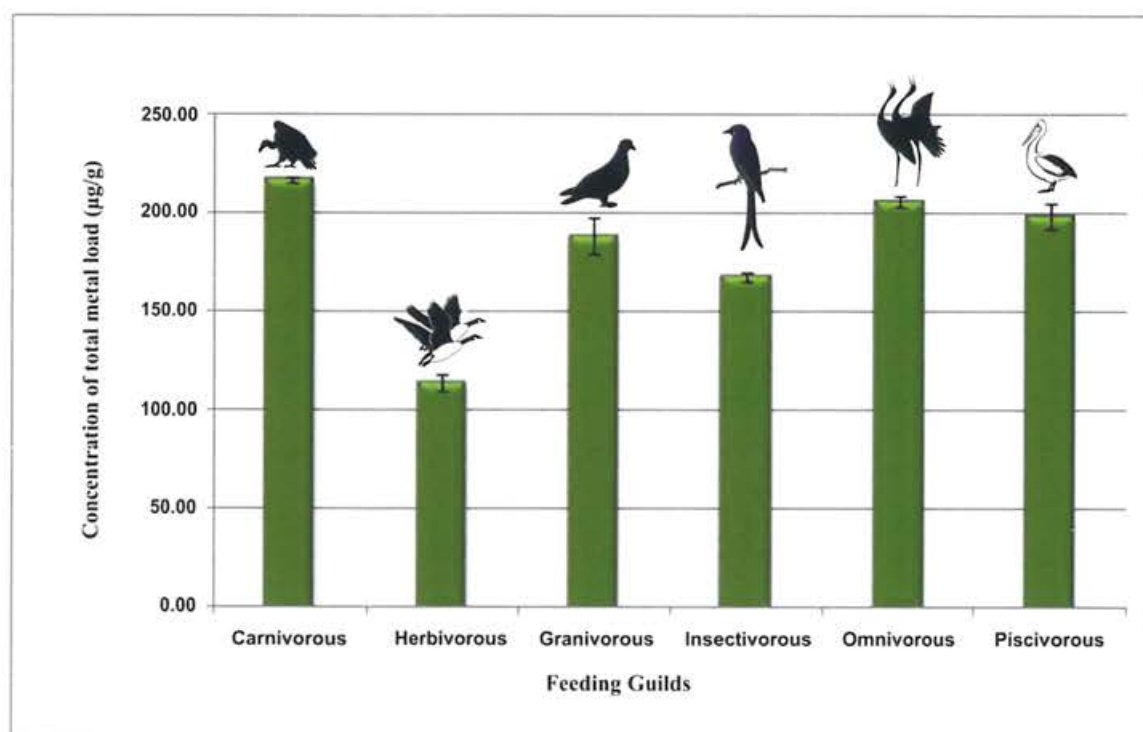


Figure 5.4 Variation in total metal contamination among birds belonging to different feeding guild (One-way ANOVA $F= 341.509$; $df=5$; $p<0.05$).

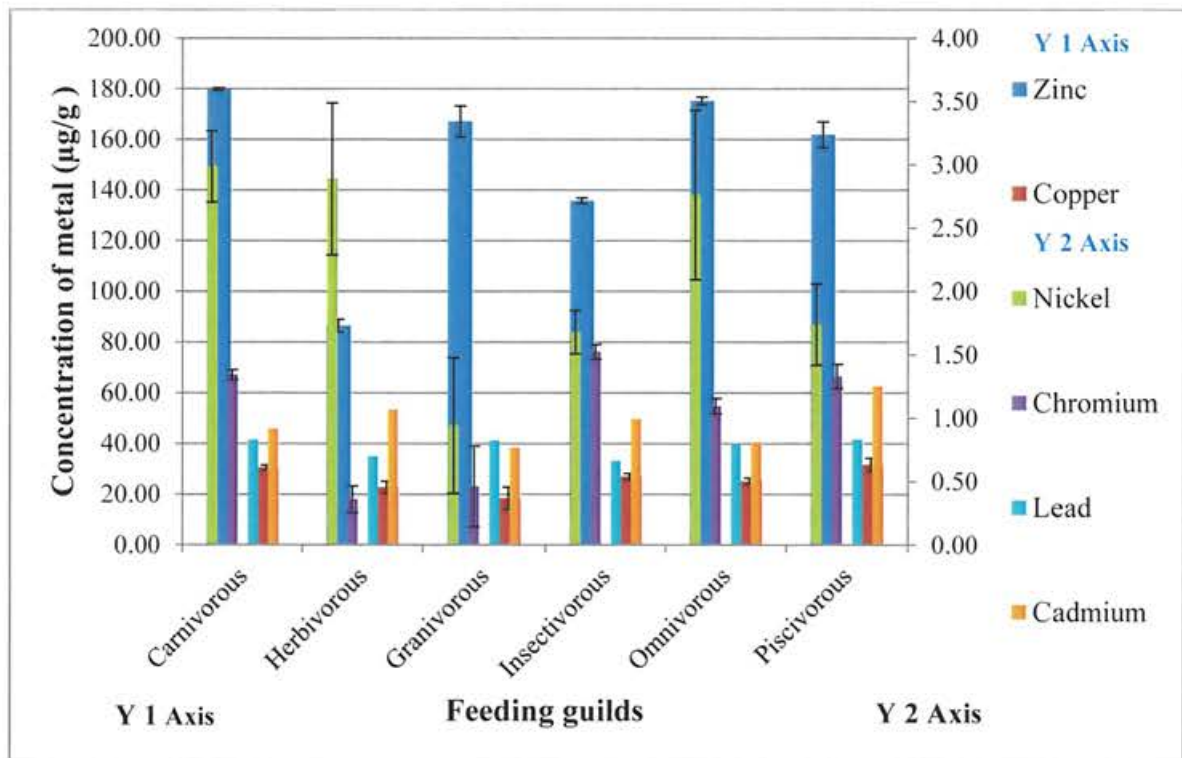


Figure 5.5 Variation of individual metals analysed among birds belonging to various feeding guilds collected between 2012 and 2014

These food items may transmit high doses of pesticides and metals from agrochemicals (Beyerbach et al. 1987; Purchart and Kula 2007). Cadmium concentration in nestlings of Rooks, including nestlings without any ingested food from several rookeries in north-eastern Poland was high in the tissues (liver, kidney, lung, muscles and bones; ranged between 15.52 & 18.8 µg/g) of the adult birds (Orłowski et al. 2012). Further, the levels of Cd in kidney was positively correlated with the number of food items (primarily ground-dwelling Coleoptera and their larvae) present in their gizzard content (Orłowski et al. 2013). Metals get bio-accumulated in the tissues of animals that are final consumers (Bilandzic et al. 2011). Cadmium is also known for its ability to bioaccumulate in some terrestrial food chains (Lodenijs 1990).

Kramer and Redig (1997) found that the prevalence of lead poisoning in eagles did not decrease after the implementation of 1991 non-toxic shot regulations for hunting waterfowl and American Coots *Fulica americana* in the United States. The authors suggested that carcasses of animals still hunted with lead ammunition, such as small

mammals and birds, and deer, could be additional sources of exposure (Kramer and Redig 1997). Lead poisoning of wintering Bald Eagles and Golden Eagles *Aquila chrysaetos* were due to secondary poisoning, as they feed on lead shot dead waterfowls and lead exposure and poisoning has been reported in a various eagle species and other raptors throughout the world (Pain et al. 2009). Studies carried out by Garcí'a-Ferna'ndez et al. (1997) indicate higher tissue concentrations of Pb in carnivorous birds particularly raptors than in fish-eating, insectivorous, omnivorous and carrion-eating birds, probably because some of these species live in areas where there is human activity.

The published records of 37 wild Bald Eagles that died of lead poisoning reported lead residues of 5-61 $\mu\text{g/g}$ in liver and 5 and 12 $\mu\text{g/g}$ in the kidneys tested (Mulhern et al. 1970; Kaiser et al. 1980; Reichel et al. 1984; Frenzel and Anthony 1989; Craig et al. 1990; Langelier et al. 1991; Gill and Langelier 1994). Mean lead concentrations in liver and kidney of two Steller's Sea - Eagles and three White-tailed Sea - eagles with lead bullet fragments in their stomachs were 92 and 156 mg/kg, respectively (Iwata et al. 2000). Five White-tailed Sea - eagles diagnosed with lead poisoning had mean lead concentrations of 22.4 and 9.6 mg/kg in liver and kidney respectively (Krone et al. 2004; 2006 and 2009).

Top-level piscivorous birds are liable to consume prey containing high level of pollutants and Hence, can accumulate higher levels of contaminants than birds that are placed lower on the food chain (Burger and Gochfeld 2002). Abbasi et al. (2015) reported piscivorous birds with 2.90 $\mu\text{g/g}$ and granivorous birds with 0.83 $\mu\text{g/g}$ of lead from Pakistan.

Dietz et al. (2000) had reported that Pb does not accumulate thorough trophic levels in terrestrial or marine ecosystem. Lead accumulation in considerable quantities also holds a certain threat for organisms at the end of the food-chain, including humans (Kalisí'nska and Salicki 2010). A similar pattern of metal accumulation was found in diverse organisms in ecosystem, where Pb is not biomagnified along the successive trophic levels of the food chain (Szefer 1991; Barwick and Maher 2003). Larger raptors select larger prey than smaller raptors and are more likely to ingest more lead (Pain et al. 1993; Garcia-Fernandez et al.1997).

Lead levels in soft tissue associated with mortality in doves and pigeons ranged between 20 and 60 $\mu\text{g/g}$ or even higher, as in the case of a wild Mourning Dove that died of lead poisoning with 72 $\mu\text{g/g}$ in the liver (Locke and Bagley 1967). Clausen and Wolstrup (1979) reported liver and kidney with lead residues of 48 and 200 $\mu\text{g/g}$ respectively in a wood pigeon (*Columba palumbus*) that died due to lead shot. Mourning Doves that died due to consumption of shot had mean lead residues of 80-93 $\mu\text{g/g}$ in the liver, 230-300 $\mu\text{g/g}$ in the kidney, and 116-192 $\mu\text{g/g}$ in bone (Buerger et al. 1986). In another lead shot dosing study, a Mourning Dove that died had lead residues of 267 $\mu\text{g/g}$ in the liver, 1901 $\mu\text{g/g}$ in the kidney, 11 $\mu\text{g/g}$ in the brain, and 403 $\mu\text{g/g}$ in bone (Kendall et al. 1983).

Bargagli et al. (1998) studied metal concentrations in a food web in the Mediterranean Sea and found that at high trophic levels, Cd concentrations are lower than that were observed at the bottom of the food chain, concluding that there is no evidence of biomagnification of Cd in this marine food chain. Similarly, Barwick and Maher (2003) found no evidence of magnification of Cd in a temperate estuarine ecosystem from Australia.

Copper and zinc levels in fish-eating coastal birds including pelican, Great Cormorants, and Herons, which stand on top of the food chain were reported to be very high (Zamani-Ahmadmahmoodi et al. 2009). The birds *Pelecanus occidentalis* and *Phalacrocorax brasilianus* (Neotropic Cormorant), are known to consume extensively fish and shrimp. The levels of Cd, Pb and Zn in fish were 0.3; 1.0; 18.4 $\mu\text{g/g}$ respectively. Similarly, shrimp, the levels were (Cd-0.5; Pb-0.9; Zn-61 $\mu\text{g/g}$). The Brown Pelican Cd-0.7.; Pb-4.2; Zn-23.3 $\mu\text{g/g}$ and *Neotropic Cormorant*, (Cd-1.2; Pb-1.7; Zn-35 $\mu\text{g/g}$) had levels which were higher than the above referred levels of Cd, Pb and Zn in fish and shrimp from Mexico (Meji'a-Sarmiento., 2001). It proves that the concentration of metals were transferred through food chain, and depends upon the feeding guild.

There was a gradual increase of levels from granivorous, piscivorous and carnivorous in the present study similar to the study in Japan by Horai et al. (2007). They also reported the levels of metals in various species of birds collected from Japan. The levels of Cr in Feral Pigeon, Grey Heron and Black Kite were 0.20 $\mu\text{g/g}$; 0.38 $\mu\text{g/g}$

and 0.39 µg/g respectively. Similarly the levels of Cu were 48.6 µg/g; 7.91 µg/g and 16.1 µg/g in Feral Pigeon, Grey Heron and Black Kite respectively.

The concentrations of metals in omnivorous birds, including *corvids*, have been rarely investigated. However, some remarkable comparative data on the *corvids* in Japan were provided by Horai et al. (2007), who studied Zn and Cu concentrations in the liver tissues of the Jungle Crow, *Corvus macrorhynchos*. In Poland, Komosa et al. (2012) studied the accumulation of metals (Cu, Zn, and Pb) in the liver tissues of omnivorous species such as Rook *Corvus frugilegus*. Dos Santos et al. (2006) found a small increase of Zn content with increasing trophic level that could be an evidence of biomagnification.

Binkowski and Meissner (2013) reported that the copper concentrations in tissues of Black Kites to be lower in muscles compared to liver as reported by earlier researchers. As body weight increased, concentration of copper also increased in muscles followed by liver. The concentration of copper in tissues especially, liver is regulated below 50 µg/g by homeostatic control (Pyle et al. 2005). Animal flesh is the best source of readily bio available zinc (Soriano-Santos 2010) with red meat being the richest. Black Kite *Milvus migrans govinda* is neither an aviary bird nor it feeds on red meat (it gets plenty of poultry left overs i.e. white meat in urban area). The possibility of any zinc toxicity in Black Kite is remote in the present environmental conditions. Hence, monitoring contamination in food web is necessary.

5.1.e Spatial variations in the levels of metal contamination among various species of birds

Since the sampling method adopted in this study was opportunistic, the possibility of getting the same species of dead birds from all states was remote. Hence, the levels of metals were pooled irrespective of species and the comparisons were made among three states, namely Gujarat, Tamil Nadu and Kerala to get a rough idea. Since, same species and adequate individuals could not be obtained from Assam it was not possible to include the state for the comparison. Overall, Gujarat (181.94 ± 2.87 µg/g) had the highest load of metal contamination, while Tamil Nadu (176.75 ± 3.98 µg/g) the least. The variation in total metals in the birds among states were significant (One-way ANOVA $F= 8.78$; $df = 3$; $p < 0.05$). Among the states, birds collected from Gujarat had the maximum

concentration of zinc ($181.85 \pm 1.93 \mu\text{g/g}$), whereas the least was recorded in Tamil Nadu ($144.03 \pm 3.52 \mu\text{g/g}$). The maximum level of copper was recorded in Tamil Nadu ($27.13 \pm 1.00 \mu\text{g/g}$) and Kerala ($27.02 \pm 1.49 \mu\text{g/g}$), while Gujarat had the least concentration ($26.22 \pm 0.88 \mu\text{g/g}$). The highest level of Nickel was recorded in Tamil Nadu ($2.78 \pm 0.42 \mu\text{g/g}$) and the lowest levels were noted in Kerala ($1.19 \pm 0.16 \mu\text{g/g}$). The levels of chromium, lead and cadmium analyzed in birds did not show any variation among states (Table 5.8).

Due to their industrial use, metals can be present in the environmental and are available for bio-magnification through air, water and food and through food-chain steps (Lagadic et al. 1998). As a result of bioaccumulation, birds can accumulate higher levels of metals, and it is known that once metals enter a bird, they can be stored in internal tissues such as the kidney and the liver (Braune and Gaskin 1987a; 1987b), which compares well with the present findings. In the present study, the levels of metals recorded higher in birds' species sampled from the states with higher industrial pressures.

Industrial and mining activities continuously contaminate the surface water with a variety of toxic materials including metals. Such compounds reported to degrade the physicochemical properties of environment and subsequently different species of birds inhabiting the environment. Rajamani and Muralidharan (2015) reported that the vulture from Gujarat had the maximum metal contamination than any other state. Further, Wastewater from industries and cities are used for irrigation without adequate treatment which is causing accumulation of metals in birds which use these habitats. Ahmedabad, Vadodara, and Surat are declared as a critical city in Gujarat. Industrial effluents are directly discharged into major rivers such as Tapi, Sabarmati, Amalkhadi and Kharicut canal. In Vadodara, about 45 borewells are being contaminated by industrial effluents. Data availability for industrial effluents is very hazy. There is a vast data gap on quantum of effluent generated and without this data it is not viable to draw the future plan for industrial waste management. The high accumulation of metals in the birds in Gujarat may be due to high levels of contamination due to the presence of different sources of industrial pollutions. The remaining states, namely Tamil Nadu and Kerala have relatively lesser industrial activities so as the levels of metal accumulation in birds as well.

Table 5.8 Levels of metals (mean \pm SE, $\mu\text{g/g}$) in various species of birds collected from four states between 2012- 2014.

States	Zinc	Copper	Nickel	Chromium	Lead	Cadmium	Total Metal
Gujarat	150.65 \pm 2.31	26.22 \pm 0.88	2.21 \pm 0.20	1.31 \pm 0.08	0.65 \pm 0.03	0.89 \pm 0.06	181.94 \pm 2.87
Kerala	150.37 \pm 2.42	27.02 \pm 1.49	1.19 \pm 0.16	1.02 \pm 0.13	0.64 \pm 0.06	0.83 \pm 0.12	181.07 \pm 3.22
Tamil Nadu	144.03 \pm 3.52	27.13 \pm 1.00	2.78 \pm 0.42	1.18 \pm 0.08	0.77 \pm 0.04	0.85 \pm 0.06	176.75 \pm 3.98
One-way ANOVA	<i>F</i> value	1.87	3.04	1.79	7.50	0.51	8.78
	<i>P</i> value	<0.05	>0.05	>0.05	<0.05	>0.05	<0.05

5.2. Feathers: A better tool to understand metal contamination in birds

Totally 130 individuals comprising 19 species of birds were collected from three states in India, namely Gujarat, Tamil Nadu and Kerala. Different types of feathers such as breast feathers, primary feathers, secondary feathers and tail feathers were collected and analyzed for select metals, namely cadmium, lead, chromium, nickel, copper and zinc. The mean and standard error were calculated.

The data have been compiled to look at the variation in the concentrations of individual metal and total metal load in feathers among all the species. Further, an attempt has been made to find out which feather type reflects the metal accumulation the best. Towards this, data on all species of birds have been pooled under each type of feathers.

5.2.1. Variation in the accumulation pattern of metals in the feathers of various species of birds

The variation in total metal load among all the species was significant (Oneway ANOVA, $df = 524$; $F=76.313$; $p<0.05$). To understand the total load of metals on each species of birds, levels of all the metals included in the study were pooled irrespective of feather type. The maximum load was in Indian Black Ibis ($280.05 \pm 62.01 \mu\text{g/g}$) followed by Spot-billed Pelican ($239.59 \pm 52.92 \mu\text{g/g}$) and minimum in Asian Koel ($50.85 \pm 1.68 \mu\text{g/g}$) (Figure 5.7). Discussion on this aspect is made metal wise as detailed below.

5.2.1.a Variation in the accumulation pattern of cadmium in the feathers of various species of birds

When cadmium was considered alone, the maximum concentration was found in the breast feathers of Spot-billed Pelican ($7.09 \pm 2.01 \mu\text{g/g}$) followed by of Indian White-rumped Vulture ($7.08 \pm 0.58 \mu\text{g/g}$), Sarus Crane ($6.08 \pm 0.25 \mu\text{g/g}$) and Shikra ($5.05 \pm 0.19 \mu\text{g/g}$) (Table 5.9). One - way ANOVA showed that there was significant variation in cadmium levels among different species of birds (One Way ANOVA - $F=50.962$; $df=524$; $p<0.05$). The least concentration of cadmium was recorded in tail feathers of Asian Koel ($0.0123 \pm 0.0008 \mu\text{g/g}$).

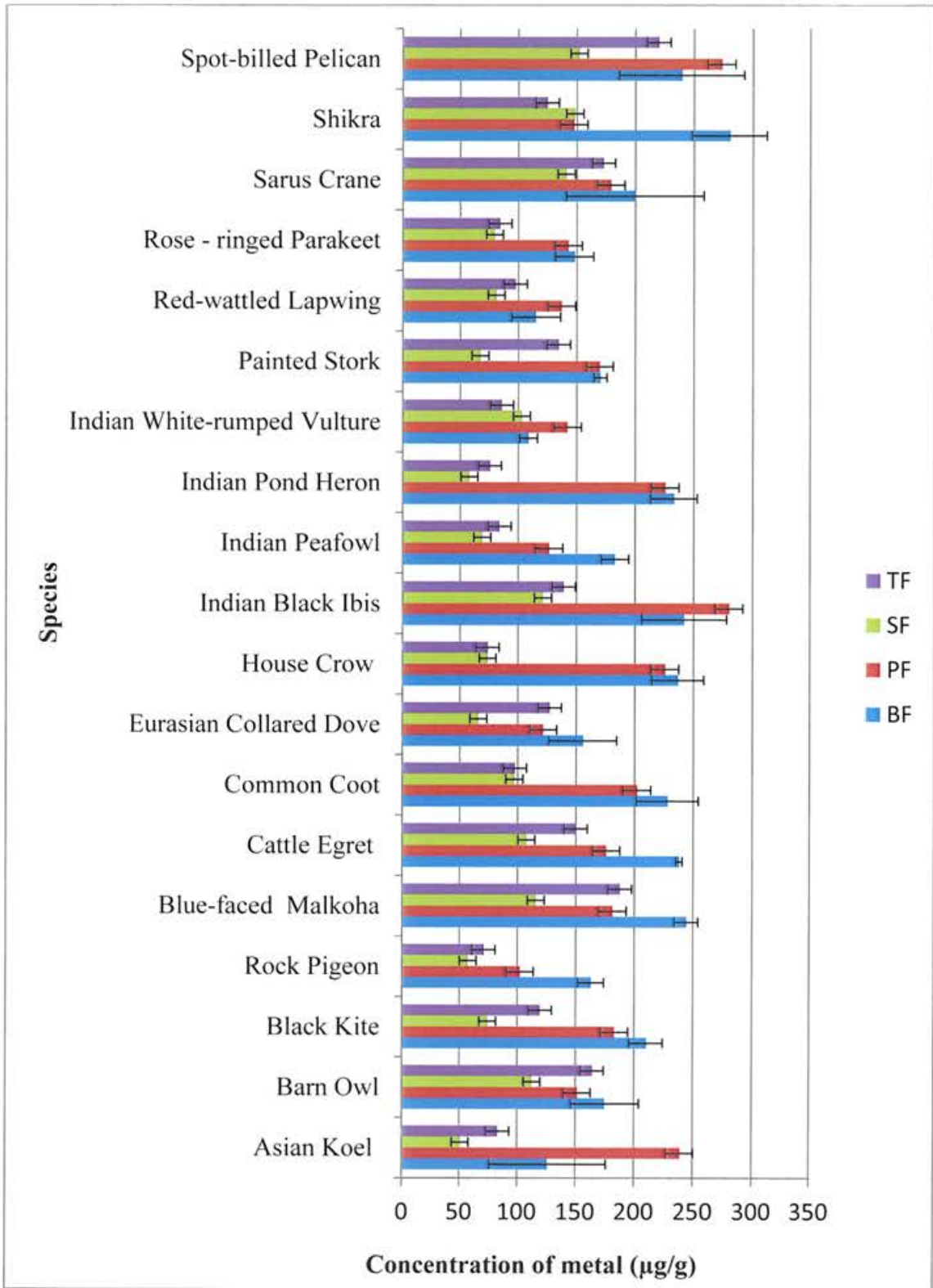


Figure 5.7 Levels of Total Metal Load in four types of feathers in different species collected from various states in India during 2012-2014

The recorded levels of cadmium in feathers in this study are comparable with levels reported from India and elsewhere in different species of birds (Abdullah et al. 2015; Manjula et al. 2015). Levels, when ranged between 0.6 and 25.4 $\mu\text{g/g}$ in feathers, the effects such as in feathers reduced growth rate and poor fledgling success were observed (Burger and Gochfeld 1993b). Levels of cadmium in pectoral feathers of Laggar Falcon *Biarmicus jugger* (0.10 $\mu\text{g/g}$) in Italy (Movalli 2000) and mantle feathers of the Little Owl *Athene noctua* (0.050 $\mu\text{g/g}$) and Common Buzzard *Buteo buteo* (0.060 $\mu\text{g/g}$) in Italy were reported (Battaglia et al. 2005). In Germany, concentrations of Cd in primary feathers of Goshawk *Accipiter gentilis* were 0.3 ± 0.7 $\mu\text{g/g}$ (Hahn et al. 1989) and 0.05 ± 4.20 $\mu\text{g/g}$ (Dietrich and Ellenberg 1986). Feather levels of cadmium known to have caused adverse effects in the birds had ranged from 0.1 $\mu\text{g/g}$ to 2 $\mu\text{g/g}$ (Burger and Gochfeld 1993). In addition to being toxic above certain levels in feathers of fledglings, Cd may also induce deficiencies of essential elements through competition at active sites in biologically important molecules (Walker et al. 1996). Hence, within the levels recorded in the present study (7.19 $\mu\text{g/g}$) only the maximum concentration, in a single individual of breast feather of Indian White-rumped Vulture is to be viewed with concern as it may cause serious consequences in long-run.

Yamac et al. (2019) reported 0.25 ± 1.59 $\mu\text{g/g}$ of Cd in the calamus, and a higher level in the vane 0.33 ± 0.16 $\mu\text{g/g}$ of the feathers of Cinereous Vulture. Besides these data, Cd concentrations up to 1.6 $\mu\text{g/g}$ in feathers of birds of prey, namely Northern Goshawk were reported in an area in New York considered to be unpolluted by Lodenius and Solonen (2013). Adverse effects such as reduced growth rates were reported have been found at lower levels ranging from 0.1 to 2 $\mu\text{g/g}$ (Burger 1993). Hence, it is possible that the sampled birds in this study are experiencing mild adverse effects because of the Cd burden in their feathers.

Table 5.9. Levels of cadmium (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast feathers	Primary Feathers	Secondary feathers	Tail Feathers
1.	Asian Koel	04	4.62 \pm 0.542	0.85 \pm 0.71	0.21 \pm 0.05	0.012 \pm 0.001
2.	Blue - faced Malkoha	05	3.234 \pm 0.49	1.570 \pm 0.514	1.936 \pm 1.041	0.453 \pm 0.392
3.	Black Kite	13	3.089 \pm 0.144	0.428 \pm 0.136	0.229 \pm 0.088	0.366 \pm 0.104
4.	Barn Owl	03	3.610 \pm 0.316	0.310 \pm 0.010	0.670 \pm 0.501	0.247 \pm 0.133
5.	Rock Pigeon	20	3.118 \pm 0.511	0.770 \pm 0.205	1.386 \pm 0.432	0.128 \pm 0.025
6.	Common Coot	08	3.253 \pm 0.124	0.328 \pm 0.080	0.208 \pm 0.050	0.456 \pm 0.129
7.	Cattle Egret	13	2.364 \pm 0.074	1.348 \pm 0.312	1.398 \pm 0.427	3.468 \pm 0.627
8.	Eurasian Collared Dove	16	2.608 \pm 0.298	0.764 \pm 0.248	0.924 \pm 0.331	0.512 \pm 0.150
9.	House Crow	10	3.659 \pm 0.024	0.565 \pm 0.323	0.226 \pm 0.072	0.203 \pm 0.069
10.	Indian Black Ibis	06	3.059 \pm 0.303	1.571 \pm 0.538	2.442 \pm 1.049	2.391 \pm 1.420
11.	Indian Peafowl	05	3.943 \pm 0.216	0.370 \pm 0.090	0.270 \pm 0.0559	0.288 \pm 0.012
12.	Indian Pond Heron	06	2.917 \pm 0.055	0.137 \pm 0.047	0.242 \pm 0.087	0.218 \pm 0.059
13.	Indian White- rumped Vulture	10	7.080 \pm 0.582	2.260 \pm 0.468	1.518 \pm 0.737	3.917 \pm 3.663
14.	Painted Stork	04	3.201 \pm 0.120	1.367 \pm 0.704	2.286 \pm 0.452	2.049 \pm 1.349
15.	Rose-ringed Parakeet	06	2.499 \pm 0.097	1.785 \pm 0.338	2.037 \pm 1.078	1.154 \pm 0.104
16.	Red -wattled Lapwing	04	5.529 \pm 0.147	2.289 \pm 0.414	3.517 \pm 0.801	5.125 \pm 1.787
17.	Spot-billed Pelican	03	7.099 \pm 2.013	3.033 \pm 0.445	3.306 \pm 0.236	7.847 \pm 4.145
18.	Sarus Crane	03	3.369 \pm 0.631	3.173 \pm 0.040	3.846 \pm 0.921	6.085 \pm 0.251
19.	Shikra	03	5.054 \pm 0.192	2.375 \pm 0.398	1.298 \pm 0.146	1.618 \pm 0.274

Interestingly, near equal concentrations of Cd were observed in the primary feathers of Red-wattled Lapwing ($1.46 \pm 0.31 \mu\text{g/g}$) and Indian White - rumped Vulture ($1.63 \pm 0.35 \mu\text{g/g}$) which have different feeding habits and foraging sites. Red-wattled Lapwing is insectivorous while White-rumped Vulture is carnivorous. The near equal levels of cadmium lead us to assume that rural agricultural lands are becoming increasingly contaminated. Red-wattled Lapwing is found not only in rural pockets or forests and agricultural lands, they also extend their foraging range to the urban environment. This may be a plausible factor for measuring high levels of Cd. Further, insects too do contain appreciable levels of Cd which may be an added dose to the birds (Azam et al. 2015).

The level of cadmium recorded in the breast feathers of Franklin Gull, collected from Lake Alice US was $5.37 \mu\text{g/g}$ (Burger 1996). She justified that the Franklin Gull feeds on earthworm which lives in contaminated soil. Further, Frantz et al. (2012) assessed levels of four metals (cadmium, lead, copper and zinc) in the feathers of 91 Feral Pigeons from seven sites in the urbanized region of Paris. Metals were detected in all the pigeons, indicating that metals persist in urbanized areas. The ratio between metal concentrations in the feathers versus urbanization rate calculated using data from other studies was 2 - 90 times higher for cadmium than for other metals, underlining its ecological importance. Concentrations in the feathers depended on locality, suggesting that pigeons remain in local habitats at this restricted scale, as explained in above observations.

5.2.1.b. Variation in the accumulation pattern of lead in the feathers of various species of birds

The maximum concentration of lead was recorded in the primary feathers of the Spot-billed Pelican ($3.26 \pm 0.13 \mu\text{g/g}$) followed by the breast feathers of House Crow ($2.99 \pm 0.63 \mu\text{g/g}$). Further Rock Pigeon and Eurasian Collared Dove also recorded more than $2 \mu\text{g/g}$ of lead $2.54 \pm 0.07 \mu\text{g/g}$ and $2.30 \pm 0.04 \mu\text{g/g}$ respectively (Table 5.10). The variations in the observed levels were significant among various species of birds studied. (One - Way ANOVA $F=147.25$; $df=524$; $p<0.05$).

Table 5.10. Levels of lead contaminations (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast Feather	Primary Feather	Secondary Feather	Tail feather
1.	Asian Koel	04	2.840 \pm 0.025	1.485 \pm 0.428	0.193 \pm 0.113	0.210 \pm 0.042
2.	Blue Faced Malkoha	05	2.844 \pm 0.028	1.152 \pm 0.405	0.888 \pm 0.168	0.460 \pm 0.347
3.	Black Kite	13	2.462 \pm 0.080	1.567 \pm 0.251	0.247 \pm 0.081	0.136 \pm 0.018
4.	Barn Owl	03	2.845 \pm 0.256	2.550 \pm 0.450	0.804 \pm 0.668	0.117 \pm 0.003
5.	Rock Pigeon	20	2.548 \pm 0.079	0.800 \pm 0.163	0.467 \pm 0.074	0.624 \pm 0.112
6.	Common Coot	08	2.639 \pm 0.136	0.703 \pm 0.244	0.340 \pm 0.110	0.245 \pm 0.051
7.	Cattle Egret	13	2.335 \pm 0.058	1.481 \pm 0.260	0.778 \pm 0.186	1.778 \pm 0.619
8.	Eurasian Collared Dove	16	2.307 \pm 0.041	1.517 \pm 0.260	0.354 \pm 0.095	0.173 \pm 0.045
9.	House Crow	10	2.999 \pm 0.632	0.920 \pm 0.236	1.473 \pm 0.252	0.522 \pm 0.160
10.	Indian Black Ibis	06	2.638 \pm 0.075	2.270 \pm 0.434	1.674 \pm 0.286	1.097 \pm 0.538
11.	Indian Peafowl	05	2.983 \pm 0.274	1.999 \pm 0.326	0.481 \pm 0.154	1.824 \pm 0.939
12.	Indian Pond Heron	06	2.939 \pm 0.093	1.662 \pm 0.308	1.422 \pm 0.329	0.911 \pm 0.301
13.	Indian White - rumped Vulture	10	2.481 \pm 0.233	1.634 \pm 0.352	0.805 \pm 0.364	0.615 \pm 0.163
14.	Painted Stork	04	2.869 \pm 0.219	1.804 \pm 0.756	0.572 \pm 0.418	2.852 \pm 0.917
15.	Rose-ringed Parakeet	06	2.409 \pm 0.090	0.956 \pm 0.262	0.828 \pm 0.253	0.658 \pm 0.057
16.	Red -wattled Lapwing	04	2.228 \pm 0.017	1.460 \pm 0.314	0.154 \pm 0.042	0.141 \pm 0.039
17.	Spot-billed Pelican	03	3.188 \pm 0.484	3.269 \pm 0.135	1.273 \pm 0.574	2.825 \pm 1.511
18.	Sarus Crane	03	2.962 \pm 0.538	0.446 \pm 0.039	0.412 \pm 0.163	0.537 \pm 0.103
19.	Shikra	03	2.570 \pm 0.182	1.444 \pm 1.079	0.499 \pm 0.167	0.517 \pm 0.204

The level of lead recorded in the breast feather of the Franklin Gull collected from Lake Alice USA was 2.86 $\mu\text{g/g}$ (Burger 1996). The levels recorded in the breast feather of Spot-billed Pelican in the present study ($3.26 \pm 0.13 \mu\text{g/g}$) is higher than the levels reported in adult Black-browed Albatross (3.35 $\mu\text{g/g}$) in Argentina which also mostly depends on fishes. In our study, Spot-billed Pelican showed appreciable levels of Pb ($3.58 \pm 0.12 \mu\text{g/g}$) and it was found to be comparable with the levels reported by (Pon et al. 2011).

The lead levels in the breast (21.52 $\mu\text{g/g}$) and moulted feathers (3.15 $\mu\text{g/g}$) of House Crow were reported by Scheifler et al. (2006) in Besançon, Eastern France. Similarly, Eens et al. (1999) reported the levels of lead in tail feathers of Great Tit (16.3 $\mu\text{g/g}$) and Blue Tit (64.0 $\mu\text{g/g}$) in Belgium. The levels of lead recorded in the House Crow in the present study were lower than the levels reported above. Burger et al. (1997) documented the levels of lead in feathers (1.55– 2.01 $\mu\text{g/g}$) of Mourning Dove from two heavily contaminated agricultural landscapes in USA. The levels of lead recorded in Rock Pigeon and Eurasian Collared Dove in the present study were higher than the levels reported in the above mentioned studies. The level of lead in the present study was far less than the level recorded in Urban Pigeon (13.82 $\mu\text{g/g}$) collected from Paris (Frantz et al. 2012). Pigeons exhibit a high level of site fidelity to birth sites, generally remaining in a small area (<2 km) for their entire lives (Rose et al. 2006). This behavioral characteristic thus permits comparisons of Pb toxicity data on pigeon among micro urban environments (Frantz et al. 2012).

Lead levels in feathers of Franklin's Gulls averaged 0.96 $\mu\text{g/g}$ (young) and 2.86 $\mu\text{g/g}$ (adults) compared to 0.87 to 4.61 $\mu\text{g/g}$ for Herring Gulls from the Great Lakes (Struger et al. 1987), and 1.81 to 2.10 $\mu\text{g/g}$ for Herring Gull fledglings from Long Island (Burger and Gochfeld 1993b). Further Janssens et al. (2001) documented the levels of lead in tail feathers (230.5 $\mu\text{g/g}$) of Great Tit from urban area of Belgium. Spahn and Sherry (1999) reported the levels of lead in feathers of dead nestlings of Little Blue Heron (1.2 to 16.9 $\mu\text{g/g}$) from South Louisiana wetlands. The levels of lead recorded in Indian Pond Heron in the present study were lower than the levels reported in above-referred studies. Gochfeld et al. (1996) reported lead in pectoral feathers in the

Laughing gull to be 0.003 $\mu\text{g/g}$. Adverse effects of lead in birds begin at levels in feathers at 4 $\mu\text{g/g}$ (Eisler 1985a; Custer and Hoffman 1994; Burger and Gochfeld 2000c). Normal background levels for adult sea birds were in the range of 0.51 – 1.68 $\mu\text{g/g}$ (Mendes et al. 2008; Burger et al. 2009).

The levels of lead in pectoral feathers of the Lager Falcon (1.56 $\mu\text{g/g}$), mantle feathers of Little Owl (2.00 $\mu\text{g/g}$) and Common Buzzard (1.48 $\mu\text{g/g}$) in Italy were reported by Movalli (2000). The levels of lead in the primary feathers (1.56 \pm 0.25 $\mu\text{g/g}$) of Black Kite in the present study had similar concentrations. Common Buzzard with 47.70 $\mu\text{g/g}$ of lead in its feathers was reported to be due to acute exposure (Battaglia et al. 2005). The levels of lead reported in the present study are below any acute exposure.

The concentrations of lead in all the species included in the present study ranged from 0.2 to 3.2 $\mu\text{g/g}$. Insectivorous birds, namely Cattle Egret (1.48 \pm 0.26 $\mu\text{g/g}$) and Red wattled Lapwing (1.46 \pm 0.31 $\mu\text{g/g}$) accumulated similar concentrations in primary feathers. Likewise, Indian Pond Heron (1.66 \pm 0.30 $\mu\text{g/g}$) and Indian White-rumped Vulture (1.63 \pm 0.35 $\mu\text{g/g}$) which are carnivorous accumulated similar levels. Indian Black Ibis (2.27 \pm 0.43 $\mu\text{g/g}$) and House Crow (2.25 \pm 0.14 $\mu\text{g/g}$) had more or less equal concentrations even though they use different habitats. However, the both species of birds are omnivorous in their feeding habits. Omnivorous birds had the highest and insectivorous had the least concentrations of Pb accumulation while comparing feeding habits. The levels were less than the levels reported in previous studies by Muralidharan et al. (2004) in Common Myna (omnivorous: 2.20 $\mu\text{g/g}$) and Jungle Babbler (insectivorous: 1.63 $\mu\text{g/g}$) from the Nilgiris.

Interestingly, similar concentrations of Pb was observed in primary feathers of Rose - ringed Parakeet (2.40 \pm 0.09 $\mu\text{g/g}$) and Shikra (2.57 \pm 0.18 $\mu\text{g/g}$) which have entirely a different feeding habits and foraging sites. While the Rose - ringed Parakeet is a frugivore Shikra is a carnivorous and restricted by and large to forested ecosystems. Similar levels of Pb in both the species give us a picture that rural agricultural lands are becoming increasingly contaminated. Rose - ringed Parakeet is found not only in rural pockets or forests or agricultural lands, they also extend their foraging range in to the

urban environment as well. This may be a plausible reason for measuring high levels of Pb. Further, fruits and seeds do contain appreciable levels of Pb which is a major source.

The values reported in the present study in primary feathers for Black Kite ($1.56 \pm 0.25 \mu\text{g/g}$) were low when compared to the levels reported in Black Kite ($0.39 \mu\text{g/g}$) in Gujarat by Sandhya et al. (2012). Levels of Pb ($2.25 \pm 0.14 \mu\text{g/g}$) recorded in the House Crow which is a common urban bird, was found to be lower than the levels reported in birds ($37.02 \mu\text{g/g}$) collected from urban locations of India, Ahmedabad, Gujarat (Sandhya et al. 2012). Further levels observed in the present study were lower than the levels reported in breast feather of House Crows ($32.01 \mu\text{g/g}$) in Malaysia (Janaydeh et al. 2016) and in the breast feathers ($21.52 \mu\text{g/g}$) of House Crows (Scheifler et al. 2006) in France.

Lead concentrations recorded in the breast feathers of Rock Pigeon ($2.547 \pm 0.079 \mu\text{g/g}$) and Eurasian Collared Dove ($2.30 \pm 0.04 \mu\text{g/g}$) which are urban or semi-urban, are found to be comparable with the levels recorded in the Rock Pigeon ($2.04 - 7.95 \mu\text{g/g}$) collected in Brazil (Brait and Filho 2011). The levels recorded in present study fall within the safe range mentioned above.

Scheifler et al. (2006) indicated that Pb excretes high because availability of insects or prey items of birds may become available to birds in local environment. For Pb, adverse effects in birds occur at levels of $4 \mu\text{g/g}$ in feathers (Burger and Gochfeld 2000c). The levels of lead in the present study were lower than the recommended ($4 \mu\text{g/g}$) concentrations referred above in all types of feathers.

5.2.1.c. Variation in the accumulation pattern of chromium in the feathers of various species of birds

Among the species of birds studied, the highest concentration of chromium was recorded in primary feathers of Spot-billed Pelican (9.19 ± 3.09), followed by Red-wattled Lapwing ($6.48 \pm 0.73 \mu\text{g/g}$), Asian Koel ($4.03 \pm 2.97 \mu\text{g/g}$) Shikra ($6.47 \pm 0.58 \mu\text{g/g}$) and secondary feathers of Spot-billed Pelican ($8.33 \pm 2.79 \mu\text{g/g}$) (Table 5.11). Statistical test showed that there was a significant variation in chromium accumulation in feathers among the species of birds studied (One - way ANOVA $F=5.284$; $df= 524$; $p<0.05$).

The levels of chromium concentrations in all four types of feathers were lower than the levels reported in breast feathers of adult Great Egret, *Egretta alba* (31.2 µg/g) in Hong Kong and China reported by Burger and Gochfeld (1993). Further, the levels of chromium concentrations in the present study were higher than the levels in Laggar Falcons (2.35 µg/g) reported in Italy (Movalli 2000).

The average level of chromium recorded in the breast feathers of Franklin Gull collected from Lake Alice USA was 1.55 µg/g (Burger 1996). The levels of chromium in primary feathers of Painted Stork (7.79 µg/g) from the present study are similar to the levels recorded in one individual of adult Australian Gannet (6.25 µg/g) (Burger et al. 1994) from New Zealand. Even though Cr is an essential dietary element to most of the species, Cr concentration of 2.8 µg/g in feathers of birds has been found to induce toxic effects (Burger and Gochfeld 2000a; 2000b). Although Chromium can come from natural sources, it is increasing drastically in the environment due to various anthropogenic activities (Parekh et al. 1989).

With regard to the levels in raptors, the breast feathers of Laggar Falcons (0.12 µg/g) was investigated for metals in various places in Italy by Movalli (2000). In the present study, breast feathers of Barn Owl had (0.96 ± 0.59) levels comparable to the study conducted in the same species in Europe (0.76 µg/g) (Gruz et al. 2018). Cr concentration of 2.8 µg/g in feathers has been related to toxic effects (Burger and Gochfeld, 2000). The levels of chromium were higher than the above referred no-effects levels in six species, namely, the Indian White-rumped Vulture (CR), Painted Storks and Spot-billed Pelican (NT), Sarus Crane (VU), Red-wattled Lapwing and Shikra (LC) should be monitored.

Table 5.11. Levels of chromium contamination (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast Feathers	Primary feathers	Secondary feathers	Tail fathers
1.	Asian Koel	04	0.290 \pm 0.025	7.823 \pm 2.979	2.322 \pm 0.636	0.420 \pm 0.084
2.	Blue-faced Malkoha	05	1.236 \pm 0.511	1.534 \pm 0.438	0.928 \pm 0.071	2.477 \pm 1.160
3.	Black Kite	13	4.612 \pm 0.149	1.730 \pm 1.115	1.224 \pm 0.502	2.234 \pm 1.134
4.	Barn Owl	03	0.968 \pm 0.598	0.073 \pm 0.023	1.676 \pm 1.504	0.514 \pm 0.286
5.	Rock Pigeon	20	1.898 \pm 1.584	2.940 \pm 0.485	1.821 \pm 0.488	0.902 \pm 0.316
6.	Common Coot	08	1.064 \pm 0.286	0.932 \pm 0.260	0.631 \pm 0.229	0.473 \pm 0.112
7.	Cattle Egret	13	1.052 \pm 0.150	4.633 \pm 1.207	1.948 \pm 0.372	5.865 \pm 0.794
8.	Eurasian Collared Dove	16	1.544 \pm 1.632	3.398 \pm 0.717	1.310 \pm 0.491	1.593 \pm 0.646
9.	House Crow	10	1.048 \pm 0.848	1.473 \pm 0.743	1.758 \pm 0.926	0.373 \pm 0.097
10.	Indian Black Ibis	06	2.951 \pm 1.261	5.321 \pm 0.560	2.912 \pm 0.710	5.734 \pm 1.057
11.	Indian Peafowl	05	1.888 \pm 0.433	0.883 \pm 0.381	1.090 \pm 0.420	1.368 \pm 0.680
12.	Indian Pond Heron	06	3.136 \pm 0.313	0.780 \pm 0.460	0.527 \pm 0.149	0.916 \pm 0.432
13.	Indian White-rumped Vulture	10	4.494 \pm 0.957	4.256 \pm 0.625	4.098 \pm 0.993	2.977 \pm 2.739
14.	Painted Stork	04	4.794 \pm 1.290	6.187 \pm 1.305	4.038 \pm 1.345	4.300 \pm 1.864
15.	Rose-ringed Parakeet	06	1.005 \pm 0.189	0.728 \pm 0.062	0.425 \pm 0.085	2.020 \pm 0.182
16.	Red -wattled Lapwing	04	5.358 \pm 0.194	6.489 \pm 0.734	5.307 \pm 0.668	5.844 \pm 0.442
17.	Spot-billed Pelican	03	5.762 \pm 0.669	9.198 \pm 3.090	3.032 \pm 0.440	8.330 \pm 2.791
18.	Sarus Crane	03	2.230 \pm 2.070	4.762 \pm 0.600	5.477 \pm 0.917	3.272 \pm 0.166
19.	Shikra	03	4.785 \pm 2.715	6.471 \pm 0.584	5.000 \pm 0.309	4.654 \pm 1.089

5.2.1.d. Variation in the accumulation pattern of nickel in the feathers of various species of birds

Interestingly, Blue - faced Malkoha had the highest level of nickel in its tail feathers ($17.62 \pm 15.04 \mu\text{g/g}$). The next highest concentration of nickel was recorded in the primary feathers of the Painted Stork ($14.23 \pm 4.77 \mu\text{g/g}$) followed by Indian Black Ibis ($11.04 \pm 5.43 \mu\text{g/g}$). The least concentration of nickel was recorded in the secondary feathers of Asian Koel ($0.10 \pm 0.01 \mu\text{g/g}$). The levels did not vary significantly among the species included in the present study (One - way ANOVA $F=0.919$; $df=524$; $p>0.05$) (Table 5.12).

Nickel concentrations in this study were higher than those reported in the feathers of Eurasian Coot *Fulicaatra*, Great Cormorant and Black-crowned Night Heron from Russia (Lebedeva 1997) and in the primary feathers of American Black Duck ($5.3 \mu\text{g/g}$) from Ontario (Ranta et al. 1978), but were similar to those in the primary feathers of Kelp Gull ($5.92 \mu\text{g/g}$) from Brazil (Barbieri et al. 2010). The levels of nickel in primary feathers of Cattle Egret in the present study is higher than the levels reported by Honda et al. (1985) in the primary feathers of Eastern Great White Egret ($0.14 \mu\text{g/g}$) from Korea.

In the present study the levels of nickel observed in breast feather of Cattle Egret ($0.90 \pm 0.18 \mu\text{g/g}$) was comparable with the insectivorous species Great Tits ($2.5 \mu\text{g/g}$) collected from resident area in Beijing, China (Deng et al. 2007). Further nickel from current observation was lower than the levels recorded in Little Egret ($13.31 \mu\text{g/g}$) collected from Tiruchirapalli, Tamil Nadu, India (Manjula et al. 2015).

Table 5.12. Levels of nickel (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast feathers	Primary Feathers	Secondary feathers	Tail Feathers
1.	Asian Koel	04	0.290 \pm 0.089	0.455 \pm 0.122	0.102 \pm 0.016	0.448 \pm 0.104
2.	Blue-faced Malkoha	05	15.291 \pm 10.953	10.652 \pm 8.335	1.88 \pm 1.187	17.626 \pm 15.040
3.	Black Kite	13	0.782 \pm 0.133	0.896 \pm 0.301	3.373 \pm 1.555	4.023 \pm 1.458
4.	Barn Owl	03	0.394 \pm 0.159	0.198 \pm 0.102	1.328 \pm 1.157	7.436 \pm 0.742
5.	Rock Pigeon	20	3.093 \pm 0.718	3.043 \pm 0.545	1.290 \pm 0.268	1.416 \pm 0.362
6.	Common Coot	08	1.064 \pm 0.286	0.632 \pm 0.196	0.656 \pm 0.225	44.958 \pm 1.290
7.	Cattle Egret	13	0.901 \pm 0.185	4.024 \pm 1.078	3.355 \pm 1.383	8.592 \pm 2.224
8.	Eurasian Collared Dove	16	27.401 \pm 7.593	9.477 \pm 6.658	0.850 \pm 0.327	2.186 \pm 1.944
9.	House Crow	10	38.709 \pm 36.428	11.009 \pm 9.572	8.063 \pm 5.097	0.945 \pm 0.386
10.	Indian Black Ibis	06	7.467 \pm 4.028	11.049 \pm 5.433	7.669 \pm 3.534	8.854 \pm 7.721
11.	Indian Peafowl	05	1.966 \pm 0.548	1.697 \pm 1.083	1.161 \pm 0.309	2.318 \pm 0.830
12.	Indian Pond Heron	06	2.192 \pm 0.891	2.525 \pm 0.828	3.292 \pm 0.880	1.792 \pm 0.623
13.	Indian White-rumped Vulture	10	10.101 \pm 5.007	8.949 \pm 3.750	23.693 \pm 14.301	9.185 \pm 8.279
14.	Painted Stork	04	3.683 \pm 0.803	14.237 \pm 4.770	2.842 \pm 1.5112	2.139 \pm 1.247
15.	Rose-ringed Parakeet	06	5.751 \pm 1.597	8.111 \pm 3.002	5.456 \pm 1.866	4.011 \pm 0.839
16.	Red -wattled Lapwing	04	7.303 \pm 2.209	5.168 \pm 1.233	6.265 \pm 1.281	4.299 \pm 0.603
17.	Spot-billed Pelican	03	5.957 \pm 1.386	5.742 \pm 0.090	11.416 \pm 5.312	10.244 \pm 2.988
18.	Sarus Crane	03	5.282 \pm 1.560	4.160 \pm 1.489	1.032 \pm 0.509	2.558 \pm 1.100
19.	Shikra	03	7.156 \pm 5.960	6.708 \pm 1.658	5.105 \pm 3.138	6.247 \pm 2.697



5.2.1.e Variation in the accumulation pattern of copper in the feathers of various species of birds

While the highest concentration of copper was recorded in the primary feathers of Indian Black Ibis ($176.61 \pm 60.77 \mu\text{g/g}$) followed by breast feathers of Spot-billed Pelican ($106.27 \pm 28.27 \mu\text{g/g}$), the least levels of copper were recorded in secondary feathers of Asian Koel ($22.22 \mu\text{g/g}$) and Indian Pond Heron ($22.96 \mu\text{g/g}$) (Table 5.13). Significant variations were observed in the levels of copper detected among the species (One - way ANOVA $F=17.699$; $df=542$; $p<0.05$).

The values reported in the present study for Shikra ($45.82 \pm 11.17 \mu\text{g/g}$) were low when compared to the levels reported in Sparrow Hawks ($3.16 \mu\text{g/g}$) and Little Owls ($8.98 \mu\text{g/g}$) studied at Bird Sanctuaries in Flanders (Dauwe et al. 2003). Levels of copper in Black-browed Albatross ($48.6 \mu\text{g/g}$), a sea bird which feeds mainly on the fishes from Argentina are comparable with the levels in breast feathers of Painted Stork (48.79 ± 4.50) included in the present study (Pon et al. 2011). Levels recorded in the present study were much lower than the concentrations reported above. But the levels of copper in the present study were similar to the levels documented from Tamil Nadu, India by Manjula et al. (2015) who studied feathers of House Crow ($139 \mu\text{g/g}$), House Sparrow ($148.3 \mu\text{g/g}$) and Indian Peafowl ($155.6 \mu\text{g/g}$). The levels of copper in the House Crow ($66.81 \pm 7.70 \mu\text{g/g}$) and Indian Peafowl ($58.65 \pm 19.81 \mu\text{g/g}$) were lower than the values reported by Manjula et al. (2015).

In the present study, the levels of copper measured in Cattle Egret ($63.84 \pm 3.27 \mu\text{g/g}$) was comparably lower ($82.15 \mu\text{g/g}$) than the levels recorded in Cattle Egret collected from Nilgiris District, Tamil Nadu, India (Muralidharan et al. 2004). Further, the levels recorded in Indian Pond-heron (72.102 ± 3.872) is almost equal to the levels reported ($71.58 \mu\text{g/g}$) in primary feathers of the species collected from Nilgiris district (Vishnu 2001). The levels of copper in secondary feathers of Cattle Egret ($39.0 \mu\text{g/g}$) were 10 times higher than the study reported by Malik and Zeb (2009) in feathers of Cattle Egret Chick ($3.9 \mu\text{g/g}$) from Pakistan. The levels of copper in tail feathers of Cattle Egret (57.15 ± 8.67) are comparable to the insectivorous bird Great Tit ($54.89 \mu\text{g/g}$) from Belgium (Janssens et al. 2001). Copper concentrations in this study were lower than those levels reported from Georgia and the Indian Ocean on several species of birds (Kim et al. 1998; Anderson et al. 2010).

Table 5.13. Levels of copper (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast feathers	Primary feathers	Secondary feathers	Tail feathers
1.	Asian Koel	04	84.168 \pm 1.681	293.725 \pm 227.59	22.227 \pm 0.4564	48.486 \pm 3.3801
2.	Blue-faced Malkoha	05	64.653 \pm 5.265	48.815 \pm 8.100	32.844 \pm 6.3821	52.382 \pm 9.293
3.	Black Kite	13	83.617 \pm 1.139	68.261 \pm 2.057	24.762 \pm 1.417	43.988 \pm 1.469
4.	Barn Owl	03	45.226 \pm 20.700	48.617 \pm 16.277	79.744 \pm 51.378	59.348 \pm 14.952
5.	Rock Pigeon	20	63.306 \pm 11.731	40.034 \pm 3.595	25.276 \pm 1.045	39.147 \pm 3.458
6.	Common Coot	08	68.093 \pm 10.637	63.707 \pm 3.394	23.707 \pm 1.008	44.958 \pm 1.290
7.	Cattle Egret	13	80.316 \pm 1.255	63.840 \pm 3.276	39.112 \pm 10.006	57.159 \pm 8.672
8.	Eurasian Collared Dove	16	56.252 \pm 18.512	54.390 \pm 7.014	32.553 \pm 6.197	44.275 \pm 2.404
9.	House Crow	10	66.818 \pm 7.705	80.498 \pm 10.754	32.478 \pm 4.100	45.184 \pm 3.548
10.	Indian Black Ibis	06	114.511 \pm 32.512	176.615 \pm 60.773	63.824 \pm 18.618	97.066 \pm 17.609
11.	Indian Peafowl	05	58.656 \pm 19.818	67.775 \pm 1.083	24.205 \pm 1.161	50.442 \pm 3.624
12.	Indian Pond Heron	06	82.199 \pm 4.137	72.102 \pm 3.872	22.966 \pm 0.594	45.932 \pm 1.864
13.	Indian White-rumped Vulture	10	41.617 \pm 16.734	72.723 \pm 9.964	35.091 \pm 4.856	34.339 \pm 7.719
14.	Painted Stork	04	48.794 \pm 4.509	45.170 \pm 15.337	35.269 \pm 1.755	67.201 \pm 8.047
15.	Rose-ringed Parakeet	06	50.977 \pm 10.146	44.716 \pm 7.248	40.728 \pm 8.451	35.158 \pm 2.357
16.	Red -wattled Lapwing	04	49.442 \pm 25.026	88.676 \pm 3.146	30.375 \pm 2.419	54.154 \pm 3.896
17.	Spot-billed Pelican	03	81.419 \pm 40.154	106.279 \pm 28.275	95.237 \pm 37.196	108.095 \pm 46.995
18.	Sarus Crane	03	68.670 \pm 26.370	44.623 \pm 3.942	47.914 \pm 11.724	53.653 \pm 10.347
19.	Shikra	03	86.942 \pm 11.808	45.820 \pm 11.179	54.899 \pm 13.041	59.281 \pm 17.991

5.2.1.f. Variation in the accumulation pattern of zinc in the feathers of various species of birds

Highest concentration of zinc was found in the breast feathers of Shikra ($173.88 \pm 23.61 \mu\text{g/g}$) followed by of Blue - faced Malkoha ($156.96 \pm 0.96 \mu\text{g/g}$) from Anaikatty, primary feathers of Indian Pond Heron ($148.60 \pm 17.20 \mu\text{g/g}$) and Spot-billed Pelican ($145.89 \pm 10.23 \mu\text{g/g}$). Breast feathers of Cattle Egret ($150.96 \pm 1.36 \mu\text{g/g}$) and primary feathers of Indian Pond Heron ($148.60 \pm 17.20 \mu\text{g/g}$) had more than $150 \mu\text{g/g}$ among all species. There were significant variations in levels of zinc detected among the species studied (one - way ANOVA $F=90.551$; $df=524$; $p<0.05$). The least level of zinc was observed in the secondary feathers of Painted Stork ($22.49 \pm 0.77 \mu\text{g/g}$) Rock Pigeon ($27.11 \pm 0.77 \mu\text{g/g}$) (Table 5.14).

The levels of zinc in the present study were higher than the values reported previously from other countries. Doi and Fukuyama (1983) found Zn mostly in the range $50.0 \pm 250.0 \mu\text{g/g}$ in feathers of wild and zoo-kept birds in Hokkaido, Japan; Scanlon et al. (1979) found mean Zn concentrations to be $76.9 \pm 124.8 \mu\text{g/g}$ in feathers of wild turkey, in Virginia, USA; Scanlon et al. (1980) found mean Zn concentrations to be $82.0 \pm 112.0 \mu\text{g/g}$ in feathers of Ruffed Grouse (*Bonasa umbellus*) in Virginia. Further, the zinc concentrations were found to be higher than the levels reported in a previous study in Tamil Nadu, India by Manjula et al. (2015). They reported zinc concentration in breast feathers of House Crow to be $114.00 \mu\text{g/g}$ while in the present study the level of zinc in breast feather of House Crow ($123.43 \pm 13.38 \mu\text{g/g}$) was slightly higher.

In the present study, the levels measured in the primary feathers of Cattle Egret ($101.96 \pm 9.96 \mu\text{g/g}$) were more or less similar to the levels recorded in Green Bee-eater ($116.0 \mu\text{g/g}$) which also mostly depends on insects (Manickam 2002) but lower than the levels in same species ($82.15 \mu\text{g/g}$) collected from Nilgiris District, Tamil Nadu, India (Muralidharan et al. 2004). This may be because of the contamination of the area live and the food they feed on.

Table 5.14. Levels of zinc (mean \pm SE, $\mu\text{g/g}$) in different types of feathers of select species of birds

S. No.	Name of the species	No. of Individuals	Breast feathers	Primary feathers	Secondary feathers	Tail feathers
1.	Asian Koel	04	84.168 \pm 8.456	134.570 \pm 31.410	25.798 \pm 2.127	33.910 \pm 1.531
2.	Blue-faced Malkoha	05	156.966 \pm 0.960	118.081 \pm 22.227	77.566 \pm 11.416	114.613 \pm 34.249
3.	Black Kite	13	119.678 \pm 14.089	110.518 \pm 8.999	44.711 \pm 5.240	69.019 \pm 16.110
4.	Barn Owl	03	122.134 \pm 48.946	99.762 \pm 63.732	28.561 \pm 1.929	29.954 \pm 3.196
5.	Rock Pigeon	20	86.166 \pm 4.465	54.943 \pm 4.0295	27.110 \pm 0.774	29.955 \pm 1.610
6.	Common Coot	08	152.000 \pm 17.381	137.004 \pm 8.377	71.842 \pm 19.882	54.958 \pm 4.274
7.	Cattle Egret	13	150.966 \pm 1.369	101.968 \pm 9.960	61.233 \pm 9.536	73.184 \pm 12.050
8.	Eurasian Collared Dove	16	63.901 \pm 9.0714	52.492 \pm 7.600	30.069 \pm 1.208	78.956 \pm 11.270
9.	House Crow	10	123.436 \pm 13.387	131.392 \pm 9.622	30.068 \pm 2.444	26.968 \pm 3.710
10.	Indian Black Ibis	06	111.255 \pm 16.092	83.231 \pm 10.707	43.088 \pm 9.888	24.132 \pm 0.815
11.	Indian Peafowl	05	113.812 \pm 11.523	67.775 \pm 7.313	42.010 \pm 15.888	28.0833 \pm 3.209
12.	Indian Pond Heron	06	141.732 \pm 17.029	148.605 \pm 17.207	29.598 \pm 3.004	26.2644 \pm 8.14
13.	Indian White-rumped Vulture	10	43.235 \pm 6.849	52.570 \pm 9.390	38.123 \pm 7.809	35.237 \pm 11.119
14.	Painted Stork	04	107.290 \pm 0.264	101.290 \pm 0.285	22.491 \pm 0.770	56.302 \pm 10.855
15.	Rose-ringed Parakeet	06	85.620 \pm 10.941	86.766 \pm 11.545	30.458 \pm 1.601	41.545 \pm 2.510
16.	Red -wattled Lapwing	04	45.552 \pm 3.283	33.351 \pm 2.268	35.751 \pm 6.104	28.308 \pm 7.792
17.	Spot-billed Pelican	03	136.172 \pm 10.167	145.892 \pm 10.235	37.808 \pm 15.449	82.856 \pm 46.412
18.	Sarus Crane	03	117.340 \pm 32.740	122.581 \pm 4.820	82.496 \pm 32.628	107.306 \pm 20.694
19.	Shikra	03	173.883 \pm 23.617	84.974 \pm 30.147	79.797 \pm 26.670	48.561 \pm 15.654

The Zn concentrations recorded in urban and semi-urban birds such as primary feather of Rock Dove ($54.94 \pm 4.02 \mu\text{g/g}$) and Eurasian Collared Dove ($52.49 \pm 7.60 \mu\text{g/g}$) are found to be comparable with the levels recorded in the Rock Pigeon (8.4 to 46 $\mu\text{g/g}$) in Southern Finland. Further Solonen et. al. (1999) recorded 5 - 250 $\mu\text{g/g}$ of Zn in Rock Pigeon. The present values fall within the range mentioned above. The levels could be attributed to zinc's affinity towards keratin. However, external contamination and topographical variation cannot be ruled out.

The common urban bird, House Crow had $131.39 \pm 9.62 \mu\text{g/g}$ of Zn in its primary feathers. During the present study period mass mortality of Crows was reported sporadically all over the country. Attempts were made to check if the mortalities were due to pesticide poisoning and or disease outbreak. Simultaneously, attempts were also made to rule out the role of metal in terms of poisoning. The levels were found to be comparably high with the levels reported in birds (114 $\mu\text{g/g}$) collected from urban areas of Tiruchirappalli, Tamil Nadu, India (Manjula et al. 2015) and with a study that reported levels of metals in urban locations of Malaysia (206.31 $\mu\text{g/g}$) (Janaydeh et al. 2016). But, levels of zinc or any other metals studied were not indicative of any poisoning.

In the present study, levels of Zn in primary feathers of piscivorous Painted Stork ($101.29 \pm 0.28 \mu\text{g/g}$) and Spot-billed Pelican ($145.89 \pm 10.23 \mu\text{g/g}$) were higher than the Black – browed Albatross (175.35 $\mu\text{g/g}$) collected from Patagonia, Argentina falling under the same feeding guild (Pon et al. 2011) and also they mentioned levels in their study was in lower levels than those reported for Black – browed Albatross from other parts of Georgia (Anderson et al. 2010). Hence, that level of pollution in Patagonia, Argentina may not be as negligible as previously thought. But zinc is one of the fundamental element essential for feather development (Sunde 1972). Even though the concentration of zinc in the present study were higher than the previous studies, it cannot be considered harmful.

Accumulation of metals in the feathers reflect the presence of pollutants in the environment. Bird feathers are considered as important bio-indicators for metal contamination in birds (Veerle et al. 2004; Naccari et al. 2009) because feather is the main location of metal accumulation and excretion, especially in molted breast feathers

(Dauwe et al. 2005; Janaydeh et al. 2018). It is also age-dependent due to the long-term exposure to environmental pollution. These results recommend that House Crows are important model species in ecological research and a suitable and cost-effective bioindicator for environmental pollution.

5.2.2. Levels of total metals in various feather types of birds

The levels of total metals varied significantly among the feathers (One - way ANOVA $F=50.962$; $df=524$; $p<0.05$). Among the four types of feathers analyzed, breast feathers had the highest concentration of total metals followed by primary feather, while secondary feather showed the least accumulation (Figure 5.8). Similarly, the levels of chromium and copper were found to be the maximum in the primary feathers than that of breast feathers, while the levels of lead, cadmium, nickel and zinc were recorded the maximum in breast feathers (Figure 5.9). Secondary feathers had the lowest level.

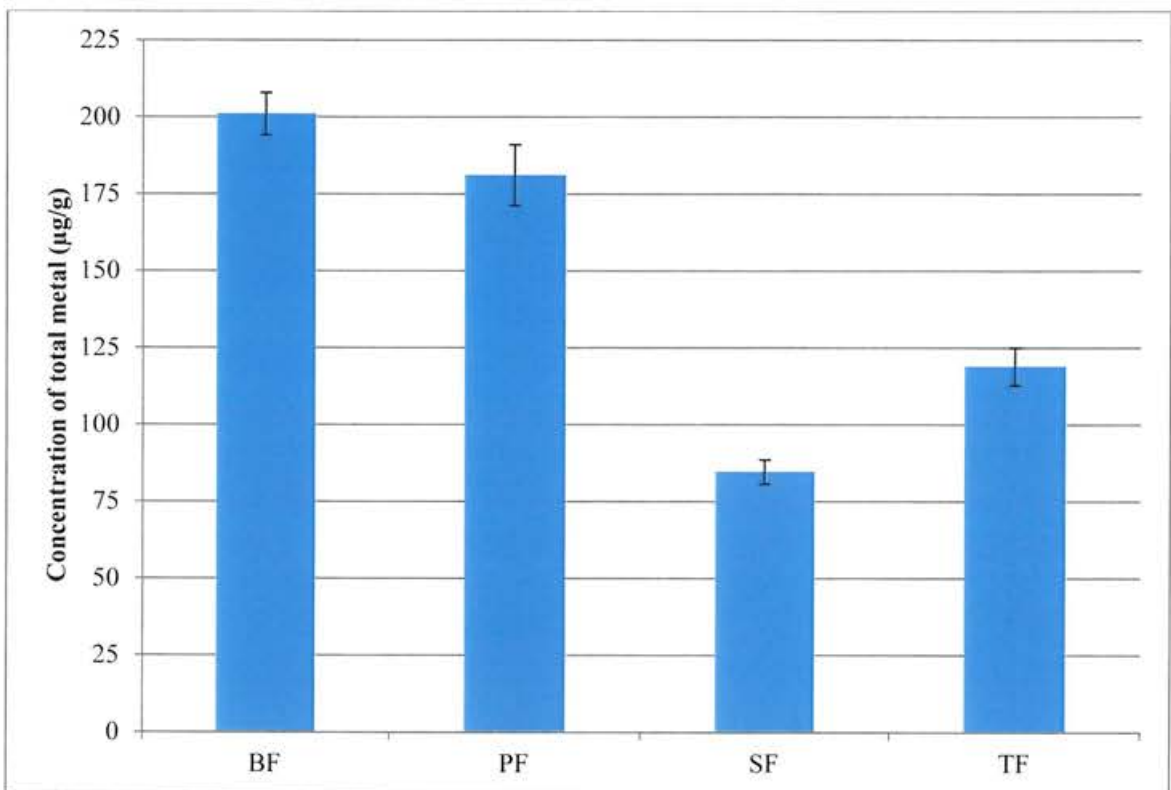


Figure 5.8 Variation in accumulation of total metal load (mean \pm SE, $\mu\text{g/g}$) among four types of feathers of birds (irrespective of species)

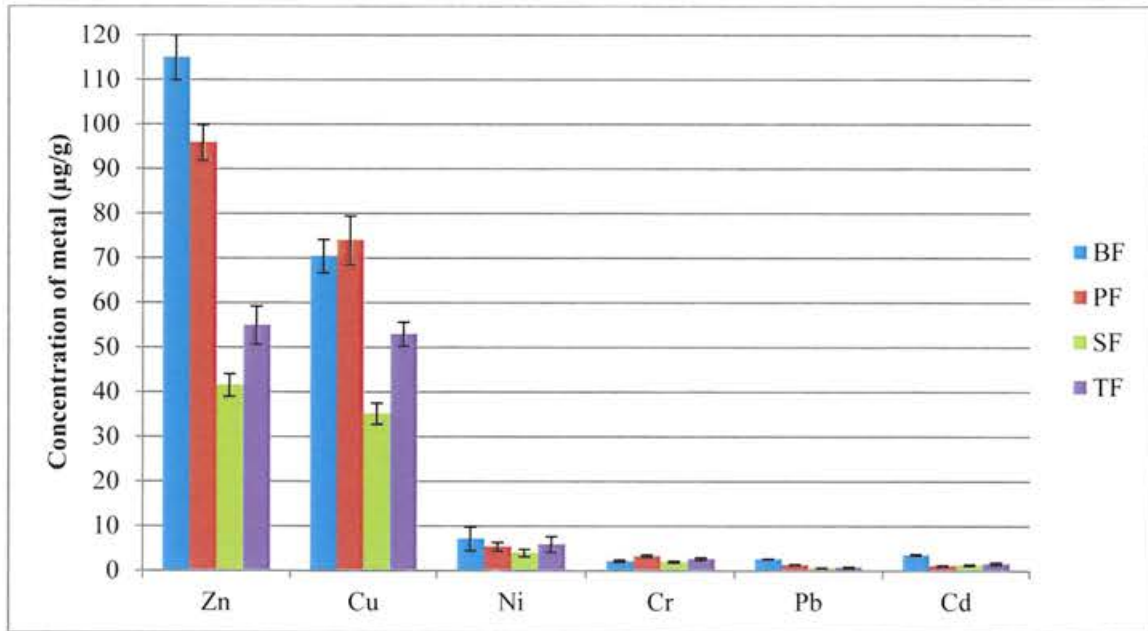


Figure 5.9 Variation levels of metal concentration in different types of feathers of birds collected between 2012 and 2014

The concentrations of Pb in breast feathers were higher than the other feathers analyzed. There are a lot of studies on the levels of metals supporting our present study indicating that breast feathers are very good indicators to understand and report metal accumulation in the birds (Movalli 2000; Lucia et al. 2010b; Tsipoura et al. 2011; Mansouri et al. 2012; Abdullah et al. 2015; Janaydeh et al. 2016; Gruz et al. 2018). But there is a lack of studies on secondary and tail feathers.

The physiological mechanisms such as secretions of uropygial glands, supra-orbital nasal glands and the affinity of metals to bind to the keratin are also notable reasons for lead, cadmium and chromium to be present at higher concentration in feathers (Howarth et al. 1981; 1982). Activities such as preening (Goede and DeBruin 1986; Pilastro et al. 1993) and presence of contaminants in external environment (Dmowski 1999) may also contribute to the concentrations of metals in feathers. The accumulation of metals in feathers are influenced by a variety of factors such as shaft- vane variation (Rose and Parker 1982), concentration of amino acids, habitat and dietary habits (Dmowski 1999), age and gender (Burger and Gochfeld 2000a), position in food-web (Solonen et al. 1999) and also moulting duration (Movalli 2000). However, in the current study, these parameters have not been investigated.

Trophic-level differences in metal levels have been reported for a number of contaminants (Burger et al. 2000a). Further, feathers of birds of prey may give useful information on environmental contamination through biomagnifications (Harmens et al. 2010; Conti and Cecchetti 2001) as they are higher on the food chain. It has been suggested that insectivorous birds accumulated more metals than those species feeding on seeds (Gochfeld and Burger 1987; Eens et al. 1999; Goutner et al. 2001) as they are higher on the food chain.

In general, metal accumulation in feathers documented in the current study were in accordance with previously reported studies (Movalli 2000; Malik and Zeb 2009; Ullah et al. 2014). The current study also substantiates the claim that the breast feather accumulated highest concentrations of metals and can be used as a bioindicator (Movalli 2000; Tsipoura et al. 2011; Mansouri and Hoshyari 2012; Manjula et al. 2015; Abdullah et al. 2015; Janaydeh et al. 2016; Gruz et al. 2018).

5.3 Correlations analysis: An attempt to study the relationship between Internal Tissue (kidney) and Feathers

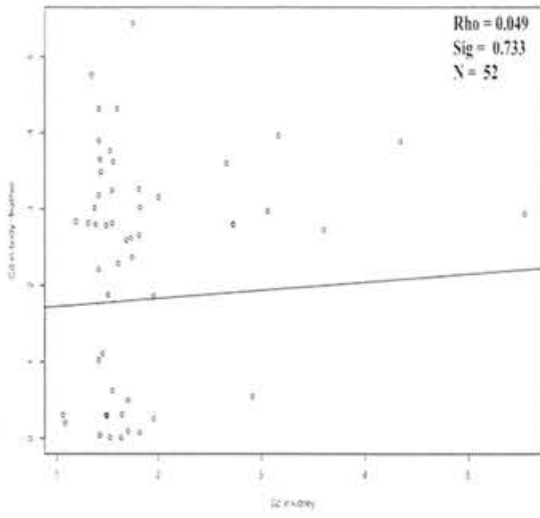
Feathers are an alternative to tissue because they are obtained non-invasively, and they reflect the metal concentration in the blood at the time of their formation and excrete totally through feathers (Goede and de Bruin 1986). A correlation study between feathers and kidney was conducted to assess if there was a relationship between the two on the basis of the possibility of re-absorption of metals in the kidneys and accumulation of divalent metals while the same is not the case in feathers (Barbier et al. 2005). Both kidney and feathers, excrete toxic metals from the body burden but re-absorption will takes plays from the kidney, whereas feather eliminates directly (Barbier et al. 2005). Metal concentrations in the breast feathers reflect the internal exposure history of the bird. The metal levels found in feathers were generally much higher as compared to internal tissues (Hahn et al. 1993). Metal levels in feathers are stable, reflecting the levels in food and blood during the feather growth (Westermarck et al. 1975; Dauwe et al. 2000), while those in soft tissues such as liver and kidney vary for various reasons (Esselink et al. 1995; Jager et al. 1996). They said, assuming that concentrations in feathers reflect circulating concentrations in the body at the time of their formation; feathers may not reflect recent

changes in contamination. Sequestering metals in feathers may be an important route for eliminating pollutants (Burger and Gochfeld 1992). Comparison of levels of metals between in feathers and other tissues in the same individual reported elsewhere are few (Swiergosz 1991; Denneman and Douben 1993; Wenzel and Gabrielsen 1995).

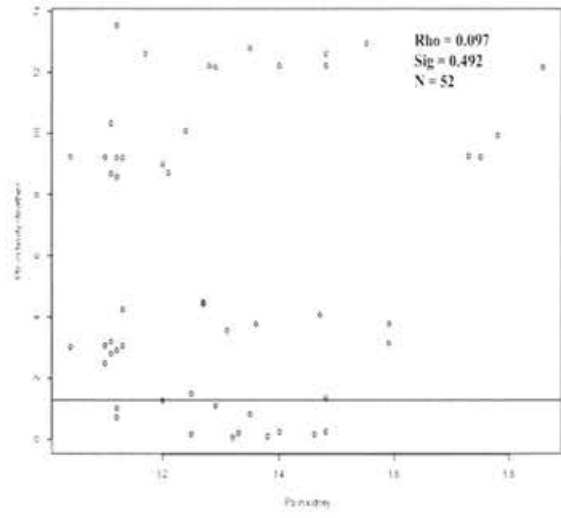
In the present study, it was found that among states, Gujarat had highest concentration of metal contamination where feather was concerned. While comparing levels among feathers namely, breast, primary, secondary and tail feathers, it was found that breast feathers had the highest load. While kidney was a better indicator internally, breast feathers had the highest levels externally. In this study, Spearman's test revealed correlations between the concentration of metals in internal organs and breast feather. Few studies have estimated the relationship of metals between breast feathers and internal tissues in wild birds (Goede 1985; Burger 1993; Markowski et al. 2013).

While a negative correlation was observed between the concentration of zinc in kidney and feathers ($r = -0.294^*$; $P=0.034$), a positive correlation was observed for Cd; Pb and Cr ($r = 0.049$; 0.097 and 0.116 respectively). Ni ($- 0.101$) and Cu ($- 0.176$) had no correlation (Figure 5.10). Non-essential metals such as cadmium and lead were positively correlated (Table 5.15). Although some authors previously reported a significant correlation between metal levels in feathers and other tissues (Gochfeld 1980; Ohlendorf et al. 1985), they found significant correlations between the levels of zinc in kidneys and feathers. Zinc was also found to be an essential element for the feather growth (Sunde 1972). Zinc has more affinity towards keratin and gets built into the feathers during growth (Muralidharan and Jayakumar 2004). Elevated cadmium concentrations in the feathers were strongly correlated with reduced growth rates of bone (Spahn and Sherry 1999).

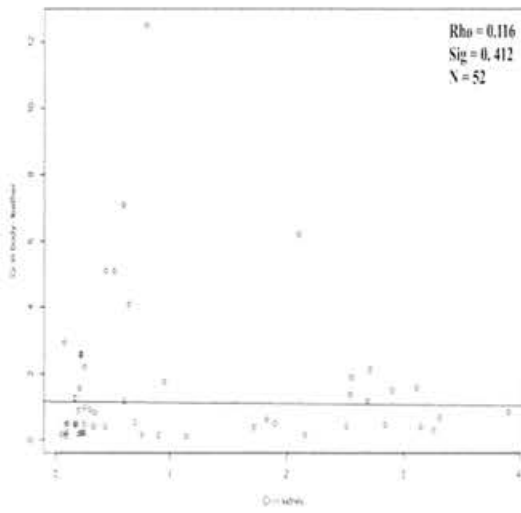
The entry of metals into the body might be from recent exposure via the diet or from mobilization from tissues from where they were sequestered (Braune and Gaskin 1987a; 1987b; Lewis and Furness 1991). Activities such as preening (Goede and De Bruin 1986; Pilastro et al. 1993) and contaminants from external environment (Dmowski 1999, Rose and Parker 1982) may also contribute. The accumulation of metals in feathers is influenced by a variety of factors such as, concentrations of amino acids, habitat and dietary habits (Dmowski 1999) age and gender (Burger and Gochfeld 2000c) and also



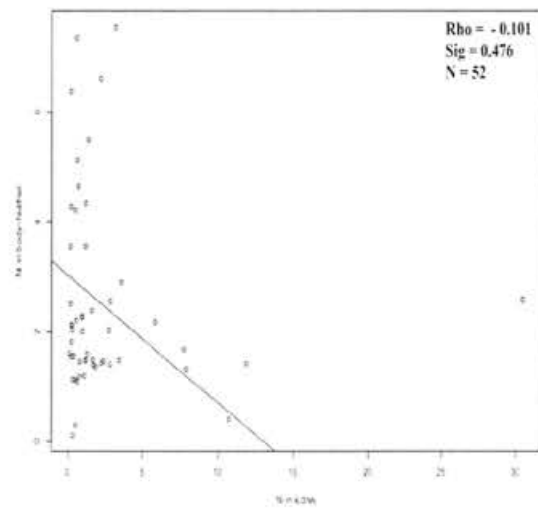
a) Cadmium



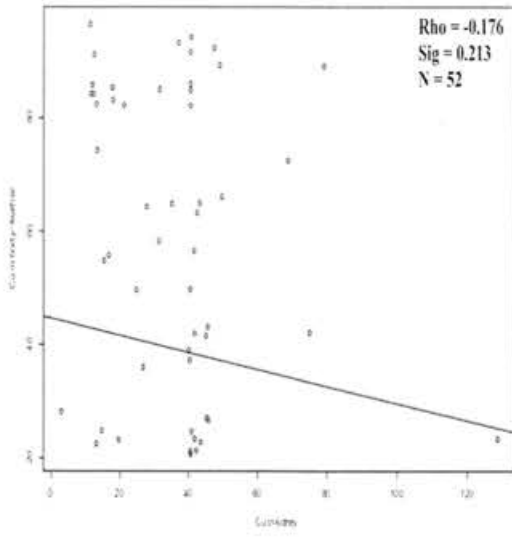
b) Lead



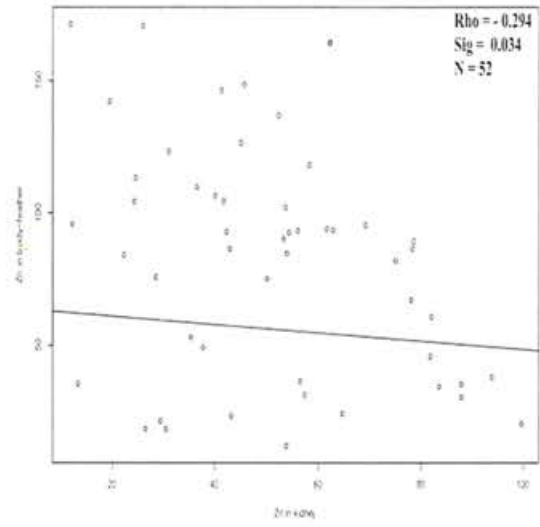
c) Chromium



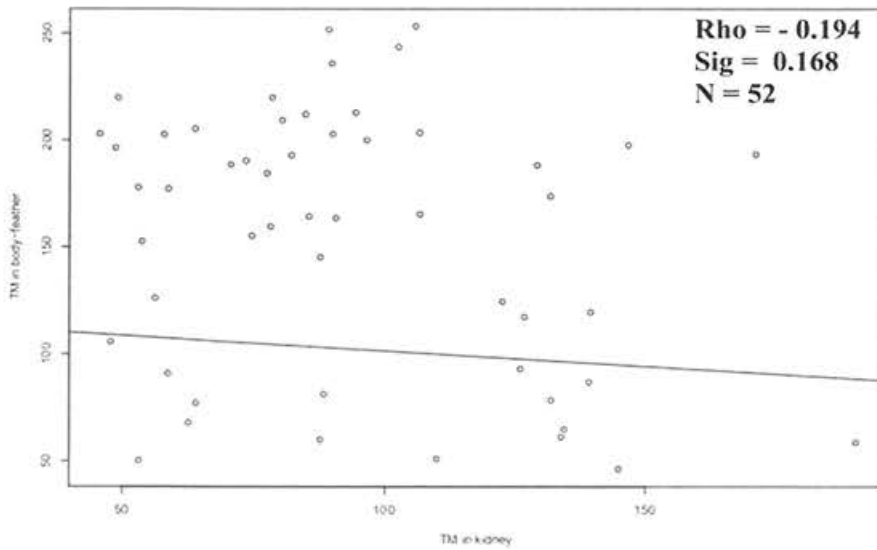
d) Nickel



e) Copper



f) Zinc



g) Total metal load

Figure 4.1. a) Cadmium b) lead c) chromium d) nickel e) copper f) zinc and g) Total metal load

Spearman's correlation of metal levels in kidney (Y axis) and body feather (X axis)

Table 5.15 Correlation of metals within kidney and feathers

Feather	Metal	Organ	Metal	Rho	Significant	N
BF	Cd	K	Cd	0.049	0.733	52
BF	Pb	K	Pb	0.097	0.492	52
BF	Cr	K	Cr	0.116	0.412	52
BF	Ni	K	Ni	-0.101	0.476	52
BF	Cu	K	Cu	-0.176	0.213	52
BF	Zn	K	Zn	-0.294	0.034	52
BF	TM	K	TM	-0.194	0.168	52

food web (Solonen et al. 1999). According to Malik and Zeb (2009), metal concentrations are greater in feathers compared with those of other tissues, therefore the metals can be easily detected and analyzed. Bortolotti (2010) has described feathers to be the “ultimate” bio-indicators of contamination.

The blood supply atrophies and the metal content in the feather remains extremely resistant to further change once it is fully grown (Burger 1993). When the feather matures, the feather remains as an indicator of the metal levels in the blood of the individual (Braune and Gaskin 1987a; 1987b).

Karimi et al. (2016) reported that lead was highest in feathers in comparison to all the other organs. Furness et al. (1986) reported that breast feathers are the best indicator of whole body burdens. It is possible however to estimate roughly the metal levels in other tissues on the basis of metal levels in feathers (Furness et al. 1986).

Lee et al. (1989) reported correlation between metal concentrations in feathers and metal concentrations in internal organs, which is comparable with our results. The advantage of the exogenic way in feather was also described by Hahn et al. (1993) and Veerle et al. (2004).

The physiological mechanisms such as secretions of uropygial glands, supra-orbital nasal glands and the affinity of metals to bind to the keratin are also notable reasons for lead, cadmium and chromium concentrations in feathers (Howarth et al. 1981; 1982). Moreover, concentrations of metals not only varied among different types of feathers, but also within a single feather itself. The differences in levels could be due to Shaft-vane variations, moulting and exposure period of feather to external contamination during flight among others (Dmowski 1999).

Contaminants are excreted from an internal system into growing feathers, and thus with the progress of moult, concentrations in internal tissues and in feathers replaced later in the same moult cycle would decrease with time (Braune and Gaskin 1987a; 1987b; Monteiro and Furness 2001). Our data further suggests that body feathers could be considered as an appropriate indicator of metal contamination.

5.4. Incidences of pesticide poisoning in different species of birds during 2012-2014

Preamble

During the study period (2012 -2014), there were 34 incidents of mass mortality of birds involving 24 species from 14 states in the country (Table 5.16).

Table 5.16. Notable incidences of mortality of birds reported across the country during the period of reporting

S. No.	Species	States	Location	Casualty	Reported cause
1	House Crow	West Bengal	Kolkata	>100	Unknown
2	House Crow	Odisha	Rourkela	30-40	Unknown
3	Spot-billed Pelicans, Painted Stork, Eurasian Spoonbills, Little Egrets, Black-headed Ibis, and Black-crowned Night-Herons	Tamil Nadu	Anna Zoological Park, Chennai	32	Phosphomidan poisoning
4	House Crow	Bihar	Ranchi	>250	Unknown
5	Flamingo	Gujarat	Khadir Island	145	Electrocution
6	House Crow	Maharashtra	Mumbai	>50	Unknown
7	Red-crested Pochard, Common Moorhen	Uttarakhand	Sitarganj Forest Range, Udham Singh Nagar District	5	Chlorpyrifos poisoning
8	House Crow	Tamil Nadu	Ettimadai, Coimbatore	Around 150	Unknown
9	House Swifts	Tamil Nadu	Madurai	150	DDT poisoning
10	Indian Peafowl	Tamil Nadu	Tiruchi	12	pesticide poisoning
11	Indian Peafowl	Tamil Nadu	Thirunelveli	17	pesticide poisoning
12	Indian Peafowl	Tamil Nadu	Vellakoil, Erode	4	pesticide poisoning

S. No.	Species	States	Location	Casualty	Reported cause
13	Indian Peafowl	Tamil Nadu	Dindigul	15	Phorate & Carbofuran poisoning
14	Rock Pigeon	Maharashtra	Pune	38	Dehydration due to temperature
15	Indian Peafowl	Tamil Nadu	Viruthunagar	6	pesticide poisoning
16	Indian Peafowl	Tamil Nadu	Thirunelveli	16	pesticide poisoning
17	Large Cormorants and Greater Pelicans	Assam	Kaziranga National Park	50	Unknown
18	Demoiselle Crane	Gujarat	Amreli	40	Phorate & rodenticide poisoning
19	Sarus Crane	Uttar Pradesh	Dadri Tehsil, Gautam Budhnagar	14	Reported to be pesticide poisoning
	House Crow	Uttar Pradesh		29	
	Common Teal	Uttar Pradesh		3	
	Ruff	Uttar Pradesh		6	
20	Indian Peafowl	Rajasthan	Jaipur	12	Poisoning
21	Indian Peafowl	Tamil Nadu	Madurai	14	pesticide poisoning
22	Black-crowned Night-heron and Purple Herons	Tamil Nadu	Sivakasi	3	Carbofuran poisoning
23	Indian Peafowl	Maharashtra	Allada	13	Poisoning
24	Indian White-rumped Vulture	Gujarat	Ahmadabad	2	Diclofenac poisoning
25	Indian Peafowl	Haryana	Nimoth village	16	pesticide poisoning
26	Indian Peafowl	Tamil Nadu	Namakkal	4	pesticide poisoning
27	Indian Peafowl	Telangana	Shabdullapuram Nalgonda	16	Reported to be pesticide poisoning
28	Indian White-rumped Vulture	Gujarat	Surendranagar district	11	Diclofenac Poisoning

S. No.	Species	States	Location	Casualty	Reported cause
29	Indian Peafowl	Telangana	Lingala Ghanapur mandal, Warangal	60	Reported to be pesticide poisoning
30	Indian Peafowl	Telangana	Allada village, Chevella	15	Reported to be pesticide poisoning
31	Indian Peafowl	Madhya Pradesh	Alirajpur	7	Reported to be pesticide poisoning
32	Indian Peafowl	Tamil Nadu	Chennimalai	10	pesticide poisoning
33	Eurasian Coot	Haryana	Sultanpur National Park, Gurgaon	33	Reported to be pesticide poisoning
	Common Moorhen	Haryana		8	
	Spot-billed duck	Haryana		1	
34	Indian White-rumped Vulture	Assam	Sivsagar	22	Reported to be poisoning
	Slender-billed Vulture	Assam		4	
	Himalayan Vulture	Assam		29	

Source: Newspaper reports: The Hindu, Times of India, Indian Express, Forest officials and NGOs in addition to our own surveys.

Based on circumstantial evidences, poisoning was suspected in the mortality of many species of birds. Subsequent laboratory analyses confirmed pesticide poisoning in seven incidents involving thirteen species of birds. Simultaneously, attempts were also made to rule out the role of metals in terms of poisoning. Only eight species of birds were considered for metal analyses because of non-availability of adequate tissues (Table 5.17). It may be noted them that in this chapter, only data on the levels of metals (Cd, Pb, Cr, Ni, Cu and Zn) are presented and the discussion is restricted except vultures. This is because under chapter 4.1, levels of metals and the negative impacts are discussed in detail. Details of the investigations carried out are furnished below;

Table: 5.17 List of species analyzed for metals

S. No.	Common Name	Number of Individuals
1	Barn Owl	1
2	Demoiselle Crane	3
3	Great Cormorants	4
4	Greater Pelicans	4
5	Himalayan Vulture	6
6	House Crow	4
7	Indian White-rumped Vulture	7
8	Slender -billed Vulture	1

5.4.1 Barn Owl

Natural predators such as Owls are environment-friendly biological waste control agents which are used to contain pest, particularly rodents (Wodzicki 1973; Singleton 1994; Johnson et al. 1996; Pande et al. 2004; 2007). The Barn Owl is one of the most widespread owls in the world and its large distribution range can be attributed to its ability to successfully compete in a variety of open habitats ranging from deserts to grasslands, marshes, agricultural lands, and metropolitan environments.

According to Marti et al. (2005), small mammals such as rodents, shrews, bats, and rabbits account for 74-100% of Barn Owl diets; other prey includes birds, reptiles, amphibians, and arthropods. Despite their smaller size, a high metabolic rate allows these owls to eat up to one-fourth of their body weight each day. Owls have a unique physiological adaptation that allows for straight forward dietary analysis. They generally swallow their prey whole or in large pieces and the indigestible parts (hair, bone, exoskeleton) are regurgitated as pellets. Hence, pellets prove useful for dietary analysis (Roest 1986).

Despite developments in the infrastructure for production and storage of agricultural produce, it is estimated that rodents damage up to 15 % of the crops annually throughout the country; (Parshad 1999). As a result, rodent control has been major concern. Use of a variety of chemical rodenticides has over a period of time led to population decline in Owls.

During this study we received one dead Barn Owl from Ahmedabad. On postmortem, we found a partially digested rat in its gizzard. Tissues and the rat were screened for a set of pesticides. But we could not identify the pesticide that might have killed the bird. Results of metal analysis are summarized below;

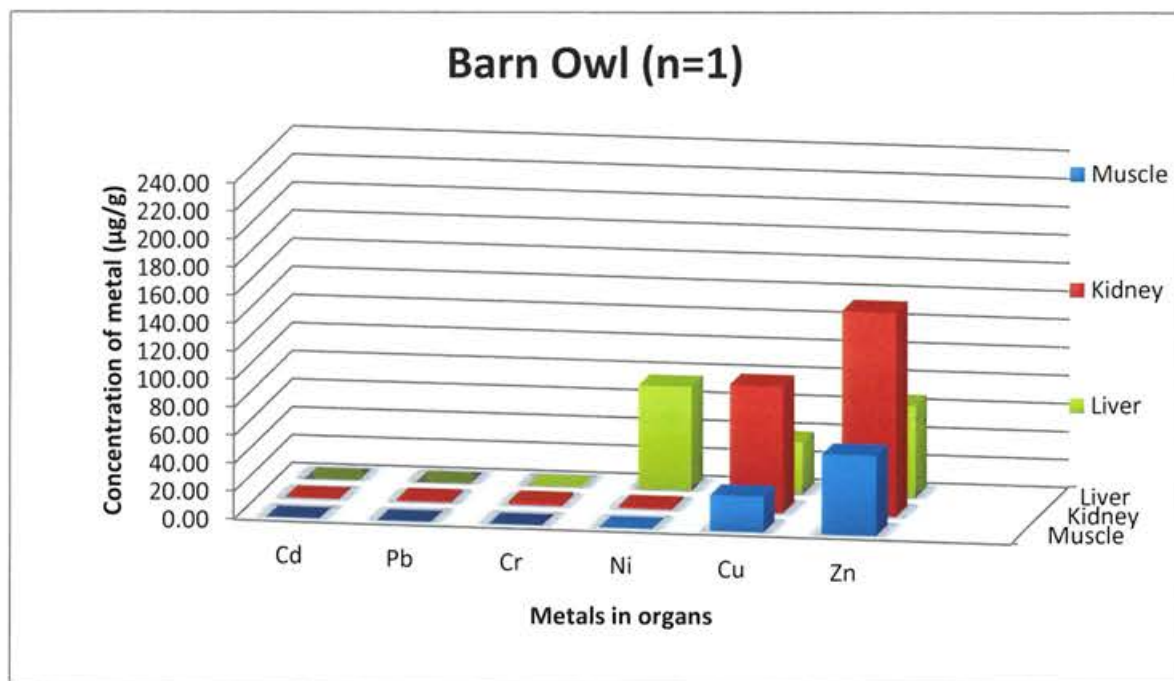


Figure 5.11. Variation in levels of metals among different organs of Barn Owl collected from Gujarat in 2013

Concentration of cadmium (0.83 µg/g), lead (1.67 µg/g), chromium (2.34 µg/g), copper (91.02) and zinc (146.06 µg/g), was found to be the highest in the kidney and lowest in the muscle (Figure 5.1). Rajamani and Subramanian (2015) reported 30.96 µg/g of zinc in the kidney of Indian White-rumped Vulture. It was almost five fold higher than what has been reported in previous literature. Since semi-digested rodent was found in gizzard (Plate 5.1), it was suspected that the bird succumbed to rodenticide poisoning. We also analyzed the semi-digested rodent for metals Cd Pb, Cr, Ni, Cu and Zn were 0.12, 0.06, 0.26, 1.68, 19.68 and 179.98 µg/g respectively (Figure 5.9). Levels of all the metals in the present study were less than level of Cd: 3.4 µg/g and Pb: 3.11 µg/g was reported in the liver of Barn Owl studied in Spain (Pe´rez-Lo´pez et al. 2008). But similar to the levels reported by Battaglia et al. 2005 from Italy. The levels of lead in muscle

(0.1 $\mu\text{g/g}$) and in liver (0.35 $\mu\text{g/g}$) were observed. Levels of cadmium in muscle and liver had (0.003 $\mu\text{g/g}$) and (0.050 $\mu\text{g/g}$) respectively.



Plate 5.1 Semi digested rat found in the gizzard of Barn Owl collected from Ahmedabad, Gujarat

Not only industrial sources, but also agricultural fertilizers and pesticides may be the source for Cd in the environment. Certain types of phosphate fertilizer were found to be rich in cadmium which was absorbed readily by crops, notably parsnips which contained 0.14 $\mu\text{g/g}$ Cd (Schroeder and Balassa 1963). Further, Zinc Phosphide is highly toxic through inhalation and oral routes (Glahn and Lamper 1983). Zinc phosphide has been used widely as a rodenticide for agricultural activities. Supporting to the current study, in Korea, Tawny Owl *Strix aluco*, cranes *Grus monachus*, *Grus japonensis*, and *Grus vipio*, Hill Pigeon *Columba repesttris*, and Oriental Stork *Ciconia boyciana* were known to scavenge carcasses and may have been secondarily poisoned by scavenging on ducks and other birds that have died from feeding in pesticide-treated fields (Hill and Menden-hall 1980; Henny et al. 1987). Unintentional poisoning appears to occur frequently and cause more mortalities than intentional poisoning. But, none of the pesticides analyzed was high enough to have killed the bird. Further, although the Zn concentration was high, it was also not in toxic range. Hence, we can presume that the bird might have died due to poisoning by some other rodenticide.

5.4.2 Demoiselle Crane

It is found in grassland habitats in close proximity to streams, shallow lakes and other wetlands, and also frequent desert areas where water is available in both its breeding and wintering (Johnsgard 1983; Ellis *et al.* 1996; Meine and Archibald 1996). Birds from Asia Mongolia and China visit India in large number during winter (Johnsgard 1983).

The diet of this species consists mainly of plant materials forage in cultivated fields such as grass seeds, feeding on ripening cereal crops in its breeding grounds (Johnsgard 1983; Snow and Perrins 1998), Peanuts, beans and other crops on migration (Meine and Archibald 1996), and wheat, chickpeas, alfalfa (Johnsgard 1983) and lucerne (Snow and Perrins 1998) in India (Johnsgard 1983; Snow and Perrins 1998), although lizards and small invertebrates such as large insects especially beetles and worms (Johnsgard 1983).

Demoiselle Crane is reported to be vanishing from the world at a disturbing rate. It is reported that about 5000 cranes are killed by sport hunters along their migration route through Afghanistan and Pakistan, every year (Sarwar et al. 2013). Extensive research and conservation efforts are being undertaken throughout the range over the last 20 years to stabilize the Demoiselle Crane population. Demoiselle Cranes are reported to undertake one of the toughest migrations in the world to reach their winter destinations in India. In late August through September, they gather in flocks of as large as 400 individuals and prepare for their flight to their wintering range and reaching altitudes of 16,000 - 26,000 feet (4,875 - 7,925 meter). In India, Gujarat is the major wintering ground for Demoiselle Cranes, and the state is extremely important for its conservation. What gets publicized and how much of it goes into official records is a minuscule percentage of the reality. Similarly, it is next to impossible to sight all the dead birds in the field for various reasons. "Forty Demoiselle Cranes found dead" was the shocking Newspaper report on the 3rd December 2012. Victor Dam at Rajula in Amreli district, Gujarat was the place which made the headlines. Between December 2012 and January 2013, a large number of Demoiselle Crane died mysteriously in several places in the districts of Rajula, Kodinar, Kutch, and Sayla. Hence, we visited the place where the Demoiselle Cranes were reported dead under suspicious circumstances and gathered circumstantial evidence. The details gathered are as follow;

During the field survey, we noticed remnants of Demoiselle Crane carcasses in two different locations in Amreli district, namely Victor and Kathiyavadar where intensive agriculture and salt pan activities take place. We also noticed large flocks in Sakriya, Thalavu, and Dharwad tanks. Water from these tanks is mainly used for irrigation. Demoiselle Cranes come in flocks for water and they remain in the tanks for long.

The flock size ranged up to 180 individuals. Apart from Demoiselles, Flamingos, Pelicans, Painted Storks and Egrets also regularly visited these tanks for drinking water and also roosting. Animals like Wild Boar, Nilgai, and domestic cattle also depend on these tanks for water. Rajula is predominantly rural with an agriculture-based economy. Around 80 % of the villagers are either agriculturists or agricultural laborers. Farmers of Rajula and its surroundings were found to be cultivating sorghum (Jowar), cotton and cereals as a major crop. Every year, the arrival of migratory birds coincides with harvesting of sorghum. Between December and January, 75% of harvesting is completed. Sorghum seeds left scattered after harvest in the fields are the major food for Demoiselle Crane. It is learnt that Acephate 75 % SP (gold star), Acetamiprid 20 % SP (prize), Phorate (Jockpot 10G), Monocrotophos 36 % SL (Monorus), Methyl parathion 2 % DP (Folidoc dust) and rodenticide such as warfarin are the major pesticides used by the farmers for insect and rodent control in this area.

On the 23rd December 2012, a Demoiselle Crane that was spotted sick the previous evening collapsed in front of us at Sakria Lake and we conducted a post-mortem of the bird in the field. Around 30 g of Sorghum seeds from the gizzard and vital organs were collected and transported to the laboratory at SACON for contaminant analysis. Excessive salivation, foam around mouth, breathing difficulties, teary eyes, inability to fly, tottered walk and bleeding in the head and beak were noticed. These symptoms are typical of rodenticide poisoning. The chemical analysis also revealed concentrations of phorate in the liver (14 µg/g) and spleen (9.68 µg/g). Hence, we did a survey among the farmers of Rajula and understood that they predominantly used warfarin, brodifacoum, difenacoum, zinc phosphide, bromadiolone, diphacinone, and chlorophacinone rodenticides. During the field study, we also observed death of Western Marsh Harrier (2 Nos) after eating poisoned Demoiselle Cranes. But, tissues could not be collected as we located the carcasses late. Although it was confirmed that the Demoiselle Cranes died due to Phorate and rodenticide, data generated on metal levels in the birds are presented briefly for further references.



Plate 5.2 Demoiselle Crane showing symptoms of pesticide poisoning

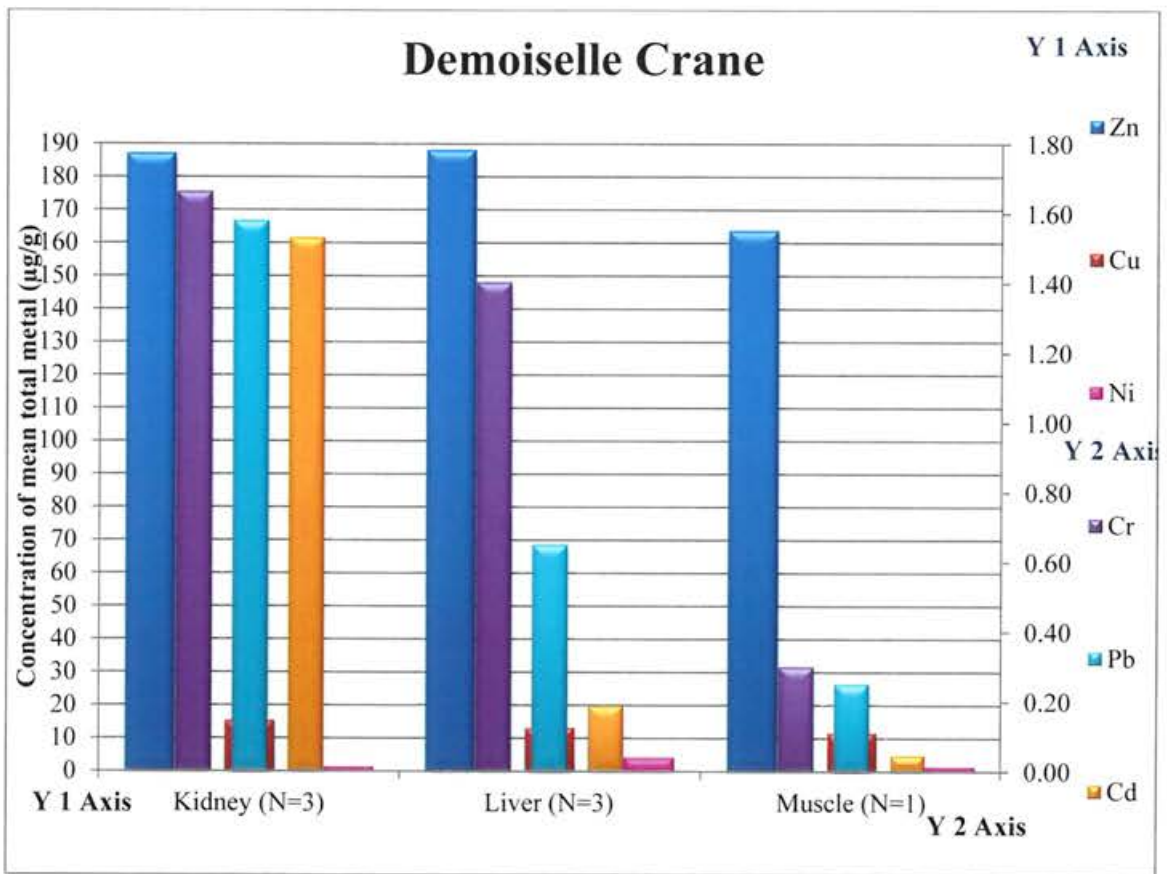


Figure 5.12. Variation in levels of metals among different tissues of Demoiselle Crane collected from Gujarat in 2012

Kidney had the highest accumulation of Cd (1.53 µg/g); Pb (1.58 µg/g), Cr (1.66 µg/g) and Cu (15.36 µg/g). Liver had highest levels of Ni (4.24 µg/g) and Zn (188.20 µg/g). Muscle had the least concentration of all the metals. There is no data available for levels of metals in internal tissues Demoiselle Crane. However, some information is available on the levels of a few metals in the eggshells of Red-crowned Cranes (Luo et al. 2014).

Extensive use of pesticides and intentional poisoning in some areas where crop damage occurs is the reason for declining population of Demoiselle Crane. Moreover, habitat loss and degradation from agriculture (e.g. conversion of steppe grassland into agricultural landscape, intensification of agricultural methods and changes in agricultural practices such as increased spring ploughing (Ellis et al. 1996; Meine and Archibald 1996) are the primary threats to this species throughout its range.

Regular monitoring programmes are ongoing in Europe but not in India. Toxicological impacts are not monitored out by both national and international agencies. Long term monitoring should be conducted not only to determine the size of the population, assess its habitat, breeding, migration and range but also on the threats from particularly pesticides, metals and other possible contaminants.

5.4.3 Great Cormorant

Great Cormorant is a gregarious fish-eater found in the inland wetlands in India. They breed locally along with other fish –eating birds colonially.

Two individuals were received dead from Assam and checked for metals. Average mean concentration of zinc (191.92 µg/g) and copper (63.46 µg/g) were the highest in liver. Remaining metals namely nickel (4.65 µg/g), chromium (5.36 µg/g), lead (2 µg/g) and cadmium (4.72 µg/g) were the highest in kidney. The total metal load was found to be 4 µg/g higher in kidney (468.11 µg/g) than liver (464.02 µg/g) respectively (Figure 5.13).

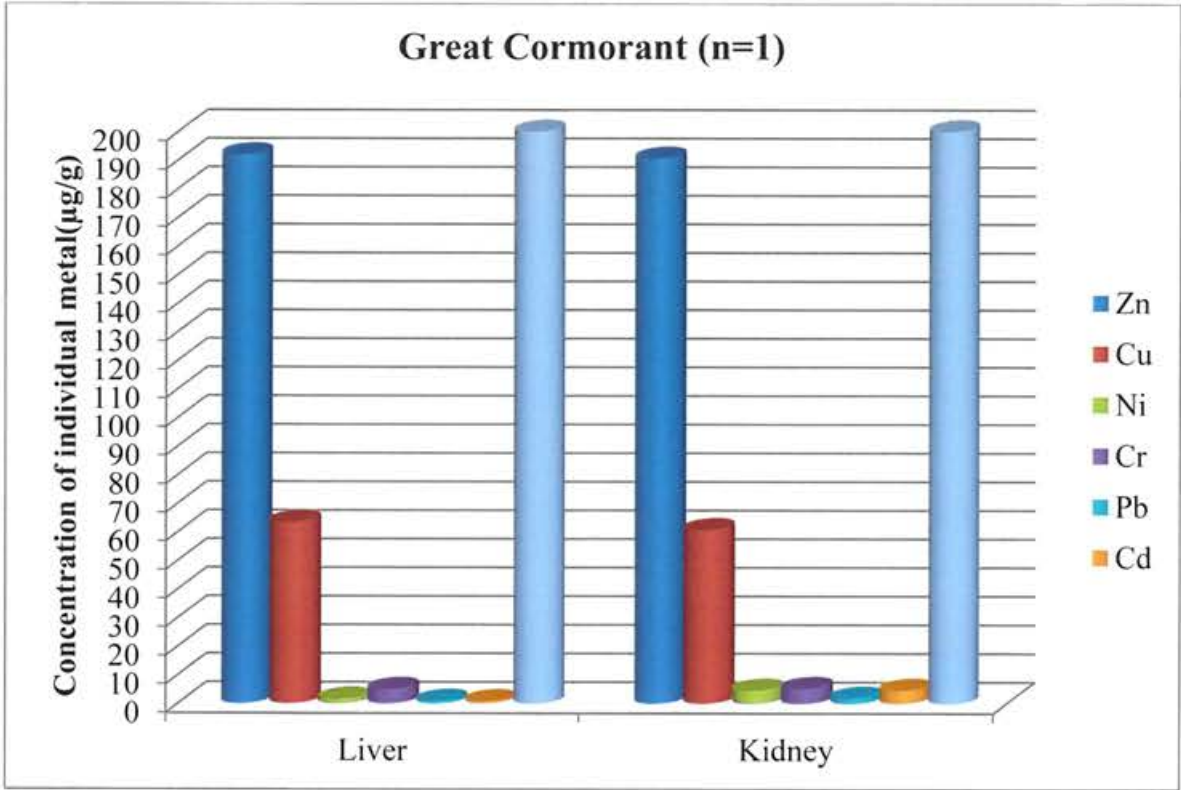


Figure 5.13. Variation in levels of metals among different organs in Great Cormorant collected from Assam in 2012

5.4.4. Great White Pelican

Great White Pelican is a piscivorous bird. It is mostly widely distributed in some parts of Africa and Pakistan. In India it is distributed in Gujarat, Madhya Pradesh, Rajasthan and Maharashtra and also in some parts of Assam, Bangladesh and Myanmar (BirdLife International (2019)).

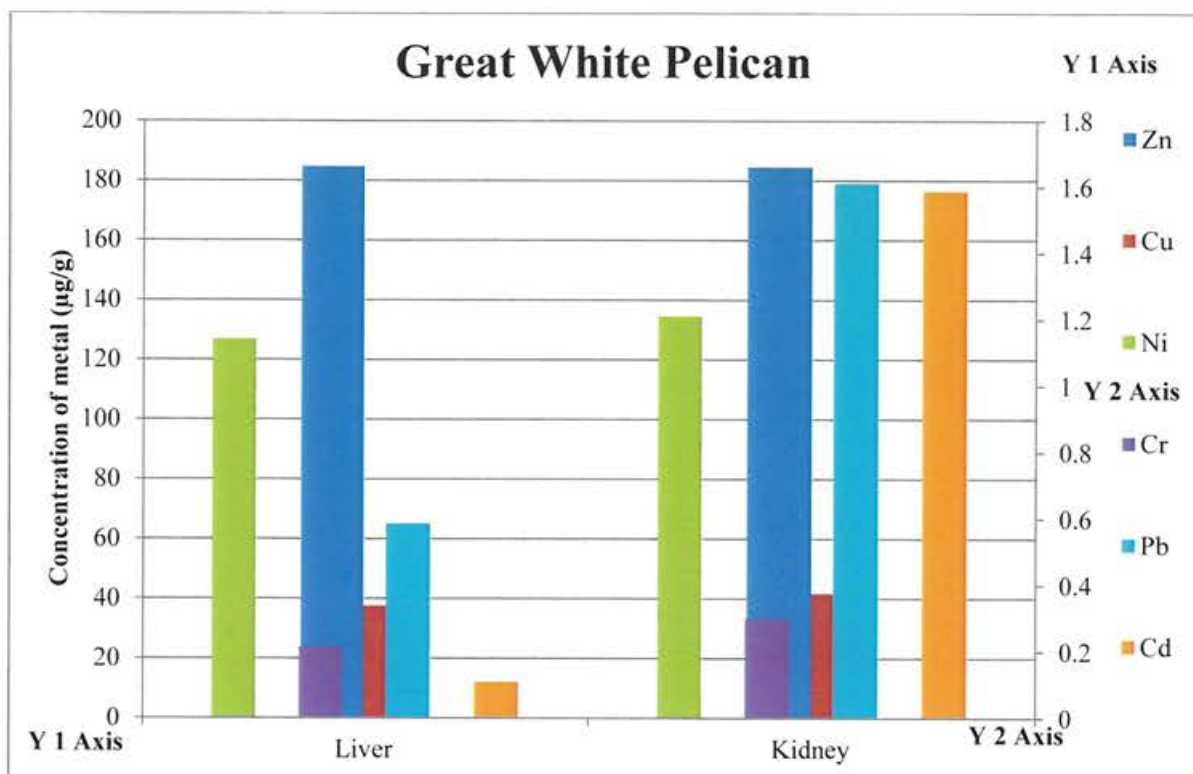


Figure 5.14 Variation in levels of metals among different organs in Great white Pelican collected from Assam in 2012

One individual was received dead from Assam. Tissues, namely liver and kidney were analyzed for metals, namely cadmium (1.58 µg/g), lead (1.61 µg/g), chromium (0.3 µg/g), nickel (1.21 µg/g) and copper (41.72 µg/g) were higher in kidney than liver. Level of zinc was the same in both liver and kidney (184 µg/g) (Figure 5.14).

5.4.5. *Gyps* species

Gyps species, namely Indian White-rumped Vulture, Indian Vulture *Gyps indicus*, Slender-Billed Vulture *Gyps tenuirostris* and Himalayan Vulture *Gyps himalayensis* are distributed in different parts of India. Populations of three *Gyps* species, namely the White-rumped Vulture, Indian Vulture and Slender-Billed Vulture endemic to South Asia, have decreased by more than 90% since the mid 1990s, and are declining at an alarming rate (Prakash et al. 2017). Since then, catastrophic declines have continued to be reported across the subcontinent.

Initially, pesticides were suspected to be the reason for the decline of vulture population but the hypothesis had to be ruled out due to lack of evidence (Prakash 1999). Oaks et al. (2004); Green et al. (2004); Shultz et al. (2004) diagnosed Diclofenac as the reason for the large scale mortality and population decline. Although subsequent studies have documented accumulation of pesticides, PCBs, PAHs, in vulture, the concentration of the referred contaminants were too low to be indicative of toxicity (Muralidharan et al. 2008; Dhananjayan and Muralidharan 2013).

Diclofenac is a non-steroidal anti-inflammatory drug (NSAID), used as an analgesic, anti-inflammatory and antipyretic (Todd and Sorkin 1988). This drug, mostly available in the form of diclofenac sodium, is widely used all over the world in palliative care, especially as a pain reliever. The drug was used by veterinarians to treat inflammation in domestic livestock extensively in the 1990s. It was evident that vultures scavenging on cattle carcasses those were treated with diclofenac shortly before death, died due to poisoning (Oaks et al. 2004; Nambirajan et al. 2018).

In the present study, between 2012 and 2014, we received 47 vultures comprising two species, namely Indian White-rumped Vulture (32) from Gujarat and Himalayan Vulture *Gyps himalayensis* (12) and three individuals of Slender-billed Vulture dead, from Assam. The sampling strategy was opportunistic.

To fix the cause of death, tissue samples were tested for a set of NSAIDs, pesticides and metals. It was established that the birds died due to Diclofenac poisoning (Nambirajan et al. 2018). Information generated on metal contamination (Cd, Pb, Cr, Ni, Cu and Zn) is summarized here.

5.4.5a Indian White-rumped Vulture

Totally six individuals were analyzed for metals. It was found that the kidney accumulated maximum of cadmium (3.16 µg/g), lead (1.90 µg/g) and copper (45.87 µg/g). The remaining three metals, namely chromium (2.25 µg/g), Ni (10.33 µg/g) and Zn (179.76 µg/g) were higher in liver than kidney. Total metal load was the maximum in kidney (234.14 µg/g) followed by liver (233.16 µg/g). Minimum was in muscle (206.59 µg/g). (figure 5.6) shows zinc and copper on Y1 axis and Ni, Pb Cr and Cd on Y2 axis.

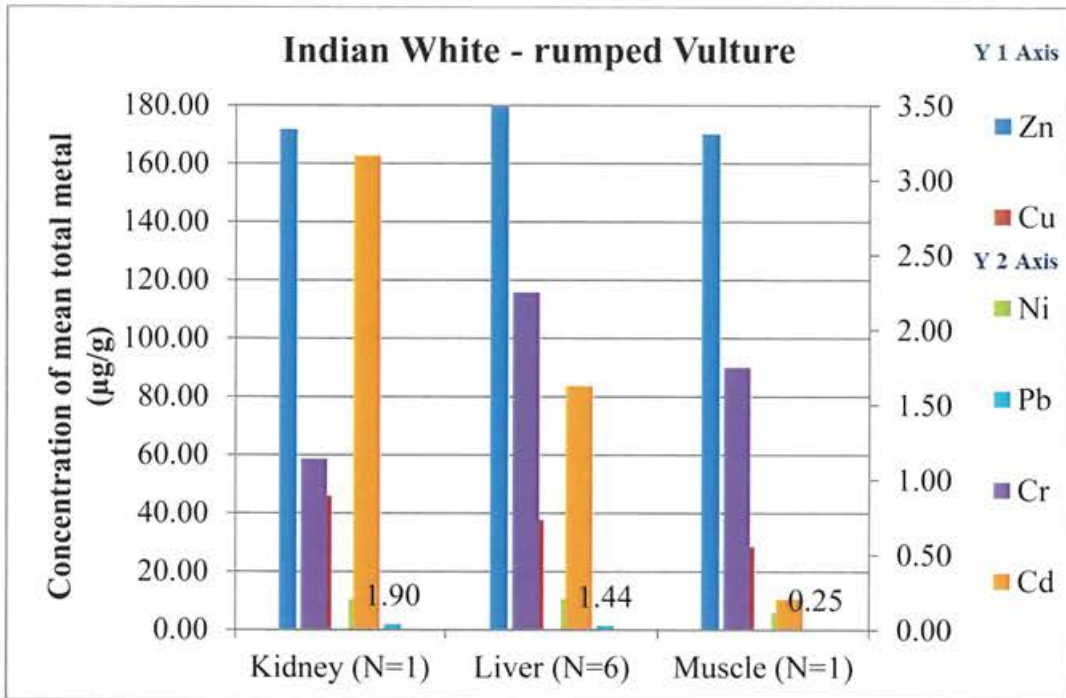


Figure 5.15 Variation in levels of metals among different tissues of Indian White-rumped Vulture (n=6) collected from Gujarat between 2012 and 2014



Figure 5.3 Indian White-rumped Vulture with visceral gout in liver

5.4.5b Himalayan Vulture

According to Birdlife International 2019, Himalayan Vulture is distributed in Nepal, Bhutan and North Asian countries. This species has native non-breeding population in Maharashtra, West Bengal, Gujarat, Chhattisgarh, Rajasthan, Uttar Pradesh, Punjab, and Himachal Pradesh. It has been up listed under Near Threatened category as it is suspected that it will undergo a moderately rapid population decline over the next three generations.

Totally six individuals were collected from Assam and analyzed for metals. The levels of cadmium (2.03 µg/g), lead (1.70 µg/g), chromium (1.93 µg/g), copper (47.19 µg/g) and zinc (191.74 µg/g) were the highest in the kidney, and the level of nickel (0.88 µg/g) was the highest in the liver. Unpredictedly chromium (3.06 µg/g) and zinc (185.78 µg/g) were found to be the maximum in muscle than liver (figure 5.14).

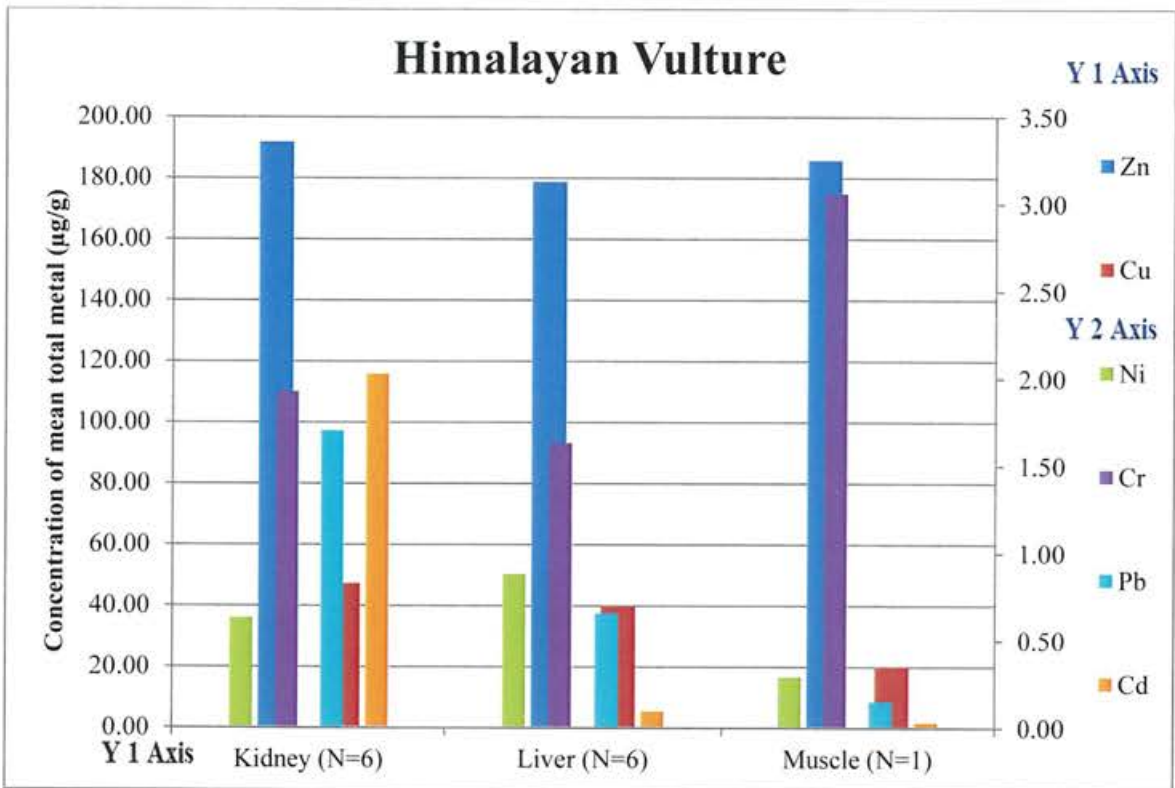


Figure 5.6 Variation in levels of metals among different organs in Himalayan Vulture (n=6) collected between 2012 and 2014

Altogether, in tissues of the three species vultures, the levels of cadmium accumulated in kidney were comparatively higher than the levels recorded in the liver. In raptors, cadmium accumulation is largely observed in kidney. However, liver also tends to accumulate cadmium owing to the availability of binding sites and extreme stability of the metal thereby reflecting the major body burden. Cadmium level recorded in kidney and liver in the present study are twice the levels reported in Cinnerous Vulture (liver-0.2 µg/g; kidney 0.13 µg/g) from Korea (Nam and Lee 2009). Scheuhammer (1987) had suggested that cadmium concentrations of >3 µg/g in liver and >8 µg/g in kidney due to elevated environmental exposure was indicative of increased risk of renal failure. However, cadmium levels recorded in the present study are not expected to create any toxicity.

Vultures belong to the order Accipetriformes. African White-rumped Vulture, *P.seudogyphs africanus* was found to be a suitable surrogate (Swan 2006) to Vultures in India. Information pertaining to metal contamination in vultures is largely lacking in India, but is available from elsewhere (Stansley and Murphy 2011) in the world. The present study attempts to assess whether metal toxicity is also a factor for population decline in this species.

In the present study, all the vultures had lead accumulation which was less than the average levels reported in African White- rumped Vulture, *P. africanus* (liver- 4.53 µg/g; kidney – 3.45 µg/g) Van Wyk et al. (2001) and Cinneroous Vulture *Aegyptius monachus* (liver-3.45 µg/g; kidney-3.45 µg/g) (Nam and Lee 2009). The mean lead levels reported in the liver (7.29 µg/g) and kidney (7.11 µg/g) are twice as high as the levels reported in the globally threatened species *A. monachus* in Southern Primoreye, Russia (Kavun 2004). It is to be noted that the reported lead levels in *A. monachus* were not considered to be toxic. Similar lead levels were observed in liver and kidney of vultures collected from carcass dumping yard at Patiala, Punjab and city market Ahmedabad, Gujarat. The autopsy report on these particular individuals also revealed that they had emaciated pectoral muscle with very little adipose tissue distended gall blader and green bile stain in the liver with lesions which are general indications of lead poisoning (Nam and Lee 2009).

Generally, vultures, outside protected area, feed on cattle carcasses and they do have ample chances of picking up contamination through food (Rajamani and Subramanian 2015). Cattle population in India is abundant and lead poisoning in bovines is also reported owing to contaminated fodder and feeding in solid waste dumping yards (Sidhu et al. 2006). The risk due to lead cannot be ignored as lead can inhibit alpha aminolevulinic acid dehydratase activity and affect the avian population by altering the hormonal regulation of calcium thereby disturbing the reproductive success, behavioral and immune responses, and physiology (Burger and Gochfeld 2000).

In the present study, in all the species of vultures, copper was found to be higher than the levels reported in the liver of Indian White-rumped Vulture from Patiala which had 57.7 µg/g (Rajamani and Muralidharan 2015). The levels reported in African White-rumped Vulture (kidney – 3.85 µg/g; liver – 9.73 µg/g; muscle- 4.78 µg/g) (Van Wyk et al. 2001) and Cinereous vulture (liver- 11.32 µg/g; kidney-3.52 µg/g) were also higher than the presently recorded levels. But these studies did not report any ill effects.

Zinc levels in the tissues of all three species of vultures analyzed in the present study are higher than the levels recorded in African White-rumped Vulture (liver – 9.84 µg/g; kidney – 38.72 µg/g; muscle – 18.91 µg/g) (Van Wyk et al. 2001), but comparable to the levels in Cinnerous vulture (liver - 42.5 µg/g; kidney - 24.76 µg/g). Further, the levels of zinc reported by Kim et al. (2008) in Brown hawk Owl Korea are higher than the levels presently reported. However, the impact of Zn on White-rumped Vulture is unclear. The levels in present study are not harmful, but absence of perceivable differences among tissues shows that essential elements may be uniformly distributed.

In this study, although levels of lead in liver and kidney of birds are indicative of toxicity, this may not ascertain the population crash in vultures in India. Nevertheless, accumulation of metals particularly lead at sub-clinical levels could make vultures susceptible to diseases and environmental stress (Perez-Lopez et al. 2008). Moreover, diclofenac and metal together may also cause synergistic effect (Rajamani and Muralidharan 2015). A continuous monitoring of the impact of contamination including metals on vultures is necessary for protecting the species in Indian subcontinent.

5.4.5c Slender-billed Vulture

Slender-billed Vulture is distributed in Nepal, Bangladesh, myanmar, Uttar Pradesh, West Bengal and New Delhi. This species is at high risk of global extinction and listed as Critically Endangered (BirdLife International 2016) and categorized under Schedule I of Indian Wildlife Protection Act 1972; 2002 (Amended). Only one individual of Slender-billed Vulture was collected from Assam. The levels of all six metals, namely Cd (1.98 $\mu\text{g/g}$), Pb (1.90 $\mu\text{g/g}$), Cr (3.49 $\mu\text{g/g}$), Ni (6.31 $\mu\text{g/g}$), Cu (42.95 $\mu\text{g/g}$) and Zn (190.11 $\mu\text{g/g}$) were higher in the kidney than liver (Figure. 5.17).

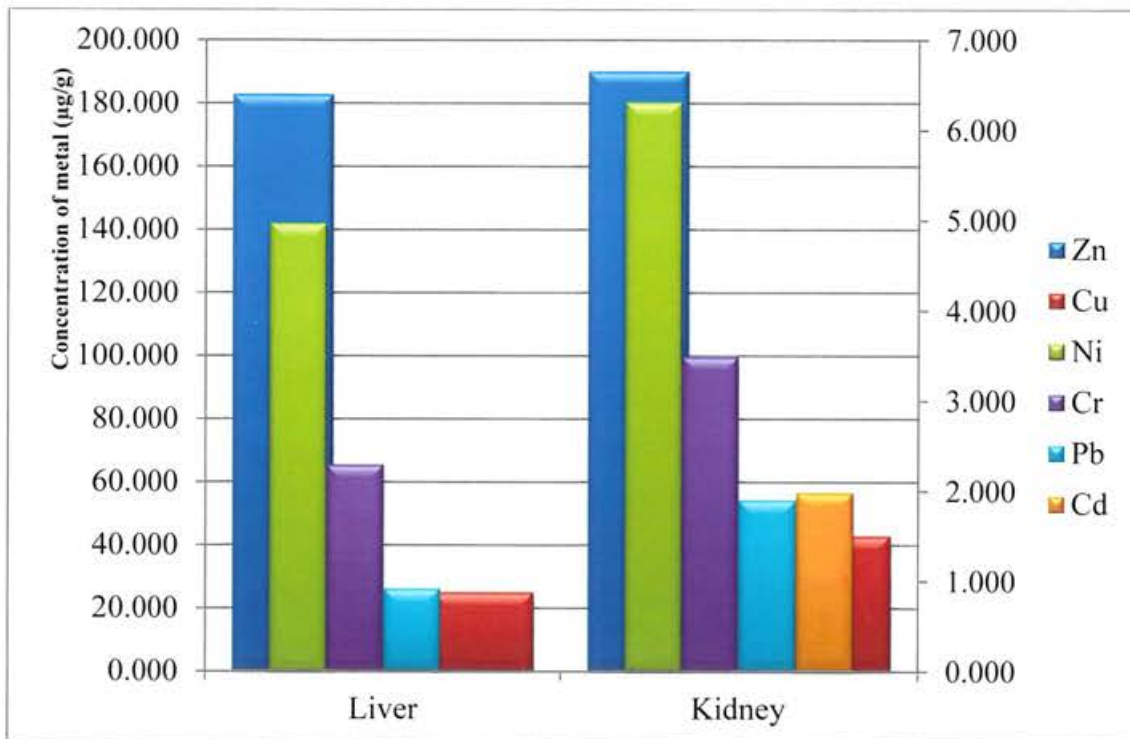


Figure 5.17 Variation in levels of metal among liver and kidney in Slender billed vulture collected from Assam in 2012

5.4.6 House Crow

House Crow is non-migratory (Birdlife International 2019) and native to an extensive area of India and central Asia from southern coastal Iran through Pakistan, India, Tibet, Myanmar and Thailand to southern China as well as Sri Lanka, the Laccadives and the Maldives (Department of Agriculture and Food 2008). It lives in human habitation and cultivation (Nyari et al. 2006).

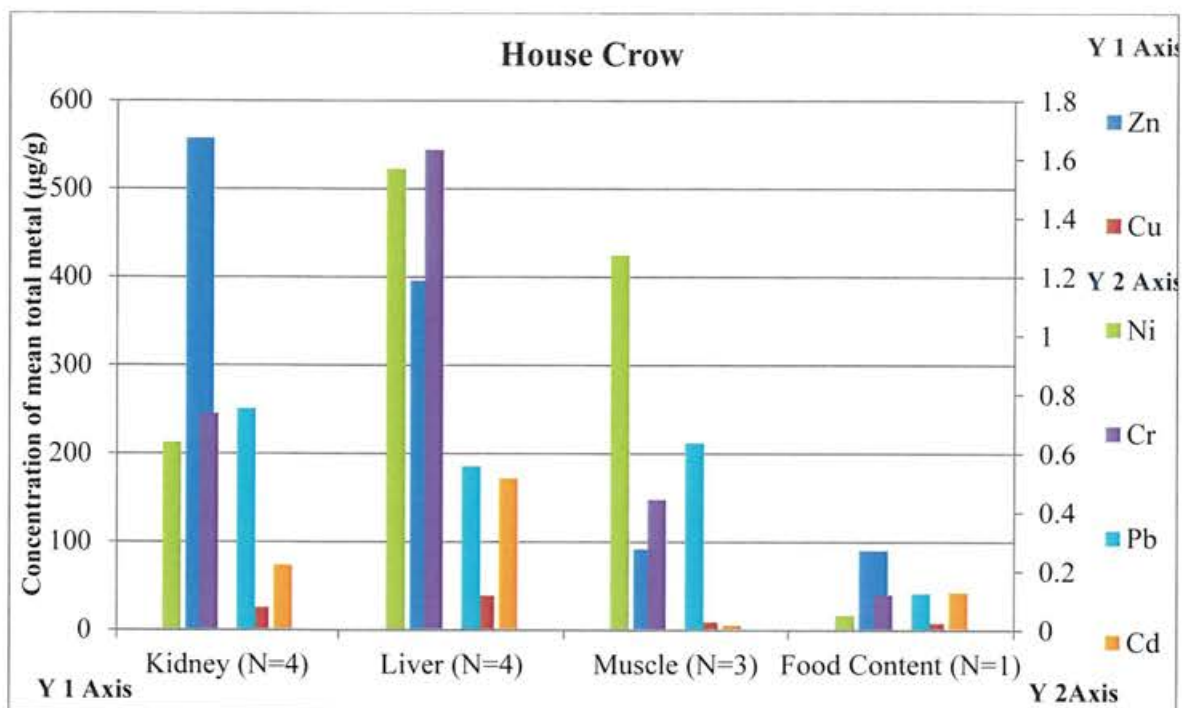


Figure 5.18 Variation in levels of metals among different organs in House Crow (n=4) collected between 2012 and 2014

It is an omnivorous species and scavenges on almost all human refuse in and around towns and cities. The diet of the House Crow can be described as broad and highly opportunistic. It includes seeds, fruits, grains, nectar, berries, birds' eggs, nestlings, mammals, reptiles, amphibians, fishes, insects, carrion and food scraps. They spend much of their time searching for food. It is not migratory to any countries in the world (BirdLife International 2019).

During 2010 and 2012, a large number of deaths were reported all over the country. In 2010, more than 100 deaths were reported from Kolkata and around 30-40 from Odisha. In 2011, more than 50 deaths were reported from Maharashtra, and more than 250 died in Bihar wherein the forest department investigated and reported flu to be the reason for the mass mortality. In 2012, around 150 individuals died in Coimbatore, Tamil Nadu (Figure 5.18). We received House Crows samples from Jharkhand and Tamil Nadu. Since the sample size, and quantity was inadequate metal analyses were carried out only for samples which were received from Tamil Nadu.



Plate 5.4 House Crow collected dead from Coimbatore during 2011 - 2012

The present study analyzed the tissues for metals which were collected from Tamil Nadu. Concentrations of lead ($0.75 \mu\text{g/g}$) and zinc ($557.26 \mu\text{g/g}$) were higher in the kidney, whereas the concentrations of cadmium ($0.51 \mu\text{g/g}$), chromium ($1.63 \mu\text{g/g}$), nickel ($1.56 \mu\text{g/g}$), and copper ($39.03 \mu\text{g/g}$) were the highest in the liver (Figure 5.18). The levels of cadmium recorded in gizzard ($0.13 \mu\text{g/g}$) in the present study were more or less equal to the levels of cadmium reported by Kler et al. (2014) in the excreta ($0.12 \mu\text{g/g}$) of House Crow in Punjab.

Zinc was found to be higher in all the tissues when compared to the levels recorded in the above referred study (Kler et al. 2014). There is no study available on metal levels in tissues of House Crow in India. House Crow could be best ecotoxicological model to predict the environmental condition since they usually live near human habitations and feed on the waste food left by humans.

Over all, the concentrations of metals do not appear to be responsible for the mortality of any of the species investigated. However, there is possibility of synergistic effects.

Summary and Conclusion

6. Summary and Conclusion

Metals are considered as critical contaminants in the environment, as they are ubiquitous, highly persistent, non-biodegradable and capable of accumulating in food chains. However, a large number of metals in trace quantities are also essential for biological sustenance in all animals including birds. Virtually, even essential metals can turn toxic to an organism. Birds are one of the major victims of metal contamination as they occupy a wide range of trophic levels. Over the years, among wildlife, birds have served as bioindicators for a number of environmental contaminants, especially metals as they are visible, widely distributed in the ecosystem, sensitive to toxins, high on the food chain, ecologically versatile, and also as they live in diverse habitats.

In India, studies on metal contamination on birds are a few. There is a growing concern to examine metal contamination in birds and interpret their levels; thereby appropriate measures may be taken to conserve the species. Metal levels in tissues can serve as an indication of the potential effects for alerting managers and the public to mitigate ecological problems. The presence of metal contaminants in tissues of birds is a problem that has not been fully resolved, given the ethical and legal impediments associated with the hunting and killing of bird species. The use of body feathers for studying metal contamination is another possible solution to this problem, as removal of these feathers does not injure the birds. In reference to seabirds, breast feathers are especially useful for establishing spatial and temporal patterns in bird populations and for evaluating metal contamination in species that are threatened or in danger of extinction.

The present study was conducted with an objective to document the levels of accumulation of metals such as Cd, Pb, Cr, Ni, Cu and Zn in different species of birds in select states of India.

The present observations and earlier reports have revealed that the chronic exposure of potential toxic metals were insufficient to produce outright mortality or other acute effects and may lead to profound consequences on birds such as decreased reproductive function, increased susceptibility to diseases or other stresses and changes in behavioral patterns. These observations further throw light that significant fluctuations in contaminated levels are present in the environment where these birds live.

The dead birds were collected from various parts of the country on opportunistic basis as explained elsewhere. Upon autopsy, select organs such as muscle, liver, kidney and gizzard were dissected out to study the metal levels. Tissues were digested in Microwave Digestion System using triple acid digestion method. Digested solutions were analyzed using Atomic Absorption Spectrometer for metals such as Cd, Pb, Cr, Ni, Cu, and Zn. Appropriate quality control measures were followed during the analyses.

Levels of metals in various species of birds

Data on 197 individuals comprising 31 species of birds are presented in detail. The maximum level of total metals was recorded in Spot-billed Pelican ($243.63 \pm 10.77 \mu\text{g/g}$) and the minimum in Emerald Dove ($97.28 \pm 8.19 \mu\text{g/g}$). The maximum concentration of cadmium ($3.54 \pm 1.68 \mu\text{g/g}$) in kidney, chromium ($4.60 \pm 1.54 \mu\text{g/g}$) and copper ($53.14 \pm 14.13 \mu\text{g/g}$) in liver tissue were recorded in the Spot-billed Pelican.

The maximum concentration of lead was recorded in kidney tissue of Indian Pitta ($2.39 \mu\text{g/g}$) followed by Slender-billed Vulture ($1.90 \mu\text{g/g}$), Indian Peafowl ($1.84 \pm 0.02 \mu\text{g/g}$), liver tissue of Indian-spotted Eagle ($1.73 \mu\text{g/g}$) and kidney tissues of Himalayan Vulture ($1.70 \pm 0.07 \mu\text{g/g}$). The levels of lead in kidney tissues of Demoiselle Crane, Sarus Crane, Painted Stork, and Griffon Vulture were 1.58, 1.48, 1.43 and 1.46 $\mu\text{g/g}$ respectively. The levels were Below Detection Level (BDL) in the muscle tissues of Emerald Dove, Demoiselle Crane, Sarus Crane, Red-wattled Lapwing and Black Kite. The levels of lead observed in Indian White-rumped Vulture included in the present study ($1.43 \mu\text{g/g}$) were lower than the previous research by Rajamani and Subramanian (2015). Levels of lead between 2.0 and 8.0 $\mu\text{g/g}$ in liver tissues and 2.0 and 6.0 $\mu\text{g/g}$ in kidney tissues in wild birds (particularly raptors) cause “toxic” and “fatal” effects respectively. Based on these guidelines, lead observed in the present study in liver and kidney tissues are not toxic. Continued monitoring of the impact of potential contaminants including metals on birds is necessary for protecting the species in the Indian Sub-continent.

Among the species analyzed, the Spot-billed Pelican had the highest concentration of chromium ($4.60 \pm 1.54 \mu\text{g/g}$) in liver followed by liver of Common Kingfisher ($4.03 \mu\text{g/g}$), kidney of Indian Spotted Eagle ($3.98 \mu\text{g/g}$), livers of Asian Koel ($3.90 \pm 1.46 \mu\text{g/g}$) and Painted Stork ($3.09 \pm 0.65 \mu\text{g/g}$). The lowest level was observed

in muscle tissues of Rose-ringed Parakeet ($0.21 \pm 0.04 \mu\text{g/g}$). Levels were Below Detection (BDL) in Himalayan Vulture, liver tissues of Slender-billed Vulture, muscle tissues of Indian Spotted Eagle, Indian Roller and kidney tissues of House Crow.

In the present study, Indian White-rumped Vulture had lower level of chromium than those levels reported by Van wyk et al. (2001) in African White-backed Vulture in South Africa. The levels of chromium recorded in various tissues of birds in the present study were lower than the levels which could cause stunted growth and abnormalities in birds.

Of all the species of birds studied, the levels of nickel in Indian White-rumped Vulture were found to be the highest in kidney tissues ($10.85 \pm 0.78\mu\text{g/g}$). The second maximum concentration of Nickel was recorded in kidney tissues of Griffon Vulture ($10.82 \pm 9.80 \mu\text{g/g}$), followed by liver tissues of Indian White-rumped Vulture ($9.37 \pm 1.38 \mu\text{g/g}$). The levels of nickel in liver and kidney tissues of Indian Pitta, liver tissues of Common Kingfisher and Painted Stork were 8.82, 7.69, 7.68 and 7.12 $\mu\text{g/g}$ respectively, and the levels varied significantly among the species. Below Detection Level (BDL) were observed in liver tissues of Bar-headed Goose and Common Coot, muscle tissues of Emerald Dove, Rock Pigeon, Asian Koel, Himalayan Vulture, Shikra and Indian Roller. The levels of nickel recorded in Cattle Egret and Bar-headed Goose included in the present study were higher than the levels reported by Honda et al. (1985) and Gochfeld and Burger (1987).

The levels of nickel observed in the muscle and liver tissues of Indian White-rumped Vulture were lower and the levels reported in kidney were higher than the African White-rumped Vulture collected from South Africa (Van wyk et al. 2001).

The highest concentration of copper was recorded in liver tissues of Spot-billed Pelican ($53.14 \pm 14.13 \mu\text{g/g}$) followed by kidney tissues of Eurasian Vulture ($52.70 \pm 8.34 \mu\text{g/g}$), liver tissues of Red-napped Ibis ($51.64 \pm 9.20 \mu\text{g/g}$) and kidney tissues of Indian Spotted Eagle ($51.11 \mu\text{g/g}$). The levels of copper in muscle tissues of Spot-billed Pelican, kidney tissues of Red-napped Ibis, Himalayan Vulture, Sarus Crane, Slender-billed Vulture, Roller and Great White Pelican were 42.43, 42.93, 43.47, 44.33, 47.18, 47.62 and 48.92 $\mu\text{g/g}$ respectively. The minimum levels of copper were recorded in muscle tissues of Black

Drongo (10.52 $\mu\text{g/g}$) and Asian Koel (11.11 $\mu\text{g/g}$). A significant variation was observed in the levels of cadmium detected among the species.

The levels of copper recorded in the Rock Pigeon and Eurasian Collared Dove in the present study were far less than the levels reported in previous studies. The levels of copper reported in Indian Pond Heron species in the present study were lower than the levels reported in the same.

Copper, although is one of the essential trace elements it can turn toxic to organisms and organs at higher concentrations. High concentration of copper can also cause public health hazards (Brito et al. 2005). Copper concentrations in birds are usually elevated in areas of human activity due to intensive copper use (Eisler 2000). Copper contamination has been reported in other wild birds without any signs of toxicity (Clausen and Wolstrup 1978; Kim et al. 1996). However, the levels reported in the present study were lower than the levels that could cause problem to birds.

Among the 31 species of birds included in the study, concentration of zinc was found to be the highest in kidney of Sarus Crane (193.67 $\mu\text{g/g}$), followed by Painted Stork (193.65 \pm 0.13 $\mu\text{g/g}$), Indian Peafowl (191.90 \pm 1.19 $\mu\text{g/g}$) and Himalayan Vulture (191.74 \pm 2.02 $\mu\text{g/g}$). The levels recorded in kidney tissues of Slender-billed Vulture, Barn Owl, Spot-billed Pelican, Demoiselle Crane and liver tissues of Demoiselle Crane, Indian Black Ibis, Painted Stork, and Indian Spotted Eagle were 190.11, 189.15, 188.08, 187.17, 188.19, 187.12, 188.14 and 187.50 $\mu\text{g/g}$ respectively. There was a significant variation in zinc concentrations among species included in the current study. The minimum levels of zinc were observed in muscle tissues of Emerald Dove (69.23 $\mu\text{g/g}$) and Eurasian Collared Dove (73.72 $\mu\text{g/g}$).

Concentrations of zinc recorded in Indian White-rumped Vulture in the present study were much higher than the previously recorded levels (Van wyk et al. 2001). The levels of zinc in Indian White-rumped Vulture included in the present study were 10 folds higher than the levels reported by Rajamani and Subramanian (2015) in the same species. However, the levels still appear to be non-toxic.

Levels of metals in different organs of birds

Among the three tissues (muscle, liver and kidney), kidney had the highest accumulation of total metal $230.62 \pm 2.367 \mu\text{g/g}$ in Black Kite and $117.31 \pm 5.42 \mu\text{g/g}$ in Rock Pigeon. Least concentration of all the metals was observed in muscle tissues. The levels of zinc and copper were found to be the maximum in kidney than that of liver and muscle, while the levels of nickel, lead, cadmium and chromium recorded were relatively similar in all the organs. Kidney had the highest level of most metals analyzed and remains a good indicator for metals in internal tissues of birds.

Levels of metals in gizzard of various species of birds

Gizzard contents from 24 species of birds (127 samples) were collected and analyzed for metal contamination. Among the species analyzed, the maximum level of total metal was recorded in Spot-billed Pelican ($250.10 \pm 31.97 \mu\text{g/g}$) followed by Blue-faced Malkoha ($220.17 \pm 10.87 \mu\text{g/g}$). The minimum level was observed in Emerald Dove ($95.50 \pm 9.42 \mu\text{g/g}$). The maximum concentration of cadmium was recorded in gizzard of Common Kingfisher ($4.05 \mu\text{g/g}$) followed by Spot-billed Pelican ($3.92 \pm 2.07 \mu\text{g/g}$) and Sarus Crane ($3.04 \pm 0.13 \mu\text{g/g}$), while the least was noted in Asian Koel.

The Common Kingfisher had the highest concentration of chromium ($4.63 \mu\text{g/g}$) followed by Spot-billed Pelican and Indian Black Ibis, whereas the lowest was observed in Indian Roller. The maximum concentration of Nickel was recorded in Blue-faced Malkoha ($7.78 \mu\text{g/g}$) followed by Spot-billed Pelican and Indian White-rumped Vulture. The highest concentration of copper was recorded in Spot-billed Pelican ($54.05 \mu\text{g/g}$) followed by Indian Black Ibis and Blue-faced Malkoha. There was a significant variation in zinc residues among the species. A higher concentration of zinc was observed in Spot-billed Pelican ($181.43 \mu\text{g/g}$) followed by Barn Owl ($179.98 \mu\text{g/g}$), whereas the least was noted in Emerald Dove.

The level of lead in gizzard contents ($0.41 - 0.79 \mu\text{g/g}$) in Rock Pigeon was reported from Bangladesh (Begum and Sherin 2013). The chromium level in gizzard contents of Common Kingfisher ($4.63 \mu\text{g/g}$) and Spot-billed Pelican ($4.17 \mu\text{g/g}$) in the present study are comparable with those of Godwin et al. (2016). Similarly, the levels of

nickel observed in gizzard contents of Indian White-rumped Vulture (value) and Indian Black Ibis were higher than those reported by Godwin et al. (2016) in Tree Swallow from Canada.

Variation in metal contamination among birds with different feeding habits

Among the 31 species studied, 30 were grouped into six feeding guilds such as granivore (04), herbivore (02), insectivore (05), piscivore (04), carnivore (10) and omnivore (06) excluding Rose-ringed Parakeet (frugivore) due to small sample size in the guild. The grouping was done based on their major food habits as mentioned in Ali and Ripley (1987). Organs of each individual were pooled to find out variation among feeding guilds.

Among the 30 species of birds analyzed, carnivorous birds had the highest concentration of total metal load followed by omnivorous and piscivorous birds. The levels of total metal load detected in insectivorous and herbivorous birds were 167.60 and 114.23 $\mu\text{g/g}$ respectively. The lowest level of total metal load was observed in granivorous birds. Similarly, the levels of zinc and copper were the maximum in carnivorous birds followed by omnivorous birds. The amount of cadmium, lead, chromium and nickel were detected more or less similar among all the feeding guilds.

Spatial variations in the levels of metal contamination among various species of birds

Since the sampling method adopted in this study was opportunistic, the possibilities of getting the same species of dead birds from all states were remote. Hence, the levels of metals were pooled irrespective of species and the comparisons were made among three states, namely Gujarat, Tamil Nadu and Kerala. Since, same species and adequate individuals could not be obtained from Assam it was not possible to include it for the comparison. Overall, Gujarat ($181.94 \pm 2.87 \mu\text{g/g}$) had the highest load of metal contamination, while Tamil Nadu ($176.75 \pm 3.98 \mu\text{g/g}$) the least. The variations in total metals in the birds among states were significant. Among the states, birds collected from Gujarat had the maximum concentration of zinc ($181.85 \pm 1.93 \mu\text{g/g}$), whereas the least was recorded in Tamil Nadu ($144.03 \pm 3.52 \mu\text{g/g}$). The maximum level of copper was recorded in Tamil Nadu ($27.13 \pm 1.00 \mu\text{g/g}$) followed by Kerala ($27.02 \pm 1.49 \mu\text{g/g}$), while Gujarat had the least concentration ($26.22 \pm 0.88 \mu\text{g/g}$). The highest level of nickel was

recorded in Tamil Nadu ($2.78 \pm 0.42 \mu\text{g/g}$) and the lowest levels were noted in Kerala ($1.19 \pm 0.16 \mu\text{g/g}$). The levels of chromium, lead and cadmium analyzed in birds did not show any variation among states.

The highest accumulation of metals in the birds in Gujarat may be due to highest level of contamination due to the presence of different types of industries. The remaining states, namely Tamil Nadu and Kerala relatively have lesser industrial houses and hence lower level of metal accumulation in birds is recorded.

When using bird species as indicators of certain environmental changes, the manager must choose species on the basis of the specific habitat factors which must be monitored. The ecology of the indicator species should be well known in order to separate the natural dynamics from the effects caused by human activity. Good indicators are specialized in their habitat needs, thus reacting rapidly to changes. Monitoring plans must be developed in such a way that they have long-term support. We argue that such support will be strongest when the overall monitoring scheme tells us about both ecological as well as human health and well-being.

Feathers: A better tool to understand metal contamination in birds

Feathers of 130 individuals comprising 19 species of birds were collected from three states in India, namely Gujarat, Tamil Nadu and Kerala. Different types of feathers, namely breast feather, primary feather, secondary feather and tail feather were collected from each individual. Metals such as cadmium, lead, chromium, nickel, copper and zinc were analyzed. When considering cadmium alone, the maximum concentration was found in breast feathers of Spot-billed Pelican ($7.09 \pm 2.01 \mu\text{g/g}$) followed by Indian White-rumped Vulture ($7.08 \pm 0.58 \mu\text{g/g}$), Sarus Crane ($6.08 \pm 0.25 \mu\text{g/g}$) and Shikra ($5.054 \pm 0.19 \mu\text{g/g}$). The maximum concentration of lead was recorded in primary feathers of Spot-billed Pelican ($3.26 \pm 0.13 \mu\text{g/g}$) followed by the breast feathers of House Crow ($2.99 \pm 0.63 \mu\text{g/g}$). The highest concentration of chromium was recorded in primary feathers of Spot-billed Pelican, followed by Red-wattled Lapwing ($6.48 \pm 0.73 \mu\text{g/g}$). Interestingly, Blue-faced Malkoha had the highest level of nickel in tail feathers ($17.62 \pm 15.04 \mu\text{g/g}$). The highest concentration of copper was recorded in primary feathers of Indian Black Ibis ($176.61 \pm 60.77 \mu\text{g/g}$), followed by breast feather of Spot-billed Pelican

(106.27 ± 28.27 µg/g). The lowest levels of Copper were recorded in secondary feathers of Asian Koel (22.22 µg/g) and Indian Pond Heron (22.96 µg/g). Highest concentration of zinc was found in breast feathers of Shikra (173.88 ± 23.61 µg/g) followed by Blue-faced Malkoha (156.96 ± 0.96 µg/g).

Levels of metals in various feathers of bird species

The levels of total metals varied significantly among the feathers. Among the four types of feathers analyzed, breast feathers had the highest concentration of total metals followed by primary feathers, while secondary feathers showed the least accumulation. Similarly, the levels of chromium and copper was found to be the maximum in primary feathers than that of breast feathers, while the levels of lead, cadmium, nickel and zinc were recorded to be the maximum in the breast feathers. Secondary feathers had lowest level of metal accumulation.

The range of metal concentrations in feathers is also greater in birds than in other tissues, which suggests that they provide a reliable indication of the different metal concentrations in the environment. Finally, other advantages of the use of feathers are that they can be obtained non-invasively and they are easier to process than other tissues.

Correlations among Internal Tissue (kidney) and Feathers

In general, the internal excretory organ, the kidney had the maximum and externally, breast feather had the maximum. When correlation was carried out to assess if the two were comparable, low correlation was observed. Overall, breast feather appeared to be the best indicator to check the metal load in body. While a negative correlation was observed between the concentration of zinc in kidney and feathers ($r = -0.294^*$; $P=0.034$), a positive correlation was observed for Cd; Pb and Cr ($r = 0.049$; 0.097 and 0.116 respectively). Ni ($- 0.101$) and Cu ($- 0.176$) had no correlation. Non-essential metals such as cadmium and lead were positively correlated.

Our data suggests that body feathers could be considered as appropriate indicators of metal contamination.

Metal contamination is perilous and once discharged in the environment, it is difficult to identify the course adopted by metals in ecosystems and even more difficult to

undo the ecological damage caused by it. Particularly with respect to toxicity in birds, it is recommended to monitor the metal contamination on a regular basis to avoid ecological hazards to local and regional avifauna.

In conclusion, the results show that feathers, when compared to kidney, are the most appropriate tool for analysis. However, further methodological studies must be carried out to establish which feathers are the most apt for bio-monitoring studies and when the samples should be collected.

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Publications

Elemental Contamination in Various Species of Birds from Select States in India

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Abstract - With the aim, to assess the magnitude of elemental contamination in birds in select states in India, samples of bird tissues were analyzed for copper (Cu), lead (Pb), cadmium (Cd) and chromium (Cr), with Atomic Absorption Spectrophotometer. Elements were detected in all the tissues and significant differences were noted in their concentrations among the species and tissues ($P>0.05$). Out of the 10 species analyzed, Bar-headed Goose and Red-wattled Lapwing accumulated the highest and Eurasian Collared-dove the lowest concentration of all the elements analyzed. Of all the tissues, kidney accumulated the maximum concentration of Pb (11.77 µg/g), Cd (17.06 µg/g) and Cr (16.08 µg/g). Apart from the organs studied, gut content also had notable levels of elements, which indicated the present contamination status of the environment. Nonetheless, the levels of most of the elements measured in the present study were considered non-toxic.

Keywords: Elements, Birds, Tissues, Bio-Indicators, Toxic, Contamination

I. INTRODUCTION

Increasing environmental contamination by elements has been a major concern in recent years in all industrialized nations in South Asia Bhuyan & Islam [7], including India. While elements are deposited on soils through geological activity, aerial deposition, flooding or anthropogenic sources, primary receptors such as microbes, plants and macro invertebrates pick them up from soil and other abiotic components, and transfer those elements to higher-level organisms [8]. In this process essential elements also can turn toxic to birds, humans and all other organisms, if exposure levels are above permissible limits [17].

Birds are one of the major victims of environmental contamination as they occupy a wide range of trophic levels in different food chains. Among wildlife, birds have been used as effective bio-indicators for a number of environmental contaminants including elements [22]. Thus, monitoring elements in birds from a delimited area periodically could be useful to assess not only the health of the species, but also the fluctuations in contamination levels in the environment where they live [30].

In India studies on elemental contamination in birds are limited. However, there is some information available on

sixteen species of aquatic birds in Koeladeo National Park, Bharatpur Muralidharan (24), House Sparrow in Jaipur Bakre and Sharma [2] and Indian White-backed Vulture Rajamani and Subramanian [27] in lead, cadmium, zinc and copper. Thus with the aim to generate more information on elemental contamination in birds in India, a study was launched, and information on 10 species of birds is presented in this article.

II. METHODS AND MATERIALS

A. Sample Collection

Samples of birds were collected on opportunistic basis from states, namely Tamil Nadu, Gujarat, Kerala and Assam between 2010 and 2014. Collected samples were transported to SACON Ecotoxicology laboratory over ice. Circumstantial evidences at the collection sites were gathered, and morphometric measurements were recorded at the time of conducting postmortem. Muscle, liver, kidney and gut content were collected and packed without any external contamination, stored at -20°C until chemical analysis.

B. Sample Digestion and Analysis

About 1 to 1.5 g (wet weight - ww) of samples were weighed with a reproducibility of 0.02 g in Mettler AE 240 balance and digested in Microwave Digestion System (Milestone, MLS 1200) using 10 mL Nitric Acid (69 % GR-Merck) for 10 min, 1 mL Perchloric Acid (70 % GR-Merck) for 5 min and 5 mL Hydrogen Peroxide (30 % GR-Merck) for 10 min at 250 W magnetron power settings. The digested solutions were filtered through Whatman No. 1 filter paper, made up to 25 mL with double distilled water and stored in polythene vials in refrigerator, and subsequently analyzed for metals, namely copper (Cu), lead (Pb), cadmium (Cd) and chromium (Cr) using a double beam Atomic Absorption Spectrophotometer (Perkin Elmer, AA 800). Standards for all the referred metals manufactured by SISCO Research Laboratories Pvt. Ltd., India were used for calibration. While the detection limit for copper was 2.0 ppb, it was 1.0, 3.0 and 2.0 ppb for cadmium, lead and chromium respectively. The

quantification limits of metals were three times the detection limit. Recovery rates for Cu, Pb, Cd, and Cr were 86, 88, 85, and 90 %, respectively. The results were not corrected for percent recovery, and expressed in $\mu\text{g/g} \pm \text{SE}$ (ww). In order to facilitate comparison with published information which are on dry weight basis, a conversion factor of three was used [10], wherever required.

C. Statistical Analysis

The data did not meet the assumption of normal distribution when checked using Shapiro-Wilk test ($p < 0.05$). Hence, Kruskal-Wallis, a non-parametric test was performed to

check the difference in elemental contamination among species and tissues. P values less than 0.05 were considered statistically significant. Statistical analyses were performed using SPSS software, Ver. 2.6.

III. RESULTS AND DISCUSSION

Totally 93 individuals comprising 10 species of birds were collected dead from four states in India (Table I). Concentrations of metals, namely Cu, Pb, Cd and Cr were compiled to look at the variations in contamination level among the species and tissues. Accumulation of elements differed among the species studied (Table II).

TABLE I SPECIES OF BIRDS COLLECTED DEAD FROM DIFFERENT STATES IN INDIA FOR THE STUDY BETWEEN 2010 AND 2014

S. No.	Common name	Scientific name	No. of individuals	Current population trend (IUCN, 2018)
1	Bar-headed Goose	<i>Anser indicus</i>	03	Decreasing
2.	Red-naped Ibis	<i>Pseudibis papillosa</i>	06	Decreasing
3	Rock Dove	<i>Columba livia</i>	24	Decreasing
4	Cattle Egret	<i>Bubulcus ibis</i>	21	Increasing
5	House Crow	<i>Corvus splendens</i>	19	Stable
6	Indian Peafowl	<i>Pavo cristatus</i>	06	Stable
7	Indian Pond - heron	<i>Ardeola grayii</i>	03	Unknown
8	Red - wattled Lapwing	<i>Vanellus indicus</i>	04	Unknown
9	Eurasian Collared - dove	<i>Streptopeli adacaecto</i>	03	Increasing
10	Rose-ringed Parakeet	<i>Psittacula krameri</i>	04	Increasing
Total			93	

On an average, Bar-headed Goose had the highest concentration of copper ($80.08 \pm 15.37 \mu\text{g/g ww}$) in gut content followed by liver ($73.58 \pm 5.47 \mu\text{g/g ww}$) while Eurasian Collared - dove accumulated the lowest ($2.95 \pm 0.57 \mu\text{g/g ww}$) in kidney. Significant variations were observed in the level of copper among the species of birds (Kruskal-Wallis test, $n = 159$; $H = 9.65$; $P < 0.05$) studied.

Waterfowl are affected by copper intoxication through ingestion of water contaminated by antifouling paints [14]. Other sources of copper include copper wire, pennies or any copper coated objects small enough to be ingested by birds [4]. But, birds appear to tolerate higher levels of copper than many mammals [28]. There have been reports on Mute Swans tolerating liver copper residues up to $1000 \mu\text{g/g ww}$ [13]. Jayakumar and Muralidharan [18] reported copper levels in a few aquatic birds in the range of $1.50 - 9.77 \mu\text{g/g ww}$ in kidney tissues which showed no sign of toxicity. The study conducted in Rock Dove from Bangladesh documented $3.44 \mu\text{g/g ww}$ in muscle and $8.72 \mu\text{g/g ww}$ in kidney of [5]. Hence, the concentrations of copper recorded in all the 10 species of birds studied were less than levels capable of inducing any toxic effects. Lead being a widespread environmental contaminant, has a history of causing mortality of millions of ducks across Europe and North America [31, 37]. However, in India there are no such reports. The maximum concentration of lead was found in

gut content of Red - wattled Lapwing ($55.85 \pm 13.09 \mu\text{g/g ww}$) followed by gut content of Bar-headed Goose ($31.92 \pm 6.70 \mu\text{g/g ww}$). The minimum concentration was in Cattle Egret ($0.29 \pm 0.03 \mu\text{g/g ww}$) in its muscle (Table II).

The levels were significantly different among the species ($P < 0.05$) studied. Levels of lead recorded in the present study were relatively lower than the levels recorded in muscle tissues of Canada Geese ($60.4 \mu\text{g/g ww}$) [34]. Kim and Koo [19] reported lead levels exceeding the threshold levels of approximately $29.4 \mu\text{g/g ww}$ in liver of Red-necked Stints and called it as indicative of chronic lead exposure. Concentrations more than 6 and $15 \mu\text{g/g}$ in kidney of Kestral was reported to be associated with impaired growth and survival respectively [29].

Lead affects birds both indirectly by reducing food supplies Eeva and Lehtikoinen[11] and directly by affecting reproduction, behavior, and hematopoietic and nervous systems [16, 35]. Although signs of lead poisoning in birds vary among species Beyer et al. [6], the same could not be ascertained as the birds included in the present study were collected only after they were dead. However, the levels of lead in Red- wattled Lapwing ($55.85 \pm 13.09 \mu\text{g/g ww}$) and Bar - headed Goose ($31.92 \pm 6.70 \mu\text{g/g ww}$) in the present study can be considered alarming when compared to earlier studies [19, 29].

Cadmium is characterized by its highest accumulation factor in living organisms [20]. At lower concentrations, Cd may lead to development of behavioral and sub lethal effects as compared to Pb and Hg [16]. Cadmium concentration was the highest in Indian Pond Heron ($27.94 \pm 10.10 \mu\text{g/g ww}$) followed by Indian Peafowl ($22.03 \pm 21.53 \mu\text{g/g ww}$) in kidney. Out of the ten species of birds studied, the Eurasian Collared Dove had the lowest level of Cd in muscle ($0.34 \pm 0.24 \mu\text{g/g ww}$).

Muralidharan [24] in his study on metal contamination in sixteen species of aquatic birds had recorded $2.12 \mu\text{g/g}$ of Cd in the kidney of Indian Pond Heron. Kidney damages have been reported in wild colonies of pelagic sea birds having Cd level of $60\text{--}480 \mu\text{g/g}$ in their kidneys [38]. Cadmium in breast muscles of mallards in Europe was $0.082 \mu\text{g/g ww}$ [32]. Cadmium levels in liver of Canada Geese, although were higher than other tissues at over $0.20 \mu\text{g/g}$ in Mill Creek and over $0.30 \mu\text{g/g}$ at Skeetkill Marsh were not anywhere close to the avian threshold ($41.02 \mu\text{g/g ww}$) for Cd poisoning [15]. Thus, the present study shows that the concentrations of Cd among the 10 species of birds were less than levels capable of causing any toxicity.

Chromium being a trace element is an essential micro-nutrient in animal diet [26]. Interestingly both maximum

and minimum levels of chromium were in Rock Dove. While kidney had the maximum ($31.92 \pm 16.64 \mu\text{g/g ww}$), muscle had the minimum levels ($3.49 \pm 1.08 \mu\text{g/g ww}$) (table 2). While Begum & Sehrin [5] reported 1.85 and $0.91 \mu\text{g/g ww}$ of Cr in kidney and muscle tissues of Blue Rock Pigeon respectively in Bangladesh, Torreset *al.*, [33] reported $3.37 \mu\text{g/g ww}$ of Cr in muscle tissues of Blue Rock Pigeon in Spain, and these levels were similar to levels recorded in the present study. Sublethal effects of Cr in birds include growth retardation, anemia and testicular damage, and it is considered to be life threatening to the organism [36].

Since Blue Rock Pigeons are also consumed by man we suspect these conditions to adversely affect and create health risk in those end users [1, 5, 23]. The study recommends monitoring the levels of elemental contaminants including Cr in tissues of Blue Rock Pigeon. United States Fish and Wildlife Service [36] suggested that Cr levels in kidney tissues in excess of $40 \mu\text{g/g ww}$ should be considered as an evidence of Cr contamination. When compared to published information, levels recorded in the present study were found to be higher in all the species. But apparently these levels did not show any toxic effects.

TABLE II LEVELS OF ELEMENTS ($\mu\text{G/G MEAN} \pm \text{SE}$) IN DIFFERENT TISSUE OF BIRDS IN SELECT STATES IN INDIA

Bird species	Organ	Copper	Lead	Cadmium	Chromium
Bar-headed Goose (n=03)	Muscle	41.61 ± 11.72	13.23 ± 3.95	8.75 ± 3.24	15.39 ± 2.86
	Liver	73.58 ± 5.47	5.74 ± 2.50	5.97 ± 0.87	6.44 ± 0.77
	Kidney	70.57 ± 15.74	7.30 ± 1.10	13.22 ± 1.63	8.15 ± 1.35
	Gut content	80.08 ± 15.37	31.92 ± 6.70	4.22 ± 0.76	9.45 ± 0.92
Red-napped Ibis (n=06)	Muscle	27.31 ± 4.50	9.71 ± 3.59	11.55 ± 2.97	5.91 ± 1.62
	Liver	25.90 ± 6.81	16.98 ± 3.89	21.55 ± 3.30	7.29 ± 0.63
	Kidney	6.28 ± 1.93	21.53 ± 4.13	25.49 ± 2.80	8.35 ± 0.60
	Gut content	25.69 ± 5.43	17.11 ± 3.56	15.29 ± 4.31	10.59 ± 2.35
Rock Dove (n=24)	Muscle	42.54 ± 4.48	3.48 ± 0.58	1.89 ± 1.06	3.49 ± 1.08
	Liver	38.75 ± 3.80	6.45 ± 0.82	8.59 ± 1.53	8.19 ± 3.60
	Kidney	17.72 ± 02.88	14.48 ± 8.56	13.19 ± 3.09	31.92 ± 16.64
	Gut content	25.91 ± 4.50	13.73 ± 3.29	8.32 ± 1.84	7.49 ± 1.08
Cattle Egret (n=21)	Muscle	18.50 ± 2.60	0.29 ± 0.03	2.03 ± 21.54	10.83 ± 2.96
	Liver	27.93 ± 4.30	4.92 ± 1.43	10.90 ± 3.13	15.85 ± 4.65
	Kidney	18.56 ± 3.21	12.87 ± 0.78	21.93 ± 2.71	18.10 ± 6.71
	Gut content	27.83 ± 7.80	29.92 ± 6.70	4.22 ± 0.76	16.10 ± 3.57
House Crow (n=19)	Muscle	42.28 ± 11.52	4.33 ± 0.45	5.544 ± 3.56	10.83 ± 2.96
	Liver	45.03 ± 10.64	8.53 ± 1.12	8.78 ± 1.87	15.85 ± 4.65
	Kidney	38.96 ± 23.96	9.20 ± 1.26	10.52 ± 5.25	18.10 ± 6.71
	Gut content	13.94 ± 2.09	5.57 ± 0.93	2.59 ± 0.64	4.60 ± 2.05
Indian Peafowl (n=06)	Muscle	10.13 ± 1.53	3.05 ± 0.51	4.12 ± 1.33	10.13 ± 1.53
	Liver	54.34 ± 24.29	8.43 ± 4.05	8.16 ± 3.53	9.56 ± 0.98
	Kidney	9.55 ± 6.92	12.09 ± 0.03	22.03 ± 21.53	10.48 ± 3.09
	Gut content	15.48 ± 10.58	1.75 ± 1.39	1.51 ± 0.60	15.51 ± 2.22

Indian Pond-heron (n=03)	Muscle	13.23 ± 3.95	5.04 ± 2.34	4.12 ± 1.33	10.13 ± 1.53
	Liver	49.31 ± 20.08	8.45 ± 6.6	16.26 ± 7.18	14.51 ± 3.64
	Kidney	46.96 ± 2.22	10.29 ± 8.66	27.94 ± 10.10	27.25 ± 9.18
	Gut content	28.50 ± 6.03	16.98 ± 1.57	6.55 ± 3.22	16.69 ± 4.64
Red-wattled Lapwing (n=04)	Muscle	13.33 ± 5.46	6.66 ± 3.59	6.53 ± 3.79	5.78 ± 1.74
	Liver	25.96 ± 11.67	11.29 ± 4.24	12.06 ± 4.94	3.69 ± 1.29
	Kidney	19.00 ± 5.19	16.73 ± 5.56	21.30 ± 7.29	15.15 ± 3.84
	Gut content	6.65 ± 6.07	55.85 ± 13.09	0.93 ± 0.24	7.74 ± 0.53
Eurasian Collared-dove (n=03)	Muscle	2.95 ± 0.57	13.07 ± 1.86	0.34 ± 0.24	2.85 ± 0.83
	Liver	6.88 ± 2.49	9.78 ± 3.70	7.15 ± 6.55	5.29 ± 1.54
	Kidney	15.27 ± 1.03	12.87 ± 0.78	21.93 ± 2.71	13.20 ± 4.21
	Gut content	12.29 ± 1.05	29.76 ± 8.34	1.86 ± 0.46	13.72 ± 2.65
Rose-ringed Parakeet (n=04)	Muscle	11.29 ± 2.84	13.07 ± 1.86	1.34 ± 0.24	7.85 ± 0.83
	Liver	19.68 ± 2.74	27.74 ± 4.64	5.93 ± 1.54	17.69 ± 4.82
	Kidney	14.42 ± 1.67	23.52 ± 3.69	2.71 ± 0.86	13.08 ± 2.07
	Gut content	20.16 ± 5.16	7.50 ± 0.29	10.68 ± 1.53	11.53 ± 0.30

The variation in levels of elemental contaminants among tissues irrespective of species was also compiled to understand the accumulation potential of the organs. Organs play vital role in accumulating metals. Particularly, liver and kidney are involved in detoxification mechanisms [3]. Even though number of individuals in each species was less because of opportunistic sampling, we tried to find out the tissue which accumulated the maximum concentrations of elements (Fig.1). One way ANOVA Post hoc test was performed to compare the levels of elemental contamination in the tissues.

Of all the tissues, muscle accumulated the minimum concentration of all the elements (Cu- 2.17 ± 1.58 µg/g ww; Pb-6.11 ± 2.36 µg/g ww; Cd-3.91 ± 2.47 µg/g ww and Cr-1.11 ± 0.46 µg/g ww). Rajamani and Subramanian [27] reported of lead 1.72 µg/g in muscle in Indian White-backed Vulture to be not toxic.

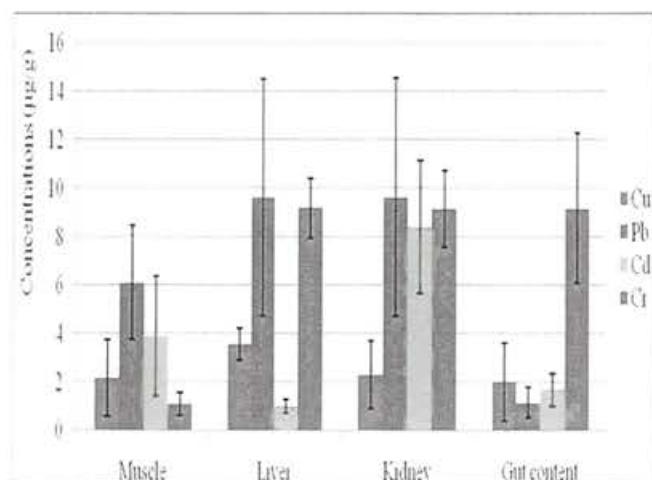


Fig. 1 Variation in elemental contamination among organs in birds collected from select states in India (µg/g Mean ± SE)

Liver accumulated only copper in higher concentrations (3.56 ± 0.64 µg/g ww among other tissues. Kidney accumulated higher concentrations of Pb, Cd and Cr (9.62 ± 4.89 µg/g ww; 1.01 ± 0.27 µg/g ww; 9.19 ± 1.24 µg/g ww) respectively. Similar levels Cd (1.10 µg/g ww) in kidney were reported in Greater White-fronted Goose (*Anseralifrons*) [39]. Apart from the organs studied, gut content also had notable levels of heavy metals, which indicates the present status of elemental contamination of the environment.

IV. CONCLUSION

Further, in order to evaluate any instance of element poisoning, normal levels of them for the species concerned must be and the known abnormal levels, which tend to produce either harm or no apparent harmful effects, must be recognized which is a conspicuous need in Indian context. Aquatic birds are particularly vulnerable to contaminants, including elements [25]. Kidney and liver are the major organs which respond to the insult of elemental contamination owing to their physiological role [18, 21]. Similarly the data generated in this study also show the liver and kidney tissues of birds to be good indicators of elemental contamination in the environment.

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