SPECIATION AND BIOAVAILIBILITY OF METALS IN MUDFLAT AND MANGROVE REGIONS OF MANDOVI ESTUARY, WEST COAST OF INDIA

A Thesis submitted to Goa University for the award of the Degree of

DOCTOR OF PHILOSOPHY

IN

(MARINE SCIENCE)

BY

(RATNAPRABHA R. SIRASWAR, M.Sc)

Goa University,

Taleigao Goa

(APRIL, 2014)

SPECIATION AND BIOAVAILIBILITY OF METALS IN MUDFLAT AND MANGROVE REGIONS OF MANDOVI ESTUARY, WEST COAST OF INDIA

A Thesis submitted to Goa University for the award of the Degree of

DOCTOR OF PHILOSOPHY

IN

(MARINE SCIENCE)

BY

(RATNAPRABHA R. SIRASWAR, M.Sc)

Research Guide

(PROF. G. N. NAYAK)

Goa University,

Taleigao Goa

(APRIL, 2014)

DEDICATED TO MY PARENTS

STATEMENT

As required under the university ordinance OB.9.9 (iv), I state that the present thesis entitled **“SPECIATION AND BIOAVAILIBILITY OF METALS IN MUDFLAT AND MANGROVE REGIONS OF MANDOVI ESTUARY, WEST COAST OF INDIA”,** is my original contribution and the same has not been submitted for any other degree, diploma, associateship, fellowship or similar thesis in any universities or institutions on any previous occasion. To the best of my knowledge, the present study is the first comprehensive work of its kind from the area mentioned.

The literature related to the problem investigated has been cited. Due acknowledgements have been made wherever facilities and suggestions have been availed of.

Place: Goa University

Date: 07.04.2014

Ms. Ratnaprabha R. Siraswar

CERTIFICATE

This is to certify that the thesis entitled, **“SPECIATION AND BIOAVAILIBILITY OF METALS IN MUDFLAT AND MANGROVE REGIONS OF MANDOVI ESTUARY, WEST COAST OF INDIA”,** submitted by Ms. Ratnaprabha R. Siraswar for the award of the Degree of Doctor of Philosophy in Marine Science is based on her original studies carried out by her under my supervision. The thesis or any part thereof has not been previously submitted for any other degree, diploma, associateship, fellowship or similar titles in any universities or institutions. This thesis represents independent work carried out by the student.

Place: Goa University

Date: 07.04.2014

Prof. G. N. Nayak
Research Guide
Head, Department of Marine Sciences,
Goa University, Goa

Acknowledgements

It’s really an impossible task to acknowledge each and every one who has helped in preparing this thesis. Foremost, a big “thank you” to almighty God, the driving force who gave me the strength, blessings and has endowed me with an excellent research guide.

Prof. G. N. Nayak patiently guided me throughout. Words are inadequate to express my sincere gratitude to my research guide. Special thanks to him for suggesting this topic and giving me the opportunity to research on it. I thank him for his scientific advice and knowledge and many insightful discussions and suggestions. His constant motivation, encouragement, critical suggestions has been immensely helpful.

I wish to place on record my thanks to Dr. S. R. Shetye, Vice Chancellor, Goa University and Deans of the faculty for their support and encouragement.

I would also like to thank Dr. Savita Kerkar (Co-guide) in extending help in formulating some experiments. I would like to express my gratitude to the FRC comprising of Dr. B.N. Nath, Dr. A. Muzamdar, Dr. Savita Kerkar and Prof. G. N. Nayak, for their suggestions and encouragements. I would also like to thank Dr. A. L. Paropkari for giving me suggestions.

I wish to express my gratitude to the Centre of Excellence, Ministry of Earth Sciences for granting me Junior Research fellowship under in house project sactioned to Prof. G. N. Nayak during the three years of my Ph.D. I also pledge to thank all the people who were actively involved in the field surveys.

My sincere gratitude to Shri. Rashik Ravindra, Director, National Centre for Antarctic and Oceanic Research (NCAOR), Goa for giving me an opportunity to participate in the Third Southern Ocean Expedition during my research period. Special thanks go to Dr. M. Sudhakar, Chief Scientist for his encouragement and support.

I’m thankful to faculty members of Department of Marine Sciences, Goa University viz Prof. H. B. Menon, Dr. S. Upadhyay, Dr. M. V. Matta, Dr. C. U. Rivonkar, Dr. Aftab Can, for their encouragement. I’m also thankful to the non-teaching staffs of Department of Marine Sciences viz Mr. Rosario, Mr. Yashwant, Mr. Ashok, Mr. Narayan, Mr. Samrat, Mr. Shatrugan, Mr. Atchut, Mrs Sanjana, Mrs Matilda, Mrs Mangal for their kind help. Also, my thanks go to Mr. Serrao, Mr. Martin, and Mr. Ulhas for rendering help during the period.

I owe a lot to my research colleagues, Ms. Lina, Mrs. Deepti, Ms. Maria, Mrs. Cheryl, Ms. Purnima, Ms. Bhavya, Ms. Soniya, Ms. Samida, Mrs. Shilpa, Ms. Sweety, Ms. Conchita, Mrs. Tomina, Ms. Michelle, Ms. Amruta, Ms. Geeta, Ms. Teja, Ms. Anumol, Ms. Aruna, Ms. Sahina, Ms. Veliosa, Ms. Lilian, Mrs. Nutan, Mr. Maheshwar, Mr. Mahabaleshwar, Mr. Shrivardan, Mr. Dinesh, Mr. Rahul, Mr. Renosh, Mr. Santosh, Mr. Tomchou, Mr. Anant, Mr. Sunil, Mr. Vinay, Mr. Kuldeep, Mr. Milind, Mr. Pramoda, Mr. Shuvankar and Mr. Kirti. I thank them all for their support.

Every word falls short of my gratitude to my parents and my family for their unwavering support, love, help and everything. I honestly thank each and every one of my family members for bearing with me. I thank my brother Vishwanath R. Siraswar for constantly helping me in developing positive motivation. I am glad towards my guru to provide me such wonderful “Appa” and “Amma” who was constantly there in any circumstances especially my “Appa”.

Ms. Ratnaprabha R. Siraswar

Contents

List of Tables

List of Figures

Preface

Title No's	Contents	Page Numbers
	Chapter 1 Introduction	***1-20***
1.1	Introduction	1
1.2	Metal speciation	3
1.3	Factors affecting metal speciation	5
1.4	Metals and microbes	6
1.5	Literature review	7
1.6	Objectives	18
1.7	Study area	18
	Chapter 2 Materials and Methods	***21 - 34***
2.1	Introduction	21
2.2	Field methods	21
2.2.1	Collection of water samples	21
2.2.2	Collection of sediment samples	23
2.3	Laboratory analysis	25
2.3.1	Water analysis	25
a)	Salinity	25

b)	Total suspended matter	25
c)	Particulate metals	26
2.3.2	Sediment analysis	26
a)	Sediment component (Sand, Silt, Clay)	26
b)	Estimation of organic carbon	27
c)	Metal analysis	28
d)	Speciation of metals	29
e)	Microbiological analysis	31
2.4	Data processing	32
a)	Statistical analysis	32
b)	Pollution indices	32
	Chapter 3 Total Suspended Matter	***35-58***
3.1	Introduction	35
3.2	Salinity	35
3.3	Total Suspended Matter (TSM)	36
3.4	Metals	40
3.5	TSM *Vs* Metals	46
3.6	Inter-seasonal variation	49
3.6.a	Isocon diagram	49
3.6.b	Cluster analysis	51
3.6.c	Pearsons correlation	54

Chapter 4 Surface Sediments (Seasonal Variation) *59-103*

4.1.	Sediment components	59
4.1.A.	Mudflats	59
4.1.B	Mangroves	66
4.2.	Pollution level based on bulk sediment chemistry	75
4.2A.	Mudflats	75
4.2A.a.	Geoaccumulation Index (Igeo)	76
4.2A.b.	Pollution Load Index (PLI)	77
4.2B.	Mangroves	78
4.2B.a.	Geoaccumulation Index (Igeo)	79
4.2B.b.	Pollution Load Index (PLI)	80
4.3.	Speciation- Mudflats	82
4.4.	Pollution level based on bioavailability of metals - Mudflats	94
4.4.a.	SQUIRT	94
4.4.b.	Risk Assessment Code (RAC)	96
4.5.	Variation in bioavailability between seasons - Mudflats	99

Chapter 5 Sub-surface sediments (Variation with depth) *104-163*

5.1.	Sediment components	104
5.1A.	Mudflats	104
5.1B.	Mangroves	112
5.2.	Speciation of metals	121
5.2A.	Mudflats	121
5.2B.	Mangroves	131

5.3.	Mudflats v/s Mangroves Environments	138
5.3.a.	Mudflats vs Mangroves (Sediment components)	140
5.3.b	Mudflats v/s Mangroves (Metal species)	142
5.4.	Metal Enrichment	144
5.4.a.	Geoaccumulation Index	145
5.4.b.	Pollution Load Index (PLI)	146
5.4.c.	SQUIRT	148
5.4.d.	Risk assessment code (RAC)	150
5.5.	Metal tolerant microbes in mudflat sediments	153
5.5.a.	Lower estuary (Betim)	153
5.5.b.	Lower middle estuary (Karyabhat)	154
5.5.c.	Upper middle estuary (Ribander)	154
5.6.	Sediment parameters and microbes	156
5.7.	Metal species and metal tolerating bacterial population	158
	Chapter 6 Summary and Conclusion	***164-169***
	References	***170-196***

LIST OF TABLES

Table No's	**Contents**	**Page Numbers**

Table 1.1.	Literature survey on related topics of research	7
Table 2.1.	Details of sediment core samples collected	25
Table 2.2.	Time schedule used for pipette analysis	27
Table 2.3.	Geoaccumulation indices (after Müller, 1979)	34
Table 3.1a.	Pearsons correlation in surface waters (Monsoon)	56
Table 3.1b.	Pearsons correlation in bottom waters (Monsoon)	56
Table 3.2a.	Pearsons correlation in surface waters (Post-monsoon)	56
Table 3.2b.	Pearsons correlation in bottom waters (Post-monsoon)	57
Table 3.3a.	Pearsons correlation in surface waters (Pre-monsoon)	57
Table 3.3b.	Pearsons correlation in bottom waters (Pre-monsoon)	57
Table 4A.1.	Distribution of sediment components within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	60
Table 4A.2.	Distribution of Fe, Mn and Al within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	61
Table 4A.3.	Distribution of Total heterotrophic counts within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	61
Table 4B.1.	Distribution of sediment components within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	68
Table 4B.2.	Distribution of Fe, Mn and Al within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in	69

	monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	
Table 4B.3.	Total heterotrophic bacterial distribution within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	69
Table 4A.4.	Geoaccumulation Index (Igeo) values for lower, lower middle and upper middle regions of mudflat sediments of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	76
Table 4A.5.	Pollution Load Index (PLI) values for lower, lower middle and upper middle regions of mudflat sediments of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	77
Table 4B.4.	Geoaccumulation Index (Igeo) values in lower, lower middle, upper middle and upper estuarine regions of mangrove sediments of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons	79
Table 4B.5.	Pollution Load Index (PLI) values in lower, lower middle, upper middle and upper estuarine regions of mangrove sediments of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons	80
Table 4A.6.	Screening quick reference table for metals in marine sediments (Buchman 1999)	94
Table 4A.7.	Sediment guidelines and terms used in SQUIRT	94
Table 4A.8a.	Average concentration of total metals and bioavailable fractions in lower estuary in the three different seasons	95
Table 4A.8b.	Average concentration of total metals and bioavailable fractions in lower middle estuary in the three different seasons	95
Table 4A.8c.	Average concentration of total metals and bioavailable fractions in upper middle estuary in the three different seasons	96
Table 4A.9.	Criteria of Risk Assessment code (Perin et al., 1985)	97
Table 4A.10a.	Sum of exchangeable and carbonate fractions in percentage - Lower estuary	98

Table 4A.10b.	Sum of exchangeable and carbonate fractions in percentage - Lower middle estuary	98
Table 4A.10c	Sum of exchangeable and carbonate fractions in percentage - Upper middle estuary	98
Table 5A.1.	Distribution of sediment components within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)	106
Table 5A.2.	Distribution of metals (Fe, Mn and Al) within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)	107
Table 5A.3.	Total heterotrophic bacterial distribution within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)	108
Table 5B.1.	Distribution of sediment components within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)	114
Table 5B.2.	Distribution of metals (Fe, Mn and Al) within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)	115
Table 5B.3.	Total heterotrophic bacterial distribution within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)	116
Table 5A.4.	Geoaccumulation Index (Igeo) values at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of Mandovi estuary	145
Table 5B.4.	Geoaccumulation Index (Igeo) values at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary	146
Table 5A.5.	Pollution Load Index (PLI) values at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of	147

	Mandovi estuary	
Table 5A.6.	Bulk metal concentration at different sediment depths (cm) of lower, lower middle and upper middle estuarine regions in mudflat sediments of Mandovi estuary	147
Table 5B.5.	Pollution Load Index (PLI) values at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary	148
Table 5B.6.	Bulk metal concentration at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary	148
Table 5.7	Screening quick reference table for metals in marine sediments (Buchman 1999)	148
Table 5.8.	Sediment guidelines and terms used in SQUIRT	149
Table 5A.9.	Average concentration of metals in bioavailable fractions at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of Mandovi estuary	149
Table 5B.9.	Average concentration of metals in bioavailable fractions at different sediment depths (cm) of lower, lower middle and upper middle regions in mangrove sediments of Mandovi estuary	150
Table 5.10.	Criteria of Risk Assessment code (Perin et al., 1985)	150
Table 5A.11.	Sum of exchangeable and carbonate fractions in % of the total in lower, lower middle and upper middle regions in mudflat sediments of the estuary	151
Table 5A.11 a.	Sum of exchangeable and carbonate fractions in % of the total metal	151
Table 5A.11 b.	Sum of exchangeable and carbonate fractions in % of the total metal	151
Table 5A.11 c.	Sum of exchangeable and carbonate fractions in % of the total metal	152
Table 5B.11.	Sum of exchangeable and carbonate fractions in % of the total in lower, lower middle and upper middle regions in mangrove sediments of the estuary	152
Table 5B.11a.	Sum of exchangeable and carbonate fractions in % of the total metal	152
Table 5B.11 b.	Sum of exchangeable and carbonate fractions in	152

	% of the total metal	
Table 5B.11 c.	Sum of exchangeable and carbonate fractions in % of the total metal	153
Table 5A.12.	Relation between bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment fractions of lower estuary	158
Table 5A.13.	Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment fractions in lower estuary	159
Table 5A.14.	Bioavailable Co and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Co in different sediment fractions in lower estuary	159
Table 5A.15.	Bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment fractions of lower middle estuary	161
Table 5A.16.	Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment fractions of lower middle estuary	161
Table 5A.17.	Bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment fractions of upper middle estuary	162
Table 5A.18.	Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment fractions of upper middle estuary	162

LIST OF FIGURES

Figure No's	**Contents**	**Page Numbers**
Fig 1.1.	Location of study area – Mandovi Estuary	19
Fig 2.1.	Schematic diagram showing sampling and laboratory analysis	22
Fig 2.2a.	Map showing the water sampling locations	24

Fig.2.2b.	Map showing the sediment sampling locations	24
Fig 2.3.	Schematic diagram representating sequential extraction procedure	30
Fig 3.1.	Distribution of salinity and TSM profiles along the estuary from mouth (station 1) towards the head (station 5) in Monsoon (a, b), Post monsoon (c, d) and Pre monsoon (e, f)	38
Fig 3.2.	Plots of salinity and TSM in surface and bottom waters in monsoon (a and b); post monsoon (c and d) and pre monsoon (e and f) in Mandovi estuary	39
Fig 3.3.	Distribution of particulate metals (Fe, Mn, Zn, Ni and Co) along the estuary from mouth (Station 1) towards the head (Station 5) in surface and bottom waters of Mandovi estuary during different seasons	46
Fig 3.4.	Isocon diagram (Grant, 1986) between seasons where, individual points represent average value of salinity, TSM and particulate metals (Fe, Mn, Zn, Ni, Co) in surface and bottom waters	51
Fig 3.5.	Dendograms of average values of variables - salinity, TSM, particulate metals (Fe, Mn, Zn, Ni and Co) in surface and bottom waters	54
Fig 4A.1.	Trace metal (Cr, Cu, Co, Zn, Ni and Pb) distribution within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons	62
Fig 4B.1.	Trace metal (Cr, Cu, Co, Zn, Ni and Pb) distribution within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons	70
Fig 4A.2.	Distribution of Mn, Co, Pb and Zn in mudflat sediments of lower estuary in different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons	88
Fig 4A.3.	Distribution of Mn, Co, Pb and Zn in mudflat sediments of lower middle estuarine region in	89

	different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons	
Fig 4A.4.	Distribution of Mn, Co, Pb and Zn in mudflat sediments of upper estuarine region in different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons	90
Fig 4A.5.	Isocon plot for samples collected from mudflats of lower estuary in different seasons	100
Fig 4A.6.	Isocon plot for samples collected from mudflats of lower middle estuary in different seasons	101
Fig 4A.7.	Isocon plot for samples collected from mudflats of upper middle estuary in different seasons	102
Fig. 5A.1.	Distribution of metals (Cr, Cu, Co, Zn, Ni and Pb) within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)	109
Fig. 5B.1.	Distribution of metals (Cr, Cu, Co, Zn, Ni and Pb) within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)	118
Fig. 5A.2.	Geochemical fractionation of Fe, Mn, Co, Zn and Pb with depth in lower estuary (LE), Lower middle estuary (LME), Upper middle estuary (UME) of mudflat sediments in Mandovi estuary	125
Fig 5A.3.	Isocon plot for cores collected from mudflats of lower, lower middle and upper middle estuary	131
Fig. 5B.2.	Geochemical fractionation profile of Fe, Mn, Co, Zn and Pb with depth in lower estuary (LE), Lower middle estuary (LME), Upper middle estuary (UME) of mangrove sediment in Mandovi estuary	135
Fig 5B.3.	Isocon plot for cores collected from mangroves of lower, lower middle and upper middle estuary	137
Fig. 5.4.	Isocon diagram (Grant, 1986) individual points represent average value of sediment component and element of the core, mudflats vs mangroves	141
Fig 5.5a.	Isocon plot for cores collected from mudflats and	142

	mangroves of lower estuary	
Fig 5.5b.	Isocon plot for cores collected from mudflats and mangroves of lower middle estuary	143
Fig 5.5c.	Isocon plot for cores collected from mudflats and mangroves of upper middle estuary	144
Fig. 5A.6.	Total heterotropic counts in Zobell marine agar (ZMA) and Minimal media (M.M) in a) Lower estuary b) Lower middle estuary c) Upper middle estuary	155
Fig. 5A.7.	Metal tolerant bacteria in media containing metals such as Fe, Mn, Co, Zn and Pb in different concentrations (25, 50 and 100 ppm) from sediments of a) Lower estuary b) Lower middle estuary c) Upper middle estuary	156

PREFACE

Estuaries are unique environments of the coastal zone where freshwater from land meets and mixes with saline oceanic waters. The estuarine processes are governed by several forcing factors such as river inflow, tides, waves, wind making the system dynamic, thus temperature, salinity, turbidity and other parameters fluctuate on a tidal, daily, fortnightly and seasonal basis. Estuaries cover various sub-environments viz. shallow marine waters, freshwater, marshes, swamps, sandy islands, mud and sand flats, oyster's reefs, mangroves, tidal pools and sea *grasses. Intertidal areas comprising of mud/sand flats and mangroves are constantly influenced by varying energy conditions. Mudflats and sheltered intertidal sand flats reflect low energy conditions which are characterised by particles of small to medium diameter, high water content, low porosity, high organic content that provide high reducing conditions. Mudflats are also known for high carbon to nitrogen ratio, high microbial population and high sediment stability. On the other hand, mangroves are one of the world's most productive ecosystems and form an important part of the coastal and estuarine environment. They produce organic carbon well in excess of the ecosystem requirement and contribute significantly to global carbon cycle. They have enormous ecological value. They protect and stabilize coast lines, enrich coastal waters, yield commercial products and support coastal fisheries.*

Inorganic and organic components are discharged into the estuarine waters by rivers and sea through natural inputs and several anthropogenic activities. In an aquatic environment, many pollutants, especially metals, are easily adsorbed on the suspended particles and the flocculation process allows settling of suspended matter and metals to become part of sediments. Therefore, estuarine environments such as mudflats and mangroves, acts as a sink for particle associated contaminants. Fine sedimentary deposits or mud with high organic matter content are the most characteristic feature of these sub environments which play an important role in mobilisation of trace metals.

Within the sediments, metals are present in different forms, and generally exhibit different physical and chemical behaviour in terms of chemical interactions, mobility, biological availability and potential toxicity. Thus, information on the total concentration of metals alone is not sufficient to assess the environmental impact of sediments. Further, marine microorganisms that inhibit estuaries can tolerate higher metal concentrations than those living in open Ocean. This is because there are greater concentration of inorganic and organic metal complexing ligands e.g. Fe and Mn hydroxides, humic and fulvic acids or because of historical exposure has lead to the evolution of tolerance mechanisms. In the present study an attempt has been made to understand bioavailability of metals and metal tolerance of organisms in estuarine environment.

The objectives of the present study are

1) *To study the abundance and distribution of sediment grain size and selected metals in sediments from Mudflats and Mangroves.*
2) *To understand the role of various parameters in the distribution and concentration of metals and their species in Mudflats and Mangroves.*
3) *To investigate metal tolerance of associated bacterial isolates.*

The thesis is divided into five chapters

Chapter 1 details brief introduction on the estuarine ecosystem, associated processes and possible sources of sediments/metals. A general background on the literature survey carried out in the study area and also in the country and around the world, which triggers most of the work currently undertaken on enrichment of metals in different forms and their effect on associated biota is presented here. The chapter also includes the objectives of the present work and description of the study area along with different sampling sites.

The second chapter provides the methodology, covering in detail the sampling protocols and various analytical techniques used in the investigation. The sampling locations and the visual characteristic of the cores are described. The principles of the methods used, followed by the procedure employed are also explained. Methods followed in the present study include analysis of salinity; total suspended matter and particulate metal analysis of water samples and grain size; organic carbon; bulk chemistry and speciation of metals for sediment samples.

Statistical analysis comprising of Pearson's correlation matrix was used to understand the various geochemical sources and processes. In order to determine pollution level of the region, Geoaccumulation Index (Igeo) and Pollution Load Index (PLI) were computed. Further, to understand the risk of the studied metals to sediment dwelling organisms, the data set of both total metals in sediment and metals in bioavailable fractions were compared with Sediment Quality Values (SQV) using Screening Quick Reference Table (SQUIRT). Also, using metal speciation results, Risk Assessment Code (RAC) has been computed to assess the environmental risks to benthic organisms caused by the contaminated sediments.

Chapter 3 describes the total suspended matter (TSM) in both surface and bottom samples along with salinity from Mandovi estuary. Further, the particulate metal concentration for Fe, Mn, Zn, Ni and Co analysed using Atomic Absorption Spectrophotometer (AAS). Statistical analysis both cluster and Pearson's analysis were employed to understand the sources and matrix on to which metals are adsorbed and deposited.

Chapter 4 deals with the results and discussion on sediment characteristics, organic matter, bulk chemistry and total heterotrophic counts in mudflat and mangrove environments along Mandovi estuary in different seasons. Different pollution indices were used to understand level of pollution. Further, speciation of sediments was investigated to understand seasonal distribution of metal species (Fe, Mn, Zn, Co and Pb) within different regions of the estuary.

In chapter 5 distributions of sediment characteristics, organic carbon, metals and heterotrophic counts within the sediment cores collected at different estuarine regions of two sub-environments are discussed. Further, the speciation of metals (Fe, Mn, Co, Zn and Pb) in mangroves and mudflats are presented in this chapter. Through the laboratory experiments metal tolerance of bacteria in mudflat sediments are studied for Fe, Mn, Co, Zn and Pb. The results obtained are correlated with bioavailability of metals. Based on values of RAC, Mn, Co and Pb were selected for the study of bioavailability and metal tolerance as they are falling in high risk to very high risk category.

The sixth chapter, summary and conclusion, presents salient features of the present investigation.

Complete list of references cited in the text, tables and figures in alphabetical order are presented at the end.

CHAPTER ONE

INTRODUCTION

1.1. Introduction

Estuaries are unique environments of the coastal zone where freshwater from land meets and mixes with saline oceanic waters. The estuaries are governed by several forcing factors such as river inflow, tides, waves and winds, thus, temperature, salinity, turbidity and other parameters fluctuate on tidal, daily, fortnightly and seasonal basis. Estuaries cover shallow marine waters, freshwater, marshes, swamps, sandy islands, mud and sand flats, oyster's reefs, mangroves tidal pools and sea grasses (Rini, 2002). Intertidal areas comprising of mud/sand flats and mangroves are constantly influenced by varying energy conditions. Mudflats and sheltered intertidal sand flats reflect low energy conditions which are characterised by sediment particles of a small to medium diameter, high water content, low permeability and generally low porosity, high organic content and therefore high reducing conditions, high carbon to nitrogen ratio, high microbial population and high sediment stability. On the other hand, mangroves are one of the world's most productive ecosystems and form an important part of the coastal and estuarine environment. They produce organic carbon well in excess of the ecosystem requirement and contribute significantly to global carbon cycle (Bouillon et al., 2008). They have enormous ecological value. They protect and stabilize coast lines, enrich coastal waters, yield commercial products and support coastal fisheries.

Marine and fresh water flow brings in nutrients and sediments into the estuary (Verlaan et al., 1997; Hossain and Eyre, 2001). Marine water flow is regulated by the tidal currents which are effective at moving sediment via tidal asymmetry from areas of higher current energy towards areas of lower energy (Chen et al., 2006; Scully and Friedrichs, 2007). The sediment movement can be either through suspended, saltation or by rolling. Inorganic and organic components are discharged into the estuaries by rivers and sea which includes anthropogenic inputs in addition to natural material. Mixing of river and sea water regulates processes like coagulation, disaggregation, selective sedimentation, resuspension, adsorption, desorption, dissolution and co-precipitation (Anagnostou et al., 2000). In an aquatic environment, many pollutants, especially metals, are easily adsorbed on the suspended particles and flocculation process allows settling of suspended matter and metals. Suspended materials serve as a carrier for both organic and inorganic elements. Further, total suspended matter (TSM) helps in maintaining chemical and biological gradients in the estuary. Estuarine environment and its associated sub- environments, such as mudflats and mangroves, acts as a sink for particle associated contaminants. Factors like size of sediment components, organic carbon content play a crucial role in geochemical behaviour of metals within sediments (Zhang et al., 2002; Bilali et al., 2002). Fine sedimentary deposits, or mud with high organic matter content are the most characteristic features of estuaries which play an important role in mobilisation of trace metals in the estuaries.

Metals may be recycled several times through the sediment water interface before being permanently stored in sediments or released to the overlying waters. The major biogeochemical processes, which regulate the distribution and behaviour of trace metals in water, are mixing of water bodies, particle- water interaction through biological uptake, adsorption- desorption and diagenetic processes in sediments (Bruland, 1983; Chester, 1990a). Pore water is an important intermediate in the remobilization of metals in the diagenetic mechanisms (Shaw et al., 1990; Koschinsky, 2001). Several factors are considered while dealing with fate and distribution of metals. They include partitioning of trace elements, changing redox conditions, the decomposition of organic matter due to microbial activity and complex geochemical reactions (Santos-Echeandia et al., 2009).

The source of material may be natural or anthropogenic. Natural largely includes weathering products. Anthropogenic includes industrial waste discharges such as mining, smelting and refining. Anthropogenic metal input from urban and industrial growth centres, combustion of fossil fuels, domestic waste water, sewage sludge, urban runoff and leachate from solid waste disposal sites (Mance, 1987). Other potential sources include ports, harbours, marinas and mooring sites, also recreational, commercial, and occasionally, military, boating, and shipping activities (Denton et al., 1997). Agricultural activities also contribute to the overall heavy metal loadings to the environment through the spreading of fertilisers, pesticides, wood preservatives (Alloway, 1995).

Physiochemical factors are likely to influence the concentration and distribution of different metal species. Physical processes mostly includes water circulation and sediment deposition processes (Dyer, 1972). Hydrodynamic conditions within an area determine the type of sediments deposited which can provide base for metal accumulation. Adsorption and retention mechanisms of heavy metals varied with sediment type and the metal species. Within the estuary, in addition to estuarine mixing and flocculation, particle properties are major factors which control the deposition of materials (Edzwald and O'Melia, 1975). The pH and Eh may affect the migration of metals at the sediment - water interface.

Four major chemical processes of metal retention include cation exchange; complexation with organics; precipitation as oxides, oxy hydroxides and carbonates; and precipitation as sulphides (Dunbabin and Bowmer, 1992). Sulphide precipitation is important as its formation immobilizes the metals. Under anaerobic environment, sulphate reducing bacteria produce H_2S that may precipitate metals as metal sulphides (Lacerda et al., 1993). The degree of retention of heavy metals in sediment is also affected by sediment characteristics (Tarn and Wong, 2000). The adsorption and desorption of heavy metals in sediments are influenced by many sediment properties such as organic matter, clay content, redox potential, presence of Fe and Mn oxides, presence of other metals, the quantity of heavy metals accumulated, and the alternating aerobic and anoxic conditions (Harter, 1992; Orson et al., 1992; Tarn and Wong, 1996). The organic matter appeared to be the agent through which metals are incorporated into tidal marsh sediments (Orson et al., 1992; Tarn and Wong, 1996). Humic and other organic matter plays an important role in the distribution of trace metals through chelation with non aggregating organic colloids. Organic colloids play an important role in metal transport. Changes in geochemical conditions alter the nature of carrier colloids (Mc Carthy and Zachara, 1989; Clark et al., 1998). Metal binding capacity is dependent on clay content. In addition to the above processes biological process mainly microbial activity in breaking down of organic matter which further influences the redox conditions and thus effect metal mobilisation is also important. Bioturbation also has an effect on the sediment layers in terms of sulphate reduction which cause change in redox conditions in sediments and thus effect metal mobilisation.

1.2. Metal speciation

Templeton et al. (2000) has defined a metal species as a “specific form of an element with respect to isotopic composition, electronic or oxidation state, complex or molecular structure”. Metal speciation is “the distribution of an element amongst defined chemical species in a system”.

Sediments composed of different sedimentary phases such as clay, silt, sand, organic material, oxides of iron and manganese, carbonate and sulphide complexes that act as potential binding sites for trace metals entering estuarine regions (Jonathan et al., 2004). Metals can be present in various forms and generally exhibit different physical and chemical behaviour in terms of chemical interactions, mobility, biological availability and potential toxicity. Changing nature of estuarine environment with respect to the factors i.e. salinity, pH and Eh (Paludan and Morris, 1999) results in changing metal species from one form to the other. Thus, study of metals including speciation is important in order to understand distribution of metals over the various geochemical phases and to elucidate its bioavailability within an estuarine environment.

As stated earlier, metals associate with sediments through various ways such as ion exchange, adsorption, precipitation and complexation (Tessier et al., 1979). Changes in environmental conditions, such as temperature, pH, redox potential and organic ligand concentrations, can cause mobilization of contaminated sediments and thus releasing the elements from solid to liquid phase and causing contamination of surrounding waters. The species that are available for uptake in organism are considered to be the bioavailable fractions. The term bioavailability reflects the rate and the amount of toxic substances that may be taken up in an organism. The chemical partitioning of heavy metals among different sedimentary forms is very important in determining their bio-availability (Luoma, 1983). Determination of total metals in sediments usually gives an approximate approach to assess the environmental impact caused by heavy metals in sediment. The assessment of the environmental risks requires the measurements not only for the total contents of heavy metals in sediments, but also for the amounts of heavy metals in each binding form (Lin et al., 2003).

Many studies have been carried out in geochemistry of sediments in Mandovi estuary which highlights the total metal concentration within sediments. Further (Dessai and Nayak, 2009) have focussed on the speciation of metals in different seasons in surface sediments of Zuari estuary. However, literature on speciation studies within intertidal regions is very poor elsewhere and no such literature is available for Mandovi estuary. Therefore, the chemical analyses of sediments including speciation studies were taken up. Metal speciation is of critical importance to the potential toxicity and mobility of contaminant metals released into the aquatic environment through either natural or anthropogenic processes (Macklin and Dowsett, 1989; Baruah et al., 1996; Buykx et al., 2000; Bird et al., 2005).

1.3. Factors affecting metal speciation

Metal speciation is influenced by various environmental factors

pH and Eh controls the solubility and concentration of metal species (Schubauer et al., 1993). The fractionation of a specific metal species at specific pH level is metal dependent. For metals, as water pH decreases, the partitioning between dissolved and particulate forms favours the dissolved phase (Riba et al., 2004). Changes in H^+ concentrations may cause changes in metal speciation by altering the structure of environmental ligands and metals bound to them. Although increased H^+ concentration generally causes an increase in dissolved metals, increasing in organism toxicity do not necessarily occur, as there is also increased competition between H^+ and dissolved metals for uptake sites on the organism (Wilde et al., 2006)

Water salinity has larger effect on speciation of metals due to complexation with chloride or competition with major salinity ions Cl^-, Na^+, K^+ for the biological ligands and particulates (Hatje et al., 2003). In estuaries, increase in salinity often cause the removal of significant amounts of dissolved organic carbon into particulate phase, through flocculation induced by changes in ionic strength and pH that accompany the mixing of riverine and saline waters (Fox and Wofsy, 1983). Increase in salinity often causes salt induced flocculation phenomenon (Thill et al. 2001) which is a major mechanism influencing transfer of significant amount of the metal from dissolved to particulate form (Teasdale et al., 2003). Increase in salinity results in decreasing the toxic effects of metal exposure to organisms due to competitive and complexing effects of major ions with trace metals (Riba et al., 2004).

Humic substances upon decomposition to organic matter can play an important role in speciation of many metals. Metals bind to various ligands primarily at carboxylic and phenolic functional groups, but other groups like amino and sulfidic groups may also play an important role. Metal interactions with natural organic matter currently remain an immense challenge due to its polydisperse, heterogeneous nature (Panquin et al., 2002) as the composition of natural organic matter can be highly variable, large differences in binding capacities may occur.

Trace metal speciation can be described as a function of the redox potential, in view of the fact that the latter is related to the heterotrophic bacterial activity. As the sediment depth increases, organic matter and oxidised compounds become depleted and metals increasingly exist in reduced forms. Sulphide is the dominant metal binding phase at depth, due to the reduction of sulphate, which is usually freely available to bacteria (Santschi et al., 1990). Thus microbes have an important role in sediment geochemistry.

1.4. Metals and microbes

Metals are directly and/or indirectly involved in all aspects of microbial growth, metabolism and differentiation (Gadd, 1992a). Metals and their compounds interact with microbes in various ways depending on the metal species, organism and environment, while structural components and metabolic activity also influence metal speciation and therefore solubility, mobility, bioavailability and toxicity (Gadd, 2007). Many metals are essential for life, e.g. Na, K, Cu, Zn, Co, Ca, Mg, Mn and Fe, but all can exert toxicity when present above certain threshold concentrations. Other metals, e.g. Cs, Al, Cd, Hg and Pb, have no known essential metabolic functions but all can be accumulated. Microbes are intimately associated with the biogeochemical cycling of metals, and associated elements, where their activities can result in mobilization and immobilization depending on the mechanism involved and the microenvironment where the organism(s) are located (Violante et al., 2008; Ehrlich and Newman, 2009).

Microbes possess transport systems for essential metals; inessential metal species can also be taken up. Microbes are also capable of mediating metal and mineral bio precipitation, e.g. by metabolite production, by changing the physico-chemical micro environmental conditions around the biomass, and also by the indirect release of metal-precipitating substances from other activities, e.g. phosphate from organic decomposition or phosphate mineral solubilization. Microbial cell walls, outer layers, and exopolymers can sorb, bind or entrap many soluble and insoluble metal species as well as clay minerals, colloids, oxides, etc. which also have significant metal-sorption properties. Redox transformations are also widespread in microbial metabolism, some also mediated by the chemical activity of structural components.

Metals exhibit a range of toxicities towards microbes and while toxic effects can arise from natural geochemical events, toxic effects on microbial communities are more commonly associated with anthropogenic contamination or redistribution of toxic metals in aquatic and terrestrial ecosystems. It is commonly accepted that toxic metals, their chemical derivatives, metalloids and organometals can have significant effects on microbial populations and, under toxic conditions, almost every index of microbial activity can be affected (Giller et al., 2009). However, metal toxicity is greatly affected by the physico-chemical nature of the environment and the chemical behaviour of the metal species in question (Gadd and Griffiths, 1978). Despite apparent toxicity, many microbes grow and even flourish in apparently metal-polluted locations and a variety of mechanisms, both active and incidental, contribute to resistance (Avery, 2001; Holden and Adams, 2003). Microbial resistance to toxic metals is widespread, with frequencies ranging from a few per cent in pristine environments to nearly 100 % in heavily polluted environments (Silver and Phung, 2009).

1.5. Literature review

Studies carried out on related area of research using different parameters in the recent past are detailed below in Table 1.1.

Table 1.1. Literature survey on related topics of research

Authors	Parameters analysed	Observations
a) Suspended Matter		
Kessarkar et al., 2010	SPM (suspended particulate matter), Salinity, SEM of suspended matter	Systematic seasonal variations of suspended particulate matter (SPM) along a 44 km transect of the Mandovi estuary was studied by the authors. The results revealed that the concentrations of SPM were low at river-end stations, increase generally seaward, and were highest at sea-end stations of the estuary. An estuarine turbidity maximum (ETM) occurred at sea-end stations during June–September when river discharge was high and also in February-May when river discharge was low. Wind-driven waves and currents, and bio-geochemical processes at the mouth of estuary

		likely played an important role in the formation of ETM, in resuspension and transformation of SPM into floccules and aggregates, and in their upkeep / removal. SPM was characterized by kaolinite-rich and smectite-rich clay mineral suites at the river-end and sea-end stations, respectively. Smectite concentrations increase seawards with the increase in SPM content and were not influenced by salinity.
Qiao et al., 2007	Metals in suspended sediments	They studied metals in suspended sediments from Changjiang (Yangtze River) and Huanghe (Yellow River). Their study reflected that the parent material, weathering process and anthropogenic activities along their drainage basins contributed to the metals in suspended matter. They also reported the seasonal difference in the metal concentration and they related the same to grain size.
Sadhuram et al., 2005	Suspended particulate matter	The study reported high values of total suspended matter in both surface and bottom water of Hooghly Estuary, India, showing the impact of fresh water on sediment transportation.
Stecko and Bendell – Young, 2000	Geochemistry of suspended matter and deposited sediments	The authors have reported distinct seasonality in geochemistry for suspended matter relative to deposited sediments. A major implication of their study was the bioavailability of metals in SPM which changes with the season.
Niencheski and Baumgarten, 2000	Trace metal studies with suspended particulate matter	The study focussed on the source of the SPM within Patos Lagoon estuary. The study indicated enrichment of suspended particles along with trace metals due to local inputs, industrial and harbour activities.
b) Sediment components and bulk geochemistry		
Praveena et al., 2010	Metals	This study was carried out to investigate the dynamics of heavy metals concentration. The results revealed relatively higher concentration of heavy metals at high tide compared to low tide. This observation was complexed by other factors such as redox condition, presence of hydroxides and oxy- hydroxides. The major source of heavy metals in mangrove surface sediment was of anthropogenic origin such as from agricultural, aquaculture and industrial

		activities as reported by authors.
Ratheesh et al., 2010	Texture, pH, total carbon, hydrogen, nitrogen, sulphur and heavy metals	Heavy metals in the surface sediments of two coastal ecosystem of Cochin, southwest India were assessed. The analysis of enrichment factor indicated a minor enrichment for Pb and Zn in mangrove sediments, while extremely severe enrichment for Cd, moderate enrichment for Zn and minor enrichment of Pb. The geo-accumulation index exhibited very low values for all metals except Zn, indicating the sediments of the mangrove ecosystem were unpolluted to moderately polluted by anthropogenic activities. The observations suggested that the mangrove ecosystems were relatively unpolluted but the estuarine stations were under the threat of severe accumulation of the toxic trace metals.
Mendiguchia et al., 2006	Total organic matter and metals	This study was carried out in the Bay of Cadiz (SW Spain) where several intensive marine aquaculture facilities are located. The data obtained was statistically treated to determine whether the spatial and temporal gradients in heavy metals and organic matter in sediments are related to aquaculture. The results suggested that trace metal enrichment in the sediments may be attributed to the fish farm effluents.
c) Speciation		
Ho et al., 2013	Grain size, total metals, speciation and pore water analysis, XRD analysis	The speciation and mobility of some selected trace metals (As, Cu, Mn, Pb and Zn) in sediments with depth was investigated in the Cam River-mouth (Vietnam) by the authors. Porewater and sediment composition, complemented with single (ammonium-EDTA) and sequential (BCR 3-step) extractions and mineralogical analysis (XRD) were studied. Overall decreasing trends of metals with depth in porewater obtained were related to anthropogenic input in upper sediment layers. High pore water concentrations of As, Mn and Pb in oxic and suboxic sediment layers may result in groundwater pollution. Sediment-bound Pb and Mn dominated in the reducible and the acid soluble fraction, respectively, while Cu

		and Zn distribute rather evenly between four extracted fractions. The pore water metal speciation, as predicted by a geochemical model Visual MINTEQ version 3.0, indicated the toxicity of Cu, Mn, Pb and Zn (presented by the proportions of free metal ions) decreases with depth, while the toxicity of As increased when As(III) becomes more abundant. The dissolved concentrations of trace metals were not only controlled by the precipitation/dissolution of discrete hydroxide/oxide, carbonate and phosphate minerals, but also by sorption processes on major sorbents (i.e. As on goethite, and Mn and Zn on calcite and dolomite) reported by authors. Sulphide minerals did not show any control even in the anoxic zone most likely because of the low concentration of sulphur.
Hui et al., 2012	Speciation analysis, (Indices: MRI and RAC)	Modified potential ecological risk index (MRI) was proposed based on the potential ecological risk index (RI) and risk assessment code (RAC) by modifying an index by the authors. The modified index was relevant to the chemical speciation of heavy metals. Xiawan Port, a typical region contaminated by industrial production, was selected as a case study area. The total concentrations and chemical speciation of heavy metals in sediments of Xiawan Port were analyzed. The experimental data indicated that Xiawan Port is seriously polluted by heavy metals, especially by Cd. The risks of heavy metals were evaluated by RI, RAC and MRI. The results of MRI showed that the risks of heavy metals are in the decreasing order of Cd>Pb>Cu>Zn. Comparison of results by different methods reveals that MRI integrates the characters of RI and RAC. MRI is recognized to be useful for risk management of heavy metals in sediments.
Massolo et al., 2012	pH, organic carbon, total metal concentration	Metal concentrations were near the background level except for As for which a moderate pollution can be hypothesized reported by the authors. The mobility order of the metals was: Cd>Mn>Cu>Zn>As>Co> Pb>Ni>Fe>Cr>Al. The highest percentage of

		Cd (>60%) was found in the most labile phase. Residual fraction was prevailing for Fe, Cr and Al, while Pb was mainly associated with the reducible fraction. Data were compared with Sediment Quality Guidelines to estimate the relationship between element concentrations and adverse biological effects on benthic community, finding the possibility of some toxic effects due to the presence of As in the entire studied area and Cd, only in Calcutta.
Olutuna et al., 2012	Speciation analysis, pH	The species and total mean metal concentrations of some potentially environmental toxic metals (manganese (Mn), iron (Fe), copper (Cu), zinc (Zn), and lead (Pb) in the sediments of Aiba reservoir were examined by the authors. The speciation result revealed that high levels of the metals studied (Mn, Fe, Cu, Zn, and Pb) were associated with exchangeable and carbonate bound fractions, pointing out that they are in potentially available forms and may pose serious problems to the reservoir ecosystem.
Pokrovsky et al., 2012	Grain size, DOC, alkalinity, organic carbon, pore water analysis, speciation of metals	The colloidal distribution and size fractionation of organic carbon and trace elements were studied. There was also a clear change in the vertical pattern of the percentage of colloidal Al, Ti, V, Cr, Fe, and Ni during different seasons, and the greatest proportion of colloidal forms was observed during the spring and autumn overturn. This pattern was linked to the dominance of soil (allochthonous) organic carbon, which complexes with trace metals during these periods. During the summer seasons, autochthonous production of small exometabolites or photodegradation increases the concentration of the low molecular weight fractions (1 kDa) that dominate the speciation of divalent heavy metals.
Davutluoglu et al., 2011	Grain size, Total organic matter, Metals, Speciation analysis	Chemical fractionation of seven heavy metals (Cd, Cr, Cu, Mn, Ni, Pb and Zn) was studied using modified three- step sequential procedure, to assess their impacts in the sediments of Seyhan River, Turkey. Metal fractionation showed that, except for Mn and Pb, the majority of metals were found in

		residual fraction regardless of sampling time, indicating that these metals were strongly bound to the sediments. Based on RAC classification, Cd and Cr posed no risk, Cu and Ni posed low risk, Pb and Zn were classified as medium risk metals, while the environmental risk from Mn was classified as high.
Wang et al., 2010	Speciation analysis, Total organic matter	This work investigated the distribution and speciation of Cd, Cu, Pb, Fe and Mn in the shallow sediments of Jinzhou Bay, Northeast China, which has been heavily contaminated by nonferrous smelting activities. The concentrations of Cd, Cu and Pb in sediments were found to be 100, 13 and 7 times, respectively, being higher than the national guideline (GB 18668-2002). Sequential extraction test showed that 39%–61% of Cd was exchangeable fractions, indicating that Cd in the sediments posed a high risk to local environments. While Cu and Pb were at moderate risk levels. According to the relationships between percentage of metal speciation and total metal concentration, it was concluded that the distributions of Cd, Cu and Pb in some geochemical fractions were dynamic in the process of pollutants migration and the stability of metals in sediments of Jinzhou Bay decreased in the order of Pb > Cu > Cd.
Ibhadon et al., 2004	Total metal concentration and speciation of metals	Speciation results reported by the authors showed that contamination of the estuary was mainly from anthropogenic sources. Pb and Zn are associated with the reducible, residual and oxidisable fractions. The speciation pattern of Cd was similar to those of Pb and Zn. However there were also some exchangeable and bound to carbonate fractions although these were less significant. Cu was largely associated with the oxidisable and residual fractions, with insignificant bound to carbonate, exchangeable and reducible fractions. The most bioavailable forms of the metals were the free inorganic ions.

Chandra Sekhar et al., 2003	Sediment characterisation, total metal contents in sediments and fish, speciation of metals	In this paper the metal concentrations were found greater than the background concentration of sediments indicating the anthropogenic origin of metals. Large fractions of Zn, Cd and Cu were associated with mobile fraction of the sediment and showed greater bioaccumulation in fish whereas, Ni and Co were least mobilisable. The results clearly indicate that the fish of Kolleru lake were contaminated with metals and not advisable for human consumption.
d) Microbial		
Omiema and Ideriah, 2012	Organic matter, total heterotrophic counts, counts of petroleum degrading bacteria, faecal coliforms	The distribution of microorganisms in the sediment and overlying water along the shoreline of Abonnema was studied by the authors. The results showed Total Heterotrophic count range of 1.8x10^3 – 3.4x10^3 unit/ml in water and 1.93x10^4 – 6.12x10^4 unit/ml in sediment; Petroleum Degrading bacteria count range of 1–3 unit/ml in water and 2–10 unit/ml in sediment and Total coliform count range of 1-2 unit/ml in water and none in the sediment. Faecal coliforms were absent in all the stations. The Heterotrophic Bacteria count was higher in the sediment than in the water but statistical analysis showed no significant difference ($p<0.05$) between them. There was high organic load at stations 1-3 resulting from discharge of domestic wastes. The population of Heterotrophic Bacteria was significantly above permissible limits. The results of this work showed that the shoreline of Abonnema is contaminated with microorganisms and petroleum and the water quality.
Pereira et al., 2012	Metal tolerance study, total metal analysis and pollution indices	This study focuses on the distribution of metals and related heterotrophic bacterial populations in the surface sediments (0–10 cm) of the Ribandar salterns (Goa, India) during the salt-making (January to May) and non salt-making seasons (August and November). In general, computation of “geoaccumulation index” revealed the sediments as ‘uncontaminated to moderately contaminated’ with Fe, Mn, Ni, Co, Pb and Zn

		during the salt-making season. The abundance of metal-tolerant bacteria was comparatively restricted to the salt-making season and was higher than the non salt-making season.
Ramkumar et al., 2012	pH, dissolved oxygen, salinity, temperature, total heterotrophic count	Qualitative and quantitative analysis of the composition of the microbial flora were conducted on samples from three stations by the authors. The highest bacterial densities, in water and in sediment samples, were found in December and the lowest, in August. Among Gram-negative bacteria, the predominant genus was Pseudomonas; Aeromonas, Vibrio and Flavobacterium were also recorded. Gram-positive bacilli were abundant at all sampling points.
Govindarajan et al., 2012	pH, temperature, salinity, ammonia, nitrate, total nitrogen, inorganic phosphorus, Silicate, dissolved oxygen, total heterotrophic counts	The water quality of different aquatic ecosystems viz., Effluent discharging and non-discharging sites of Uppanar estuary, Vellar estuary and Pitchavaram mangrove forest in Cuddalore coastal area, Southeast coast of Tamil Nadu, India were examined using selected physicochemical and microbiological indicators by the authors. The physicochemical characteristics such as pH, Temperature, salinity, ammonia, nitrate, total nitrogen, inorganic phosphate, total phosphorus, silicate and dissolved oxygen (DO) were examined. The microbiological indicators like total heterotrophic bacterial count (THB), *Salmonella* spp. *Shigella* spp. and *Klebsiella* spp. also were enumerated from collected water and sediment samples with using appropriate selective medium. The results of bacteriological studies revealed that THB and pathogenic bacterial densities were higher in sediments compared to water sample and were substantially high and much beyond the permissible limit of World Health Organization (WHO).
Mahalaxmi et al., 2011	pH, salinity, temperature, pathogenic total heterotrophic counts	Authors reported results of water and sediment samples of Uppanar estuary were collected in order to understand the distribution of THC and pathogenic bacteria. Results showed higher count of THC in water and sediment and more towards the monsoon season and this could be due to terrigenous material through

		land runoff carrying high bacterial population.
Nagvenkar and Ramaiah, 2010	Arsenic resistant bacteria counts and sensitivity towards antibiotics	Bacterial isolates from water and sediment samples from freshwater, estuarine and marine regions were tested for their growth in the presence of different concentrations of arsenic by the authors. Despite the generation times being longer in case of all bacterial isolates tested in nutrient broth with 200 ppm Arsenite (As +3), many of them were able to attain log phase and substantial growth between 36 and 96 h. The isolates tolerating C200 ppm arsenic (As) were found to belong to Enterobacteriaceae, Pseudomonas, Corynebaterium, Xanthomonas, Acinetobacter, Flavimonas and Micrococcus.
Ogbonna, 2010	pH, dissolved oxygen, turbidity, alkalinity, THC (Total heterotrophic counts), THF (Total heterotrophic fungi), HUF (Hydrocarbon utilising fungi), HUB (hydrocarbon utilising bacteria),	Microbial quality and physicochemical indices of an Imo river, Nigeria, receiving industrial pollutants were investigated during the periods of July 2007 and March 2009 by the authors. Results showed that mean counts for THB and THF of the river system were higher in the wet season than in the dry season. Other parameters such as TDS, total alkalinity, conductivity, free carbon and sulphate showed similar patterns of variation with low values for pH. The influence of physicochemical parameters on the microbial abundance showed that at $P \leq 0.05$, only Total Hydrocarbon, Conductivity and Nitrate correlated positively with HUF and did not show correlation for other microbial groups. Apart from the HUB, and HUF the highest counts also observed for THB and THF confirm that hydrocarbon pollution do not only enrich hydrocarbon utilizers but also for additional populations that utilize the by-products as sources of nutrients.
Gounou et al., 2010	Speciation of metals, Total metal analysis, Microbial metabolism (Quantification of CO_2)	The impact of autochthonous anaerobic bacteria on the release of metals in river sediment was studied. The results showed that metal release was correlated to the bacterial growth (carbon mineralization). In particular, a relationship between iron reduction and metal release was observed indicating that iron-reducing bacteria had a strong influence. By reductive dissolution of iron oxides, bacteria

		also released their associated toxic elements into the liquid phase. While organic analysis showed acetate and butyrate production leading to a decrease in pH and indicating a Clostridium fermentative bacteria activity, the results did not indicate any direct role of organic acids in the dissolution of iron and their associated metals.
Fontana et al., 2010	Grain size, Organic matter, Bacterial Carbon	The authors with an aim to quantify the biopolymers associated to esterase enzymes and identify bacterial respiratory activity in four cores collected in Suruí Mangrove, Guanabara Bay – RJ, Brazil. Biopolymer concentration was 1000 times lower than previously reported in the literature, indicating the need for creating and establishing eutrophication indicative rates and records compatible with tropical coastal systems. The biochemical representative relationships in the cores were equivalent to those from studies on coastal marine environments made in the Northern Hemisphere. The esterase enzymes in the sediment proved efficient in the mineralization of biopolymers, even with preferentially anaerobic metabolic physiology. Despite the lack of incipient geomicrobiological studies, the results highlighted the possible application of microbiology to a better understanding of geological processes.
Li and Wang, 2010	pH, DOC, nitrogen and phosphorus, metal analysis, speciation of metals and microbiological analysis	Results reported indicated that bacterial inoculation significantly enhanced the extractability of water-soluble Cd, Cu, and Zn (single-chemical extraction) in all metal mine soil treatments. However, inoculation of bacteria had a minor effect on As and Pb extractability. Increasing water-soluble and Olsen-P in soil solution induced by the presence of bacteria may contribute to the decrease of water-soluble Pb through sedimentation of Pb phosphate. Bacterial inoculation also increased the mobility of heavy metals (sequential chemical extraction). HOAc soluble Cd and Zn increased as the pH value decreased since the metal adsorption on colloids weakened due

		to a decrease in pH-dependent negative charge. In addition, a significant increase in water-soluble N, P, NaHCO3-extractable P, and soil acid phosphatase activity were observed. Based on the above results, the authors concluded that metal tolerant bacteria are efficient in increasing the bioavailability of Cd and Zn when expressed relative to the control treatment, which might be of great significance for the successful phytoextraction of metal polluted sites.
Boer et al., 2009	Grain size, total heterotrophic counts of bacteria	Temporal dynamics and vertical patterns in bacterial abundances and activities were studied in a shallow subtidal sand flat by the authors. Temporal variations that were mostly related to seasonal temperature change and to changes in substrate availability.
Parameshwari et al., 2009	pH, metal analysis and metal tolerant bacteria	Authors reported Chromium resistant bacteria such as *Pseudomonas fluorescens* and *Bacillus* sp isolated from heavy metal contaminated soil and examined for their tolerance to hexavalent chromium and their ability to reduce Cr (VI) to Cr (III). The influence of various factors such as pH, time interval and initial metal concentrations on the reduction of chromium by the bacterial isolates were studied. Both the bacterial isolates tolerated Cr (VI) at 100 ppm in minimal salt broth. The results indicated that the microbial consortia and the mono cultures of the above isolates can be useful for Cr (VI) detoxification of chromium contaminated environment as per authors.
Ryan et al., 2005	Soil analyses (soil matrix composition, organic content, pH and general chemical parameters), metal tolerant bacteria (MIC), plasmid screening	This paper details the isolation and characterisation of metal resistant microorganisms isolated from rhizosphere/soil samples obtained from abandoned zinc, lead and copper mine and a local unaffected site. This paper outlines the diversity of transferable resistance determinants between both sites and details the levels of heavy metal resistant bacteria and those expressing transferable multiple heavy metal tolerance. The paper concludes that the number of single and multiple heavy metal resistant bacteria

		may be an indictor of the level of contamination.
Vijver et al., 2003	Metal concentration in sediments and metal contents in larvae within sediments	Bioassays were performed to evaluate the impact of soil characteristics on Cd, Cu, Pb, and Zn uptake by larvae of Tenebrio molitor. A comparison of the findings with studies on other soil-inhabiting species shows that metal uptake patterns depend on metal type, soil type, and exposed species.
Chong and Wong, 2000	Metals concentration in mussels	The authors assessed the degree to which Cd, Cr and Zn bound with sediment were assimilated by the green mussel Perna viridis and the Manila clam Ruditapes philippinarum. The higher assimilation of Cd with increasing Cd load in ingested sediment may be due to the higher desorption of Cd in the gut of the bivalves. In the mussels, assimilation efficiency (AE) was higher from oxic sediment than from anoxic sediment. This study suggested that metals associated with anoxic sediment can be potentially available to marine bivalves.

1.6. Objectives

To identify and quantify the metal forms, to gain precise understanding of the potential and actual impacts of elevated metal levels in the sediment, the present study was carried out on sediments collected from intertidal regions of Mandovi estuary with following objectives

4) To study the abundance and distribution of metals in sediments from Mangroves and Mudflats and mangroves.
5) To understand the role of various physicochemical and geochemical parameters in the distribution and concentration of metal species in Mudflats and Mangroves.
6) To investigate metal tolerance of bacterial isolates.

1.7. Study area

Rivers Mandovi and Zuari, major rivers of Goa, together with the Cumbharjua Canal form an important estuarine system along west coast of India.

The present study is restricted to Mandovi estuary. The Mandovi is a tropical estuary located on the west coast of India draining into the Arabian Sea, known to respond to seasonal monsoonal changes quite significantly. In addition, increasing human interferences since the beginning of industrial revolution also affects Mandovi. As the continental runoff is a manifestation of monsoonal cycle, inventory of anthropogenically added components, are assumed to be strongly modulated by natural (monsoonal) forcing. The Mandovi is one of the major rivers of Goa that drains into the eastern Arabian Sea. The estuarine region is endowed with thick mangrove vegetation and several smaller streams carrying weathered material from the Western Ghats drain into the estuarine region. The Western Ghats which run parallel to the west coast with an average elevation of ~1 km, force strong rainfall during the SW monsoon over the western coastal plains. The bulk of the annual precipitation (~90%) occurs from June to September resulting in heavy continental runoff to the Arabian Sea during this period. As a result, the estuarine regions of the Mandovi river experiences extreme seasonal variability with nearly freshwater conditions prevailing during the south west monsoon and almost marine conditions during the non-south west monsoon periods (Qasim and Sen Gupta 1981).

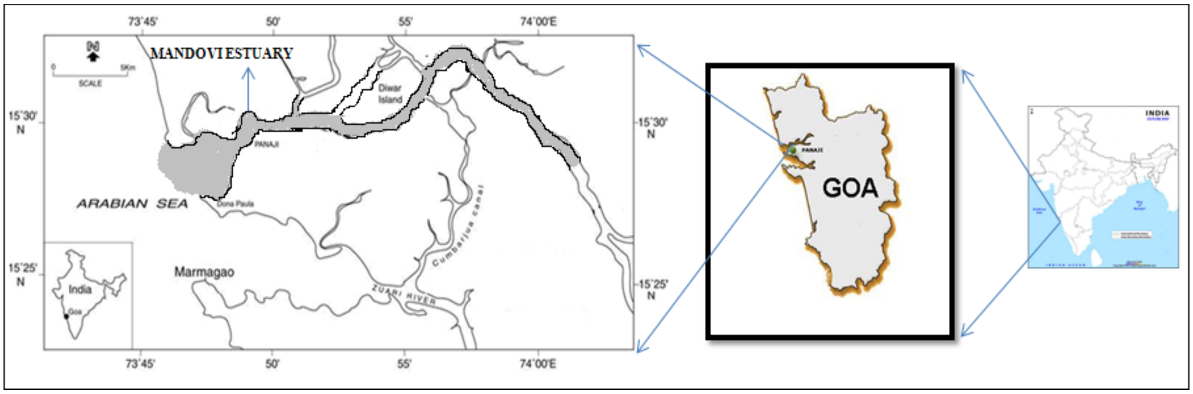


Fig 1.1. Location of the study area – Mandovi Estuary

The Mandovi estuary, is about ~50 km long and ~5 m deep, covering an area of ~29 km2 (Alagarsamy 2006). The shallow depth generally results in well-mixed conditions except at the estuarine mouth where a salt wedge exists during the south west monsoon (Qasim and Sen Gupta 1981). Based on geographical features, the Mandovi estuary can be divided into two regions i.e. the bay and the channel (Shetye et al. 2007b). The bay portion, known as the Aguada Bay is much wider (~4 km) compared to the channel giving the estuary a funnel like appearance. The city of Panjim is located on banks of the Aguada Bay and the river passing through land area is intensely used for both industrial and agricultural activities. The Mandovi estuary thus receives municipal, industrial and agricultural wastes from adjacent areas. Other prominent human activities in the Mandovi estuary include navigation by fishing trawlers, barges carrying iron and manganese ore, and passenger ferries; and recreational activities like boat cruises for tourists. Though treated sewage from Panjim city is discharged near the bottom at a location 3.93 km upstream from the estuarine mouth both the channel and bay regions receive untreated sewage as well.

More than 10 million tons of iron ore are being exported annually through Marmugao port. It is also important to mention here that Mandovi estuary, which is connected to Zuari Estuary through Cumbharjua canal, also passes through extensive mining activity. Therefore, present study focuses on the concentration and distribution of trace metals in suspended matter and sediments in order to understand the status of metal concentration in estuarine sediments both spatial and temporal. Further there are no studies on speciation of metals i.e. studies of metal distribution among various geochemical phases of sediments and metal tolerance of bacteria isolated from the same. Thus, in the present study an attempt has been made to determine the degree of association of the metals in the sediments in terms of bioavailability and to what extent microbes tolerate the metal concentration in the sediments.

CHAPTER TWO

MATERIALS AND METHODS

2.1. Introduction

The sample is a representative of a given environment, location, ecosystem etc. Selection of sample depends on the objectives of the study. It is necessary to have a thorough knowledge on sample selection, collection, preservation and subsequent analysis for generating good set of data. Care should be taken while sampling to avoid contamination, as it may lead to production of incorrect results.

Subsequent to sample collection is preservation it is an important step where there can be a possibility of alteration of chemical components in the sample which may lead to erroneous results. Further, employing standard methods to carry out the analysis is important. The experiment should be repeated to attain accuracy.

Considering the importance of every step involved in the generation of good quality data, they were strictly maintained in the present study. The methods adopted in the collection and analyses are detailed in this chapter and are also outlined in the figure 2.1.

2.2. Field methods

2.2.1. Collection of water samples

Field survey and sample collections were carried out during August, January and April months representing monsoon, post monsoon and pre monsoon seasons in the Mandovi estuary on board fishing trawler during the year 2008 – 2009. GPS was used for locating the stations.

A total of five stations were selected within the estuarine limits of Mandovi River for the collection of water samples. Near surface and near bottom water samples were collected at every selected station (Fig.2.2a) during all the three seasons using a Niskin plastic water sampler fastened on to a rope and was triggered by a messenger. Collected water samples were transferred to precleaned plastic bottles and shifted to laboratory.

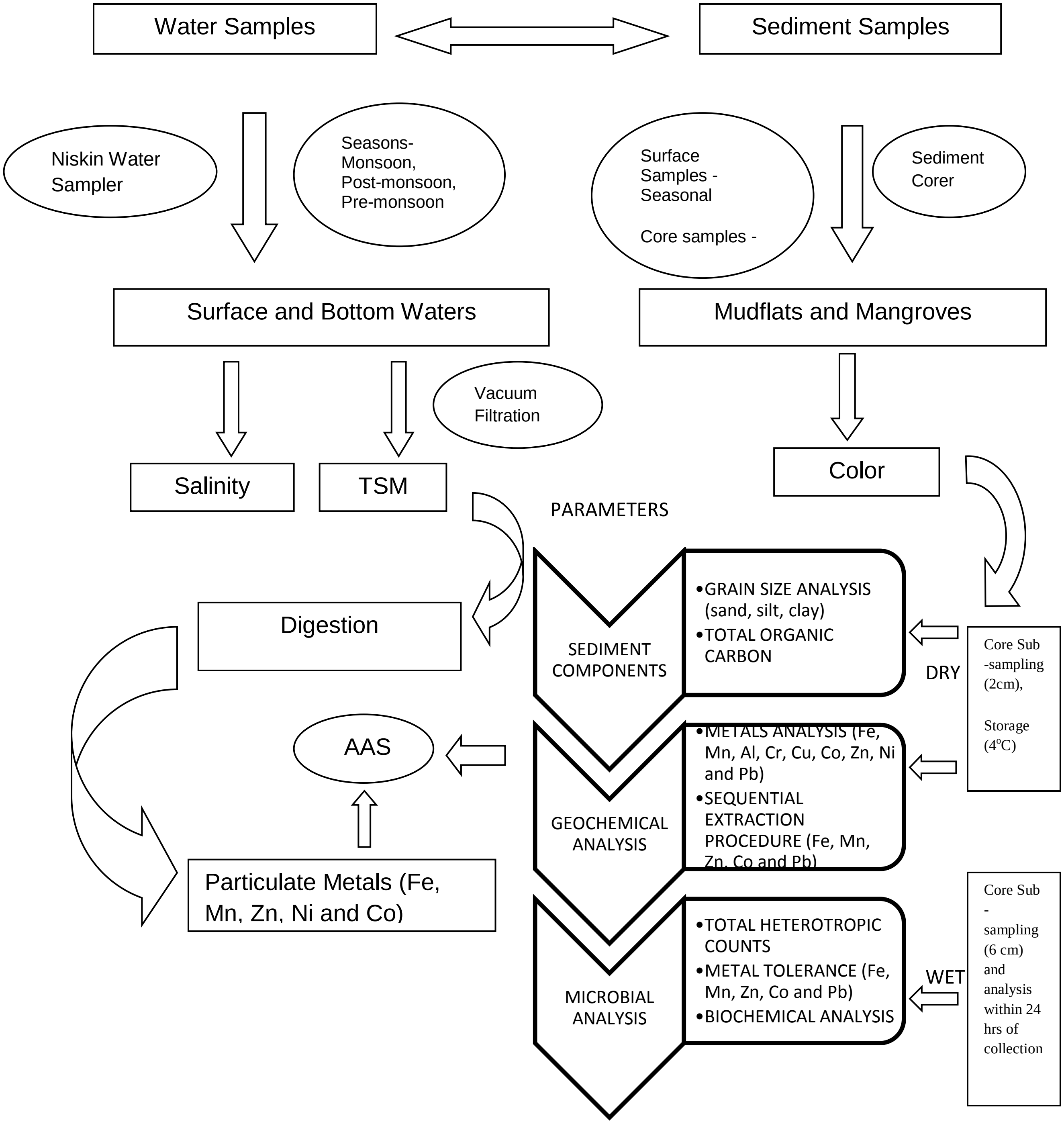


Fig 2.1. Schematic diagram showing sampling and laboratory analysis

2.2.2. Collection of sediment samples

Field studies were also carried out for a period of one year representing seasons from two sub environments namely mangroves and mudflats. Sampling was carried out during low tide representing monsoon, post-monsoon and pre monsoon seasons. Location of collection sites (Fig.2.2b) were recorded using GPS. Sediment core samples were collected representing lower estuary (Betim), lower middle estuary (Karyabhat), upper middle estuary (Ribander) and Upper estuary (Old Goa) during pre-monsoon. Of the total 7 cores, 3 sediment cores were collected from Mudflat environment (from first 3 locations) and 4 cores from Mangrove environments (from all the 4 locations). Cores were collected using hand held PVC sediment corer which was of approximately 30 cm in length and with 6.5 cm diameter. At each of the locations, collection was done in triplicates, of which one core was used for sedimentological and geochemical analysis, second one was used for microbiological analysis and third core was kept reserved as reference. In addition, surface samples were collected during monsoon and post-monsoon from the same locations in order to understand seasonal variations of sedimentological, geochemical and microbiological parameters. The cores collected during pre-monsoon season was used to understand variation of above parameters in lower, lower middle and upper middle regions in both mudflat and mangrove environment in addition to mangrove region of upper estuary with depth. Steps involved in sample collection and analysis is given in figure 2.1. Among the three mudflat sampling locations, Betim and Ribander were from main river channel whereas station Karyabhat is located in the sub channel of Mandovi Estuary.

Further, prior to the sub sampling of sediment cores, color variation was noted (Table 2.1). The sediment cores were sub sampled at 2 cm interval using plastic sterile knife to avoid metal contamination and subsamples were stored in zip lock plastic bags and transferred to laboratory in ice box. In the laboratory, the samples were stored in a deep freezer. Before the analysis they were thawed, dried at 50-60° C in an oven and part of the sediment was powdered and homogenized in an agate mortar. This part of the sample was used for the analyses of the bulk chemistry and total organic carbon. Other part of the sediment which was not powered was used for granulometric analysis to obtain sand, silt and clay components. All the sample containers and laboratory glass wares were pre-cleaned with detergent, soaked in HNO_3 (10%) for 24 hrs and rinsed with deionised distilled water before use. Acids and standard solutions used for the digestion and analyses were of suprapure grade.

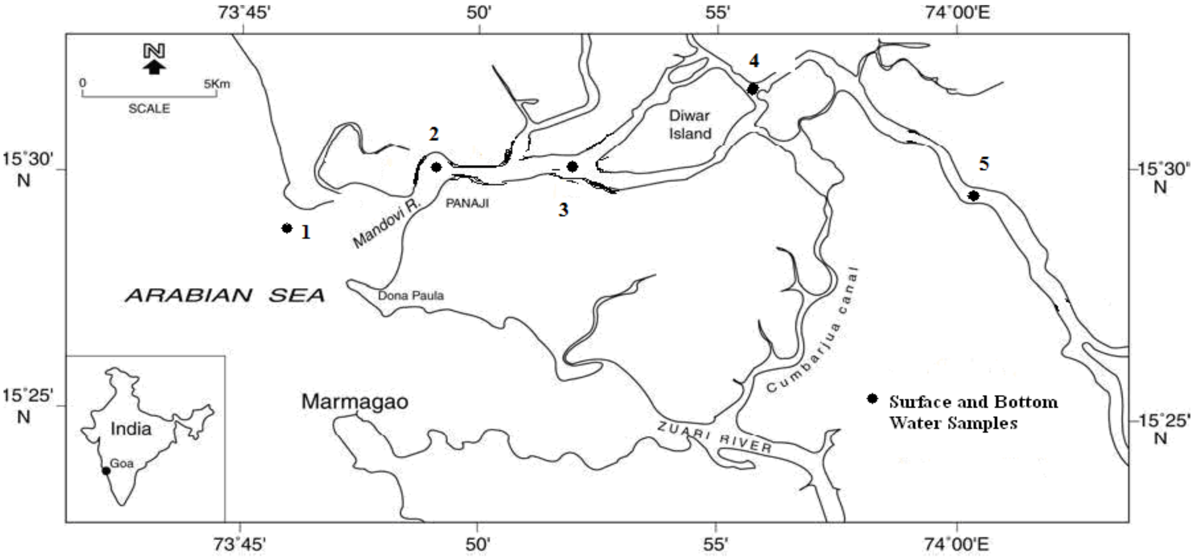


Fig.2.2a. Map showing the water sampling locations

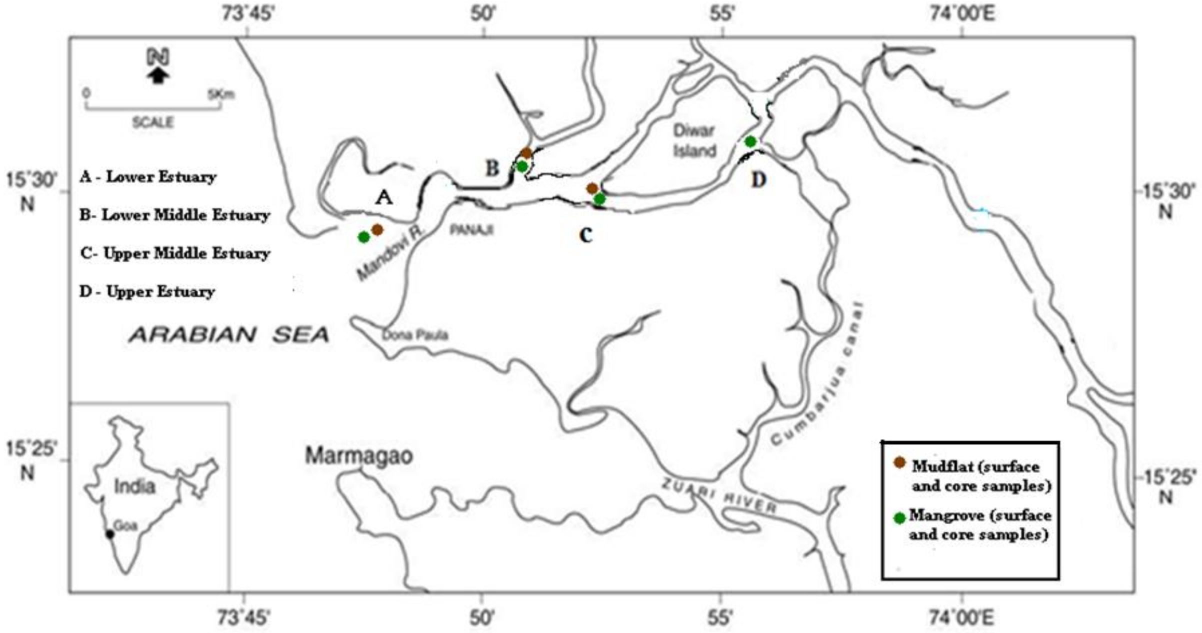


Fig.2.2b. Map showing the sediment sampling locations

Table 2.1. Details of sediment core samples collected

Station	Latitude – Longitude	Core - length	Color
BETIM	15^0 30’ 35.0” N 73^0 49’ 18.2” E	Mangrove – 18 cm Sand flat – 18 cm	Brownish black Brown
RIBANDER	15^0 30’ 15.6” N 73^0 51’ 44.7” E	Mangrove – 22 cm Mudflat – 18 cm	Brownish black Brownish black
KARYABHAT	15^0 30’ 30.3” N 73^0 51’ 16.3” E	Mangrove – 18 cm Mudflat – 18 cm	Blackish grey Brownish black
OLD - GOA	15^0 30’ 21.6” N 73^0 54’ 44.4” E	Mangrove – 18 cm	Brownish black

2.3. Laboratory analysis

2.3.1. Water analysis

Samples brought to the laboratory were used for the analysis of salinity, total suspended matter and particulate metals.

a) *Salinity*

Chlorinity was measured by Mohr Knudsen chlorinity titration method (Grasshoff, 1983), which involves the following procedure. 10 ml of sample was taken into the conical flask, 25 ml of chloride free distilled water and 6 drops of potassium chromate (K_2CrO_4) indicator were added to it. The contents of the conical flasks were mixed well and then titrated against standard silver nitrate solution until color was changed to pink. Standardization of the silver nitrate solution was done by using the standard sea water.

Salinity was then calculated using the formula

Salinity = 1.80655 *Chlorinity (Cl) psu.

b) *Total suspended matter*

A known volume of water sample (5 litres) was vacuum filtered through pre-weighed millipore membrane filter having a pore size of 0.45 µm. The filter paper was then oven dried at 60^0 C and reweighed on the four-decimal balance. Total suspended matter (TSM) concentration was then calculated using the sample volume and sample weight. TSM was expressed as mg/l.

c) *Particulate metals*

Filter paper with sample was digested using 1:1 $HClO_4$ and HNO_3 following the procedure given by Satyanarayana et al. (1985). The digested sample was diluted up to 50 ml with millique water and stored in plastic bottle for further analysis. Samples were analyzed for five selected elements viz. Fe, Mn Co, Ni and Zn using atomic absorption spectrophotometer (VARIAN – AA 240 FS model) equipped with deuterium background corrections. Blank corrections were applied for all the metals.

2.3.2. Sediment analysis

Part of the sub sampled sediment was used for granulometric analysis to obtain sand, silt and clay components as mentioned earlier.

a) Sediment component (sand, silt, clay)

Sediment component analysis was carried out following pipette method given by Folk (1968) in which sand, silt and clay percentage in sediment was estimated by wet sieving. The analysis is based on Stokes settling velocity principle. A known quantity of oven dried sediment sample (10 gm) was transferred into 1000 ml beaker and distilled water was added and stirred and kept for settling. Next day, the water was decanted by using decanting pipe without disturbing the sediment in the beaker and again distilled water was added to the beaker. This procedure was repeated for three times for desalinization. After decanting, 10 ml of 10% sodium-hexametaphosphate solution was added to dissociate the clay particles. Next day 5 ml of 30% hydrogen peroxide was added to oxidize organic matter. The sample was then passed through a 63 micron (230 mesh size) sieve. The filtrate was collected in 1000 ml cylinder for pipette analysis. Complete wash of clay and silt from the sieve was ensured by washing the contents of the sieve till solution becomes clear. Then, +230 fractions were transferred to a pre-weighed beaker and kept in the oven. The filtrate collected in 1000 ml cylinder was made to 1000 ml by adding distilled water and it was homogenized by stirring for about 2 minutes using stirrer. 25 ml of the sample representing the clay fraction (8φ) was withdrawn by inserting the pipette up to 10 ml depth and specific time at specific temperature following the table 2.2 given below.

Table 2.2. Time schedule used for pipette analysis

Size (φ)	Depth to which pipette is inserted (cm)	Time at which water is pipetted out (Hours: Minutes: Seconds)				
		28^oC	29^oC	30^oC	31^oC	32^oC
4	20	0:00:48	0:00:46	0:00:46	0:00:44	0:00:44
5	10	0:01:36	0:01:34	0:01:32	0:01:29	0:01:28
6	10	0:06:25	0:06:15	0:06:06	0:06:57	0:05:52
7	10	0:25:40	0:25:02	0:24:25	0:24:49	0:23:27
8	10	1:42:45	1:40:13	0:37:42	1:37:15	1:33:51
9	10	6:30:00	6:40:40	6:32:50	6:32:10	6:11:30
10	10	27:06:00	26:30:00			

(Standardized by NIO)

The pipetted solution was transferred to a pre weighed 100 ml beaker and dried at 60^0C. After drying, the beakers were weighed and the percentages of sand, silt and clay were computed as follows

% Sand = (Weight. of sand /Total Weight.)*100

% Clay = {[(Weight. of clay / Total Weight.) *(1000/25)] -1}* 100

% Silt = 100 – (% of sand + % of clay)

b) *Estimation of organic carbon (OC)*

Total organic carbon was determined using the standard method detailed by Gaudette et al. (1974) in which exothermic heating and oxidation with $K_2Cr_2O_7$ and concentrated H_2SO_4 are followed by titration of excess dichromate with 0.5 N $Fe(NH_4)_2(SO_4)_2.\ 6H_2O$. This method provides excellent agreement with the LECO combustion method of organic carbon. An aliquot of dried homogenised sediment (0.5 g) was treated with 1N dichromate solution (10 ml) and 20 ml of acid mixture (silver sulphate dissolved in sulphuric acid). They were mixed by gently rotating the flask for about 1 minute and allowed to stand for 30 minutes. This was then followed by addition of a mixture of 10 ml of 85% H_3PO_4 and 0.2 g NaF, and excess dichromate was then back titrated with standard ammonium solution using 15 drops (0.5 ml) of Diphenylamine as an indicator. Dextrose was taken as standard for the determination of organic carbon.

Calculation of results

Total organic carbon (%) = 10 (1- T/S)* F

Where,

T = Sample titration, ml of ferrous solution

S = Standardization blank titration, ml of ferrous solution

F = Factor, which is derived as

F = (1.0N)*(12/4000)*(100/ sample weight)

= 0.6 when sample weight is exactly 0.5 g

Where 12/4000 = m. eq. weight carbon

c) Metal analysis

The metal analysis was carried out employing open wet digestion method using Teflon beakers and combination of acids. Sediment samples for major and trace metals analysis were digested by using hydrofluoric- nitric acid - perchloric- ($HF+HNO_3+HClO_4$) mixture in (7:3:1) ratio in Teflon beakers. 0.2 g of finely ground sediment sample was transferred into a clean acid washed Teflon beaker. To this, 10 ml of above mixture was slowly added to avoid excessive frothing and was completely dried on the hot plate at 150^o C. After drying, again 5 ml of above mixture was added and dried on hot plate for 1 hr then 2 ml of concentrated HCl was added and dried completely. Final residue was dissolved in 10 ml of 1:1 HNO_3. After ensuring complete digestion (clear solution) of sediment sample, the contents from the Teflon beakers were transferred in to acid washed polypropylene volumetric flasks and solution was made up to 50 ml with millique (18.2 ohm’s) water. Same procedure was followed for digestion of two reference standards namely i) MAG-1 (Marine mud) and ii) GR-1 (Green River sediment). Samples and standards were analysed for nine metals namely Fe, Mn, Al, Cr, Cu, Co, Ni, Zn and Pb, on Atomic Absorption Spectrometry AAS (Varian AAS model AA240FS). Precision was monitored by analysing triplicates samples for some selected samples and was generally < 6% standard deviation (SD %) for major and trace metals.

d) Speciation of metals

A modified sequential extraction procedure (Tessier et al., 1979) was adopted in the present study. It is used to quantify the metals in different operationally defined geochemical phases. Core sediments of both Mudflats and Mangroves collected during pre-monsoon from lower, lower middle and upper middle estuarine regions were analysed for speciation of trace metals.

The steps involved in separating the phases are detailed in figure 2.3 and given briefly below.

1) *Fraction 1*: It is a soluble, exchangeable (Sol/Ex) fraction in which the contaminants are weakly adsorbed to the clays and organics in the sediment via electrostatic attraction. From a biological availability standpoint, metals in Sol/Ex fraction are readily available (Tessier et al., 1979; Patrick et al., 1977). Metals at the weathering site get adsorbed forming an electrical double layer. Magnesium chloride is an effective reagent for desorbing adsorbed trace metals.

2) *Fraction 2:* In this fraction the contaminants are bound to carbonates (Carb). These metals are very susceptible to changes in temperature and pH of the solution and therefore the next most readily available (Tessier et al., 1979). Buffered acetic acid and sodium acetate was applied to leach the metals in this fraction.

3) *Fraction 3:* In this fraction the contaminants are bound to Fe and Mn oxy- hydroxides (Fe/Mn). They exist in sediment as cement, nodules and concretions and tend to be thermodynamically unstable in anoxic conditions. They are most susceptible to changes in Eh (increased availability at low Eh). These contaminants are most biologically available under reducing conditions (Tessier et al., 1979; Patrick et al., 1977).

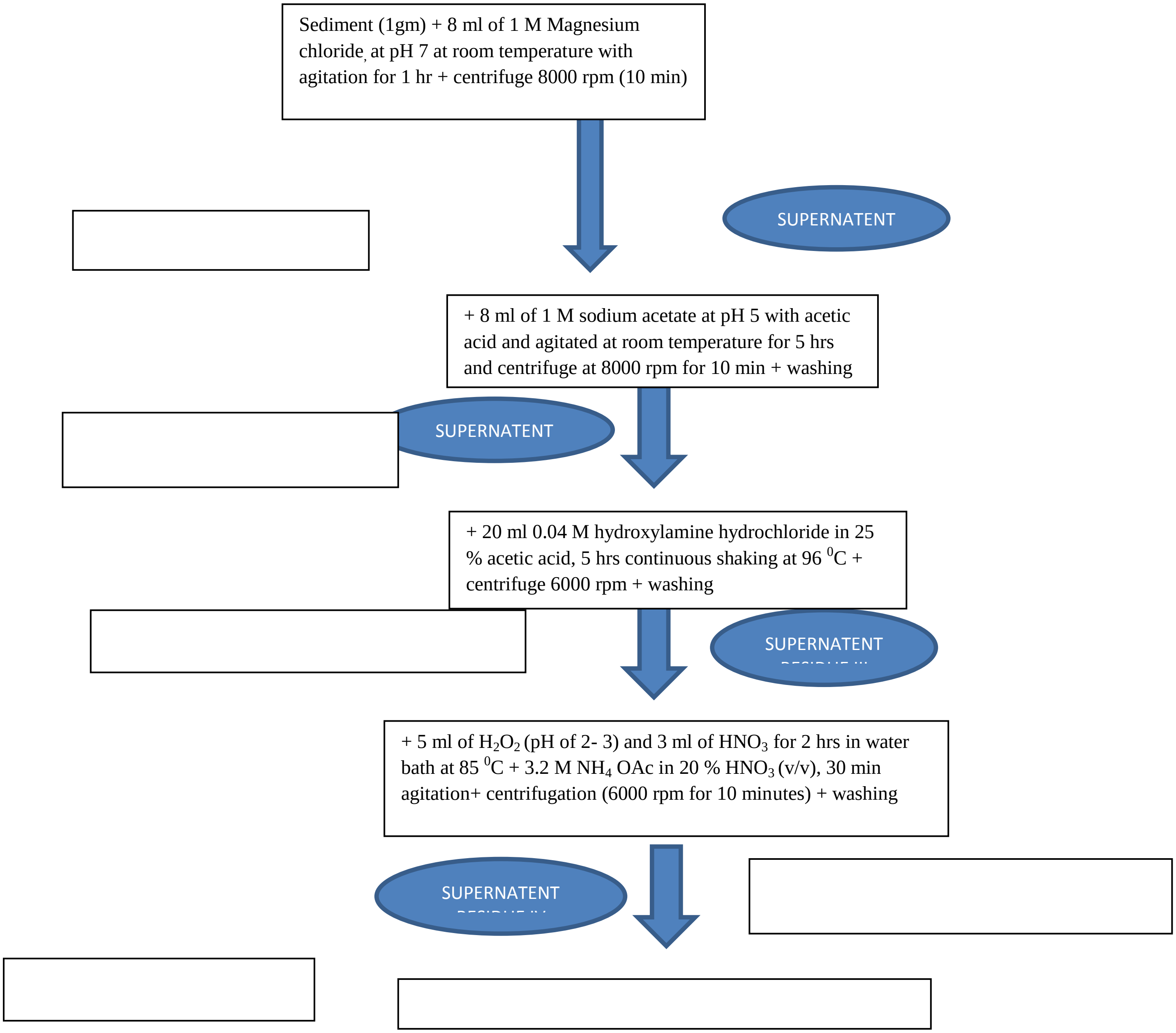


Fig 2.3. Schematic diagram representing sequential extraction procedure

4) *Fraction 4:* The contaminants here are bound to various forms of organic matter (OrgM) with strong bonds. Metals in this fraction may be associated with organic matters, such as organic coatings on inorganic particles including biotic detritus. Under oxidising conditions, these contaminants are released upon degrading the organic matter. Contamination in the OrgM fraction is the least biologically available (Tessier et al., 1979) among the first four phases. Stability of the metals in this fraction is high when compared with above three fractions and thus incorporation in geochemical cycle is difficult. Hydrogen peroxide in acid medium used to oxidise organic matter whereas, Ammonium acetate is used to prevent adsorption of extracted metals on the oxidized sediment.

5) *Fraction 5:* Residual fraction includes metals incorporated into the crystal structure of the primary and secondary minerals. These contaminants are not biologically available and can only be released with the use of a very strong acid such as HF (Tessier et al., 1979; Patrick et al., 1977). Metals in this fraction are inert and may not take part in the biochemical or chemical reactions under normal environmental conditions.

e) Microbiological analysis

The sediment cores collected for microbiological studies were analysed within 24 hrs of the collection. The sediment cores collected during premonsoon were sub sampled inside the laminar air flow into three sections i. e. (0-6 cm); (6-12 cm) and (12-18 cm). Sterilized plastic knife was used to avoid metal contamination and to maintain sterilized condition. The surface samples collected during monsoon and post-monsoon were also studied.

5 gm of sediment sample was inoculated in 45 ml of autoclaved full strength sterile sea water and were mixed thoroughly and serial dilutions were made. The dilutions (0.1 ml) were plated containing enriched media (Zobell marine agar) and Minimal media without metal and with metal: (Fe, Mn, Zn, Co and Pb) in concentration of 25, 50 and 100 ppm of the respective metal. The plating was done in triplicates to avoid manual error. Further, the plates were kept at room temperature and growth were observed after 24 hrs or until growth was obtained.

Heterotrophic Bacterial counts in the form of colony forming units (CFU) formed on to the medium were recorded after 24-48 hrs. Dry weight of the sediment used for the dilutions was determined by drying the filtered sediment in an oven at 60 ^{0}C for 48 hr.

Further, the colonies (Metal tolerant) were picked from the plates of 100 ppm and were further grown in higher concentration of metal i.e. 200; 400, 600; 800 and 1000 ppm to understand the maximum tolerance of the isolates. Further, the cultures which showed highest tolerance were stored for further study which includes Gram character and biochemical analysis.

Gram staining

The cultures were picked and smear was prepared onto the slides with one drop of autoclaved distilled water. The slides were slightly heat fixed and then kept for complete drying. Method involves following procedure. Crystal violet is added to the slides and then rinsed with distilled water after 1 minute. Next step involves addition of Grams Iodine for 1 minute and then subsequent rinsing with distilled water. Further, step involves rinsing of slide with 50 % alcohol and finally flooding the slide with saffranine for 1 minute and then rinsing with distilled water. The slide was air dried and morphology of the cells was observed under oil immersion lens.

Biochemical studies

Himedia kits were used for Biochemical studies which comprises of various sugar tests, IMVIC and ONPG, lysine, orthinine, urease, phenlylanine, nitrate and H_2S production.

2.4. Data processing

a) Statistical analysis

MS Excel and Grapher were used for computation and plotting different parameters respectively. Pearson Correlation Coefficient was employed for the better understanding of relationship between the concentrations of various metals, sediment components by using statistical package STATISTICA (StatSoft, 1999). All the values were normalized using the same software.

b) Pollution indices

The Pollution load index (PLI) proposed by Tomlinson et al. (1980) for computing level of pollution, which permits a comparison of pollution levels between sites and at different seasons, was obtained as a concentration factor for heavy metals with respect to the background value in the sediment. The world average concentration of the metals for shale (Turekian and Wedephol, 1961) was used as the background value for respective heavy metals. The formula applied is as follow

$$PLI=\sqrt[n]{CF1 * CF2 * CF3 * \ldots\ldots\ldots\ldots CFn}$$

Where CF metal= C metal/ C background

Where CF = contamination factor; n = number of
Metals; Cmetal = metal concentration in sediment
Sample; C background = background value of that metal.

According to Tomlinson et al. (1980), PLI values of zero, one, or larger than one suggest absence of baseline pollutants, presence of them, or progressive deterioration of sediment quality, respectively. According to Angula (1996), PLI is able to give an estimate of the metal contamination status.

Enrichment factor (EF) and geoaccumulation index (Igeo) were used to assess the extent of sediment contamination. EF provides the actual contamination level in the sediment (Groengroeft et al., 1998) and is a good tool to differentiate the anthropogenic and natural source of metals (Morillo et al. 2008; Selvaraj et al., 2004; Valdés et al., 2005). The Following equation was used to compute EF.

EF = (Cx/CAl) sediment / (Cx/CAl) shale

Where, (Cx/CAl) sediment is the metal concentration Cx in (µg/g dry weight) in relation to Al (% dry weight) within sediment samples. (Cx/CAl) shale is the metal concentration (Cx) within shale in relation to Al (% dry weight). Shale values are taken from Krauskopf and Bird (1995).

EF values were interpreted following Birch (2003) as EF<1 no enrichment; EF= 1-3, minor enrichment; EF=3–5, moderate enrichment; EF=5–10, moderately severe enrichment; EF=10–25, severe enrichment; EF=25–50, very severe enrichment; and EF>50 as extremely severe enrichment.

Geoaccumulation index (Igeo) was originally defined by Muller (1979) in order to determine metal contamination in sediments by comparing current concentrations with pre-industrial levels. The Igeo values for the metals studied were calculated using the equation given by Muller (1979).

$$\mathrm{Igeo} = \log_2 \frac{C_n}{1.5 \cdot B_n}.$$

Wherein C_n is the measured concentration in the sediment for the metal n, B_n the background value for the metal n and the factor 1.5 is used because of possible variations of the background data due to lithological variations. Several researchers (Subramanian and Mohanachandran, 1990; Barreiro, 1991; Sahu and Bhosale, 1991; Volvoikar and Nayak, 2013) have used the Igeo. Igeo values have been calculated using global average shale data reported by Turekian and Wedepohl (1961), in the present study.

The index of geoaccumulation consists of seven classes (Table 2.3) with Igeo of 6 indicating highest enrichment compared to background values (Müller, 1979).

Table 2.3. Geoaccumulation indices (after Müller, 1979)

Igeo	Igeo class	Pollution Intensity
>5	6	Very strongly polluted
4-5	5	Strong to very strong
3-4	4	Strongly polluted
2-3	3	Moderately to strongly
1-2	2	Moderately polluted
0-1	1	Unpolluted to moderately Polluted
0	0	Unpolluted

CHAPTER THREE

TOTAL SUSPENDED MATTER

3.1. Introduction

Estuaries are traps of suspended matters received from rivers (Turner and Millward, 2002). Metals are primarily associated with suspended particulate matter in the estuarine system due to processes of adsorption onto hydrous iron, manganese oxides, organic substrates (Turner and Millward, 2002) in addition to inorganic particulates. Therefore, suspended sediments provide pathway for transport of associated metals. Distribution pattern of Total Suspended Matter (TSM) within an estuarine region is dependent on salinity variations, terrestrial and marine input and hydrodynamic conditions prevailing within an environment. Processes like resuspension of sediments, flocculation, along with fresh and marine water influx influences, the distribution of suspended matter. TSM distribution varies with space within the estuary. During monsoon salinity, TSM as well as Fe and Mn oxyhydroxides are effective in adsorption of most of the trace metals.

Trace metals bound to suspended particles remain biologically available in the fluid medium. The trace metals are micronutrients, which are essential for the organisms, but at high doses even nutrionally essential metals can cause adverse affects. Once sorbed, they contribute to the pool of metals associated with TSM within the water column and are ultimately incorporated to bed load sediments. Upon deposition, these metals can be accumulated by benthic- dwelling organisms which live and feed on the sediment (Zoumis et al., 2001). Benthic organisms are food for several higher tropic levels and this transfer of contaminants can lead to several adverse effects (Anderson et al., 2007). Within the sediments, once metal concentration exceeds the threshold level, they may lead to severe environmental problems.

3.2. Salinity

In Mandovi estuary, the salinity ranges from 0.1 to 26 psu (Avg.10.36 psu) and 0.2 to 29.2 psu (Avg. 16.64 psu) in surface and bottom waters respectively during monsoon. Surface and bottom waters exhibit very low salinity towards the head at station 5 and show gradual increasing trend towards the mouth of the estuary. Bottom waters show higher salinity when compared to surface waters at all the five stations (Fig 3.1a). This is due to retention of higher salinity in deeper waters and flow of fresh water near the surface during this season. The variation in salinity between surface and bottom is significant at stations 2, 3 and to some extent at station 4. Salinity difference between surface and bottom waters is caused by high precipitation and runoff which is responsible for maintaining a salt wedge and also bringing salinity values low near the surface. During postmonsoon, the salinity of surface waters varies from 16.66 to 34.02 psu (Avg. 23.07 psu) and in bottom waters from 15.62 to 34.25 psu (Avg. 22.99 psu). A gradually increasing trend is observed towards the mouth region in both surface and bottom waters (Fig 3.1c). During premonsoon, salinity varies from 29 to 35.5 psu (Avg. 33. psu) and 29.66 to 35.65 psu (Avg. 33.73 psu) in surface and bottom waters respectively. Not much variation between surface and bottom waters is observed in salinity values during postmonsoon and premonsoon. However, salinity values are higher in premonsoon indicating tidal control on the estuary (Fig 3.1e) during this season. Average salinity is comparatively higher in premonsoon followed by postmonsoon and monsoon.

3.3. Total Suspended Matter (TSM)

During monsoon, TSM of surface waters ranges from 1.8 to 12.30 mg/l (Avg. 6.09 mg/l) and in bottom waters it varies from 1.88 to 36.4 mg/l (Avg. 12.73 mg/l). At almost all the stations TSM values of bottom waters are higher than surface waters with an exception at station 3 (Fig 3.1b). Increasing TSM with depth might be due to resuspension of bottom sediments (Richards, 1982) or due to sinking of larger particles upon flocculation.

In bottom waters, TSM shows gradual increase from station 5 to 3 and maintains a similar value at station 1. However, the highest value is obtained for station 2. The zone of higher suspended matter in lower portions of the estuary is related to estuarine processes such as flocculation which might be responsible in retaining maximum TSM in this region (Nayak and Bukhari, 1992; Regnier and Wollast, 1993).

During Postmonsoon, TSM exhibits a range from 2.75 to 7.72 mg/l (Avg. 4.81 mg/l) and 7.00 to 14.87 mg/l (Avg. 9.29 mg/l) in surface and bottom waters respectively. Surface TSM shows a decreasing trend from station 5 to station 4 thereafter maintaining an increasing trend up to station 2, before showing a decrease towards the mouth of the estuary (Fig 3.1d). In case of bottom waters decreasing trend is observed from station 5 up to station 3 and from station 2 to station 1. Highest suspended matter concentration is observed at station 5 which is towards the upstream of the estuary. Bottom waters maintained higher TSM compared to corresponding surface waters at all the stations indicating resuspension caused by scouring of finer sediments by tidal surge. Also, during non monsoon season, increased ore transport activities are expected to add large suspended matter to the water column. Further, one would expect turbulence related to the propulsion mechanism associated with barges and boats which must be responsible for resuspension of bedload sediments.

TSM ranges from 4.68 to 16.09 mg/l (Avg. 10.9 mg/l) and 5.38 to 66. 3 mg/l (Avg. 26.7 mg/l) in surface and bottom waters respectively during premonsoon. TSM concentration increases from station 4 up to station 2 with lower values both towards the mouth (station 1) and upstream (station 5) in surface waters. In case of bottom waters, TSM shows decreasing trend from station 4 to station 3 with highest concentration at station 4 (Fig 3.1f). Towards the downstream region an increasing trend is observed from station 3 to station 1. Increasing tidal velocity towards the upper portion of the estuary due to constriction might have helped most of the sediment to overcome gravitational settling in this zone, thus increasing residence time of the sediment in suspension, resulting in higher TSM concentration. In addition ore loading and transport must have added material.

In case of monsoon, high concentration of TSM is observed towards the lower portion of the estuary while during non monsoon months the concentration is comparatively higher in upper portions of the estuary especially in bottom waters. On an average higher TSM is observed in premonsoon followed by monsoon and postmonsoon in surface and bottom waters. During monsoon months, the river discharge is high and little time is required to flush out the materials present in the estuary. During premonsoon, suspended matter introduced into the estuary is not flushed out easily because the buoyancy of incoming fresh water is insufficient to overcome the mixing due to strong tidal currents. Suspended matter is lower in postmonsoon as compared to premonsoon in Mandovi estuary. Taylor (2000) has suggested that the presence of algal mats formed in winter months reduce the resuspension of sediments.

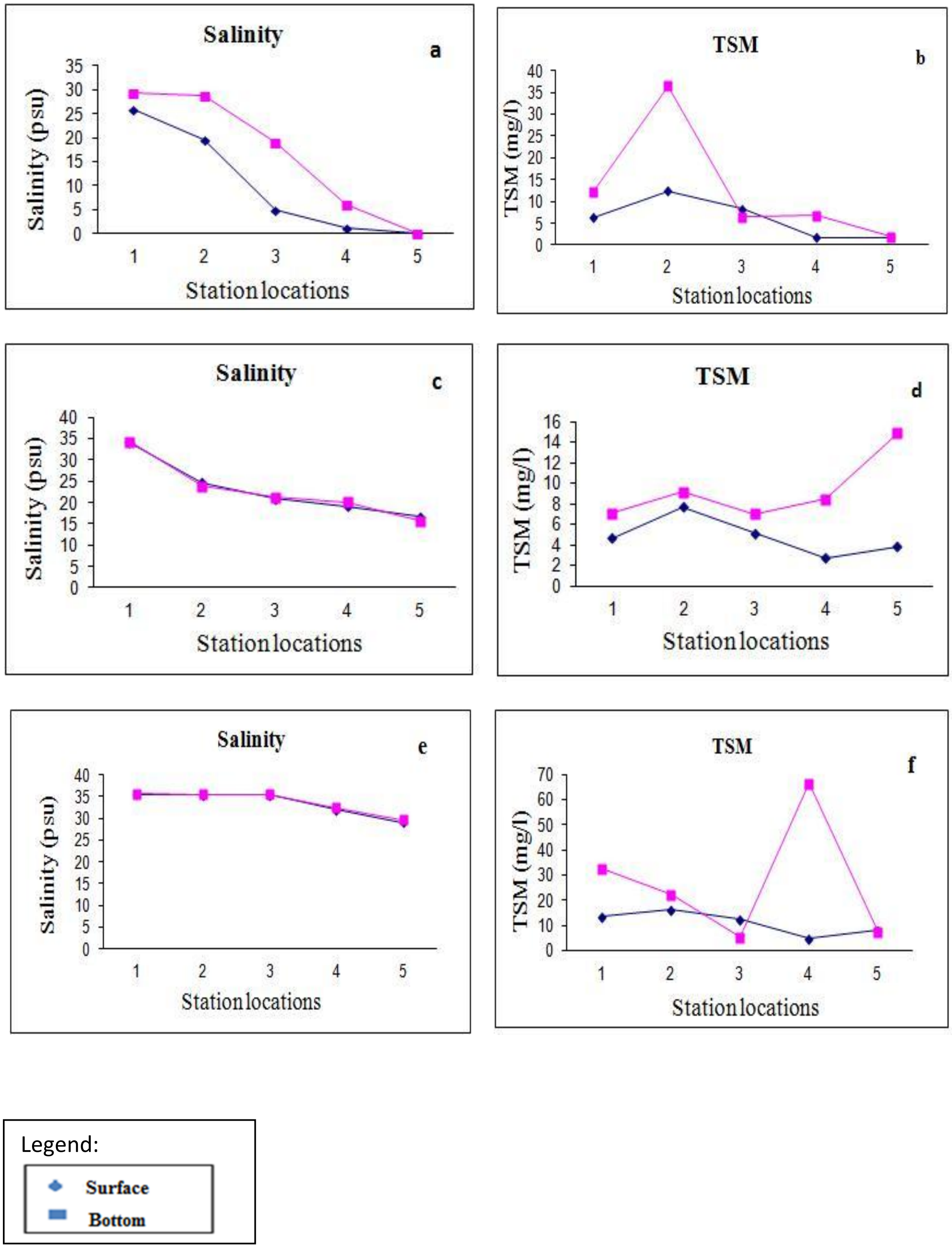
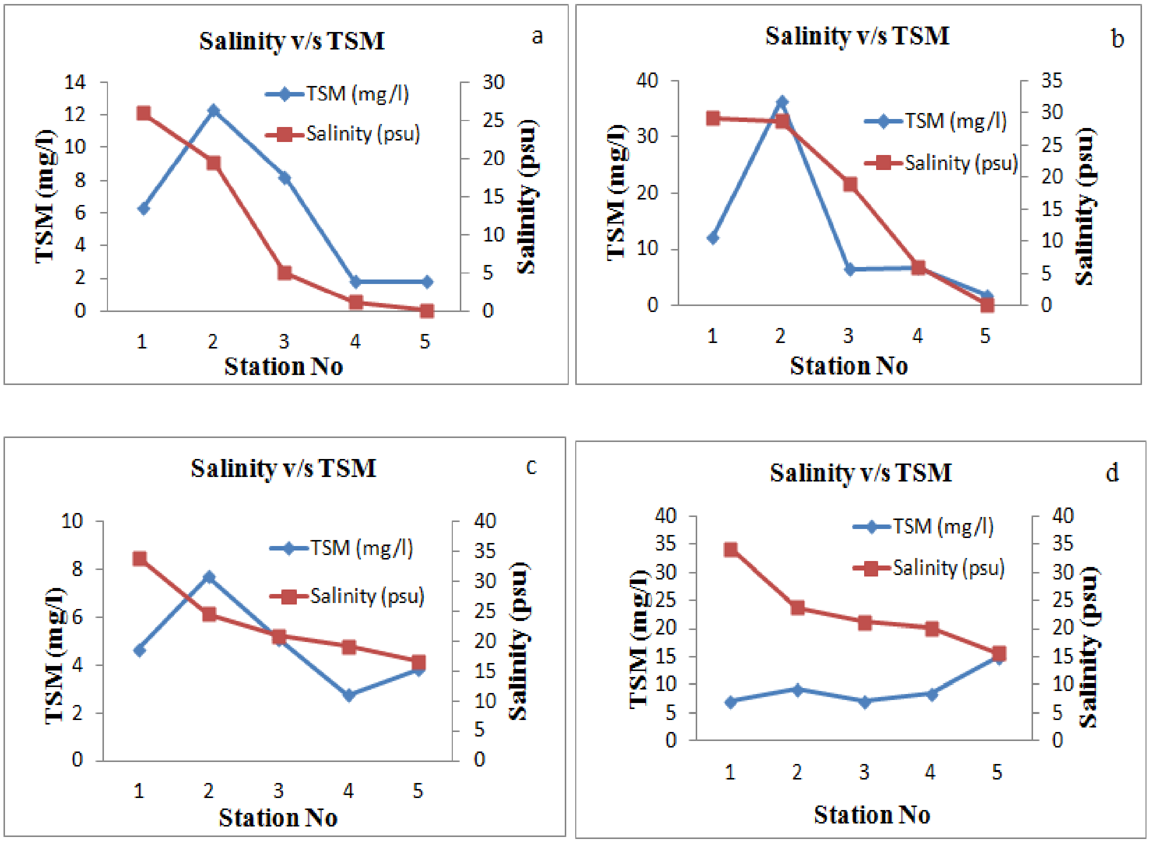

Salinity
a
Salinity (psu)
35
30
25
20
15
10
5
0
1
2
3
4
5
Station locations
TSM
b
TSM (mg/l)
40
35
30
25
20
15
10
5
0
1
2
3
4
5
Station locations
Salinity
c
Salinity (psu)
40
35
30
25
20
15
10
5
0
1
2
3
4
5
Station locations
TSM
d
TSM (mg/l)
16
14
12
10
8
6
4
2
0
1
2
3
4
5
Station locations
Salinity
e
Salinity (psu)
40
35
30
25
20
15
10
5
0
1
2
3
4
5
Station locations
TSM
f
TSM (mg/l)
70
60
50
40
30
20
10
0
1
2
3
4
5
Station locations
Legend:
Surface
Bottom

Fig 3.1. Distribution of salinity and TSM profiles along the estuary from mouth (station 1) towards the head (station 5) in Monsoon (a, b), Post monsoon (c, d) and Pre monsoon (e, f)

Salinity and TSM show an increasing pattern in both surface and bottom waters during monsoon while moving from head to mouth (Fig 3.2 a & b) and also in surface waters during postmonsoon (Fig 3.2c) and premonsoon (Fig 3.2e) seasons.



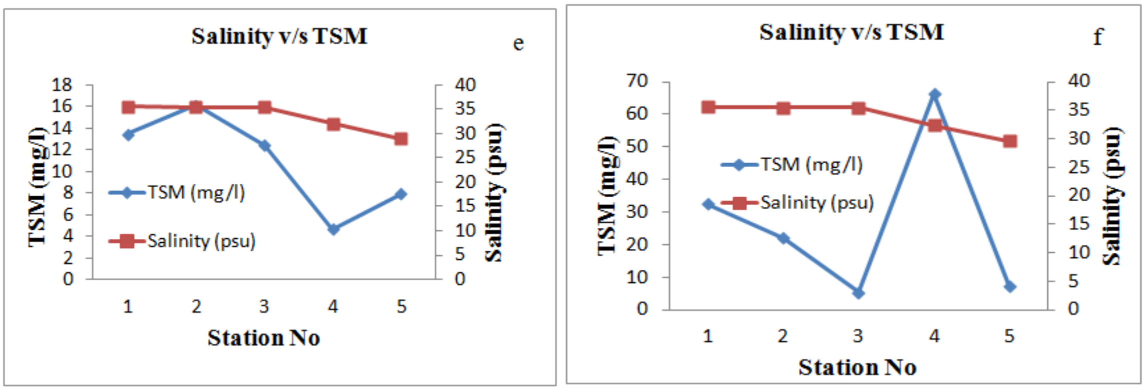


Fig 3.2. Plots of salinity and TSM in surface and bottom waters in monsoon (a, b); post monsoon (c, d) and pre monsoon (e, f) in Mandovi estuary

In general, at stations where salinity is the highest, the TSM values are comparatively low, especially near the mouth region of the estuary in both surface and bottom waters during monsoon, post monsoon and in surface waters during pre monsoon. The salt induced flocculation phenomenon is often proposed as a major mechanism influencing the deposition of the suspended matter near the mouth of rivers (Thill et al., 2001). Violent hydrodynamic conditions prevailing in the mouth region must be responsible for transporting the particulate matter towards upstream regions during flood tide.

In all the three seasons the surface waters and, during monsoon bottom waters, show high TSM concentration at station 2, supporting the transport of particulate matter from station 1 towards station 2. However, in bottom waters during both premonsoon (Fig 3.2f) and postmonsoon (Fig 3.2d), higher TSM concentration is observed towards upstream, which could be due to input from mining activities or due to resuspension of the bed sediments. Postma (1967) stated that the smaller size floccules are carried away by currents upstream where they tend to settle in calmer conditions thus increasing the TSM concentration near bottom in the upstream regions of the estuary.

3.4. Metals

Iron (Fe): Fe concentration ranges from 3.25 ppm to 18.25 ppm (Avg. 11.77 ppm) and from 5.00 ppm to 53.00 ppm (Avg. 18.18 ppm) in surface and bottom waters respectively during monsoon. In general, an increasing trend is observed from upstream to station 2 and decrease towards the mouth in both surface and bottom waters (Fig 3.3a). Highest Fe concentration is obtained at station 2 in bottom waters which coincides with higher TSM.

During postmonsoon, Fe ranges from 1.00 ppm to 11.00 ppm (Avg. 5.58 ppm) and 5.00 mg/l – 30.00 mg/l (Avg. 13.58 ppm) in surface and bottom waters respectively. Higher concentration of Fe is observed at upstream end in both surface (station 3) and bottom (station 5) waters (Fig 3.3b).

During premonsoon particulate Fe ranges from 1.25 ppm to 12.00 ppm (Avg. 3.95 ppm) and from 1.50 ppm to 34.00 ppm (Avg. 11.45 ppm) in surface and bottom waters respectively. In surface waters similar values are seen at station 5 and 4 followed by a decreasing trend up to station 2 but with higher value at station 1 (Fig 3.3c). In bottom waters concentration of particulate Fe shows a decrease from station 5 to 4 followed by an increasing trend towards the mouth of the estuary. On an average in both surface and bottom waters the concentration of particulate Fe is higher in monsoon followed by postmonsoon and premonsoon. Further, bottom waters show a higher particulate Fe as compared to surface waters.

Manganese (Mn): Mn concentration ranges from 0.50 ppm to 3.00 ppm (Avg. 1.39 ppm) and from 0.50 ppm to 6.25 ppm (Avg. 1.88 ppm) in surface and bottom waters respectively during monsoon. Distribution of Mn is similar to that of Fe in both surface and bottom TSM during this season (Fig 3.3d).

Mn concentration ranges from 0.02 ppm to 2.00 ppm (Avg. 1.06 ppm) and from 0.25 ppm to 5.50 ppm (Avg. 2.18 ppm) in surface and bottom waters respectively during postmonsoon season. Distribution pattern of Mn remains almost the same as that of Fe except for a few minor variations at station 1 and 5 (Fig 3.3e). Except for station 3, bottom water TSM maintained higher concentration than surface waters.

During premonsoon particulate Mn ranges from 1.25 ppm to 11.75 ppm (Avg. 3.95 ppm) in surface waters whereas, in bottom waters particulate Mn shows a range between 1.50 ppm to 34.00 ppm (Avg.

11.45 ppm). Largely decreasing trend is observed from station 5 towards the mouth of the estuary in surface waters except at station 1 (Fig 3.3f). In bottom waters, particulate Mn shows a decreasing trend from station 5 to 4 followed by an increase downstream. On an average higher particulate Mn is observed in premonsoon season followed by monsoon and postmonsoon in both surface and bottom waters. High concentrations of particulate Mn during premonsoon season must have resulted from enhanced oxidation at higher water temperatures (Morris et al., 1982; Cossa, 1990; Turner, 1999).

Zinc (Zn): Zn value varies from 0.05 ppm to 0.20 ppm (Avg. 0.11 ppm) and from 0.03 ppm to 0.40 ppm (Avg. 0.13 ppm) in surface and bottom waters respectively during monsoon. In surface waters particulate Zn shows an increasing trend from station 5 to station 3 followed by decrease at station 2 before showing a slight increase at station 1, whereas in bottom waters particulate Zn shows an increasing trend from station 5 to station 2 (Fig 3.3g) and lower value at the mouth, which is similar to that of Fe and Mn.

The particulate Zn value varies from 0.01 ppm to 0.1 ppm (Avg. 0.06 ppm) and from 0.04 ppm to 0.10 ppm (Avg. 0.076 ppm) in surface and bottom waters respectively during postmonsoon. In surface waters, particulate Zn shows an increase from station 5 to station 4 followed by a decrease up to station 2 and increase towards the mouth (Fig 3.3h). In bottom waters, decreasing trend is seen between stations 5 and station 3 followed by an increase up to station 2 and decrease from station 2 to station1.

During premonsoon particulate Zn ranges from 0.04 ppm to 0.14 ppm (Avg. 0.084 ppm) in surface waters and in bottom waters it ranges from 0.05 ppm to 0.13 ppm (Avg. 0.085 ppm). In surface waters an increasing trend is seen between station 5 to station 4 followed by decrease between station 4 and station 3 and also from station 2 to station 1. TSM value decreases from station 5 to station 4 followed by an increase between station 4 and station 2 (Fig 3.3i) in bottom waters. On an average higher concentration is observed in monsoon followed by premonsoon and postmonsoon in both surface and bottom waters.

Nickel (Ni): The concentration of particulate Ni varies from 0.01 ppm to 0.03 ppm (Avg. 0.019 ppm) and ranges from 0.01 ppm to 0.06 ppm (Avg. 0.025 ppm) in surface and bottom waters respectively during monsoon. Distribution pattern of Ni is similar to that of Fe and Mn in both surface and bottom waters (Fig 3.3j).

The particulate Ni value ranges from 0.008 ppm to 0.02 ppm (Avg. 0.017 ppm) and from 0.02 ppm to 0.04 ppm (Avg. 0.028 ppm) in surface and bottom waters respectively during postmonsoon. In surface waters, Ni value shows an increasing trend from station 5 to station 3 followed by decrease up to station 2 and further towards the mouth it shows a slight increasing value (Fig 3.3k).

In bottom waters, increase is seen between stations 5 and station 4 followed by a decrease at station 3 and thereafter an increase at station 2 and then decrease towards the mouth. Except for station 3, bottom TSM maintained higher concentration than surface waters.

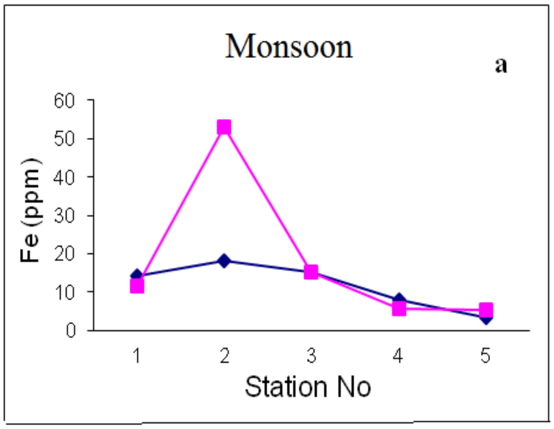
During premonsoon Ni ranges from 0.02 ppm to 0.04 ppm (Avg. 0.028 ppm) and from 0.025 ppm to 0.035 ppm (Avg. 0.033 ppm) in surface and bottom waters respectively. Generally increasing trend is observed from upstream to mouth of the estuary with some changes (Fig 3.3l). On an average higher concentration is observed in premonsoon followed by monsoon and postmonsoon in surface waters and in bottom waters higher concentration is seen in premonsoon followed by postmonsoon and monsoon.

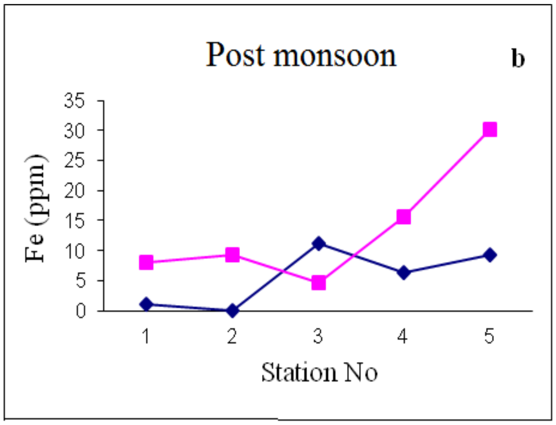
Cobalt (Co): The particulate Co ranges from 1.0 ppm to 4.25 ppm (Avg. 2.87 ppm) and from 1.45 ppm to 10.25 ppm (Avg. 3.86 ppm) in surface and bottom waters during monsoon respectively. Distribution pattern of Co is similar to that of Fe and Mn with highest value at station 2 for bottom waters (Fig 3.3m).

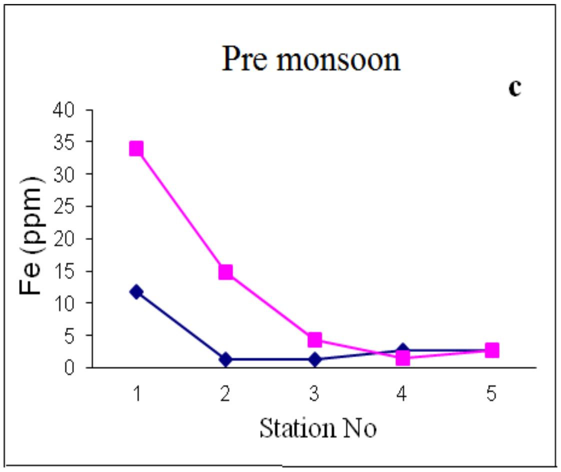
The particulate Co varies from 0.25 ppm to 3.25 ppm (Avg. 1.49 ppm) and from 1.75 ppm to 5.00 ppm (Avg. 2.93 ppm) in surface and bottom waters respectively during postmonsoon. Generally in surface waters Co shows an increasing trend from station 5 to station 3 followed by a decrease up to station 2

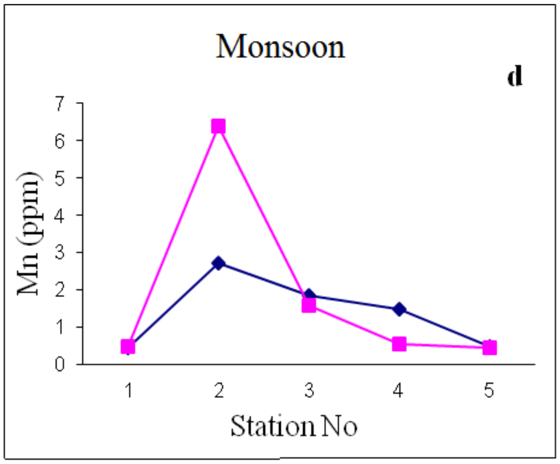
(Fig 3.3n) and increase towards mouth. In bottom waters, decrease is seen between station 5 and 3 followed by an increase at station 2 and then Co decreases towards mouth. Except for station 2 in bottom water, Co in TSM maintained higher concentration than surface waters.

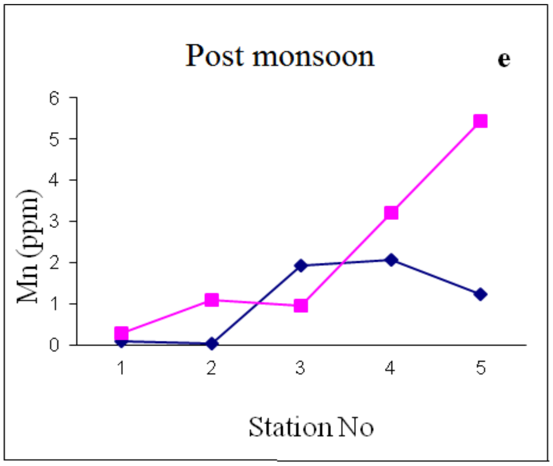
During premonsoon particulate Co ranges between 0.25 ppm to 3.15 ppm (Avg. 1.19 ppm) in surface waters whereas, in bottom particulate Co varies from 0.4 ppm to 7.65 ppm (Avg. 2.76 ppm). Increasing trend of surface particulate Co is observed between station 5 and station 4 followed by a decrease from station 4 and station 3 thereafter shows increase up to station 1 (Fig 3.3o). In bottom waters decrease in particulate Co is seen between stations 5 and station 4 followed by an increase towards the mouth of the estuary. On an average higher value of particulate Co is observed in monsoon followed by postmonsoon and premonsoon in case of surface and bottom waters.

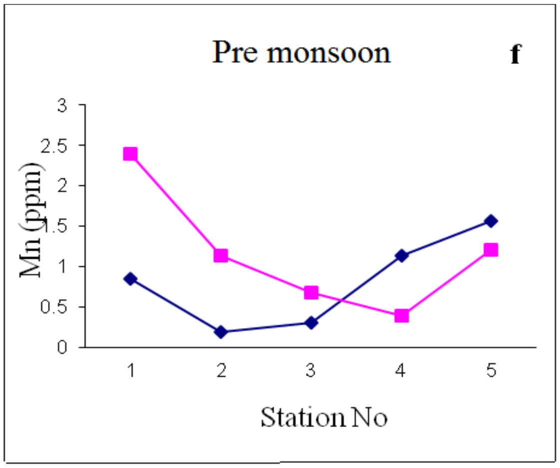


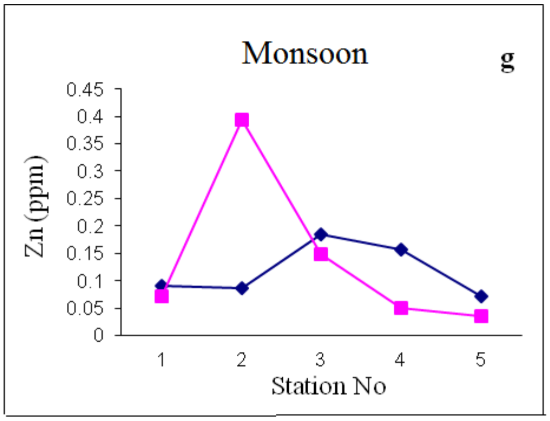


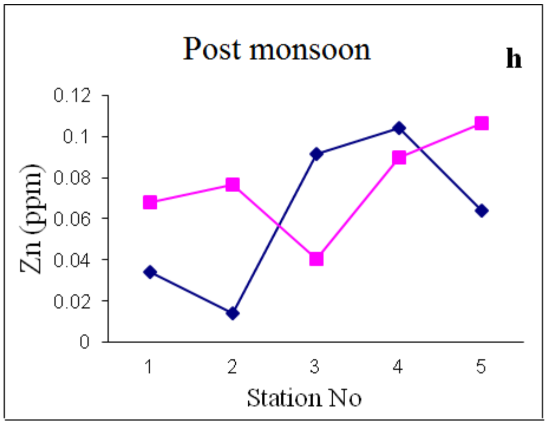

Pre monsoon
c
Fe (ppm)
40
35
30
25
20
15
10
5
0
1
2
3
4
5
Station No

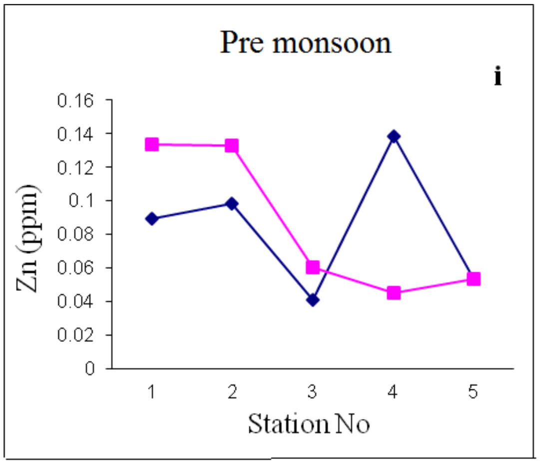

Monsoon
d
Mn (ppm)
7
6
5
4
3
2
1
0
1
2
3
4
5
Station No

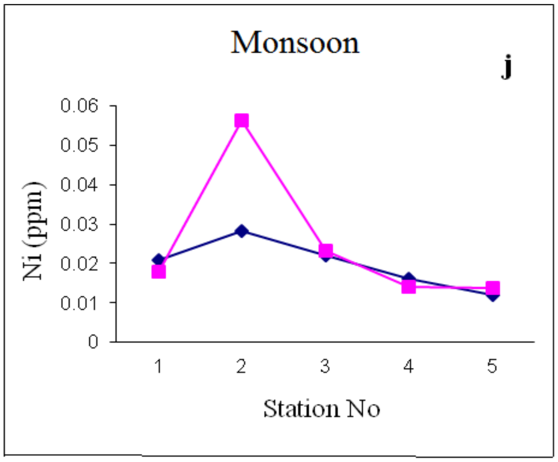

Post monsoon
e
Mn (ppm)
6
5
4
3
2
1
0
1
2
3
4
5
Station No

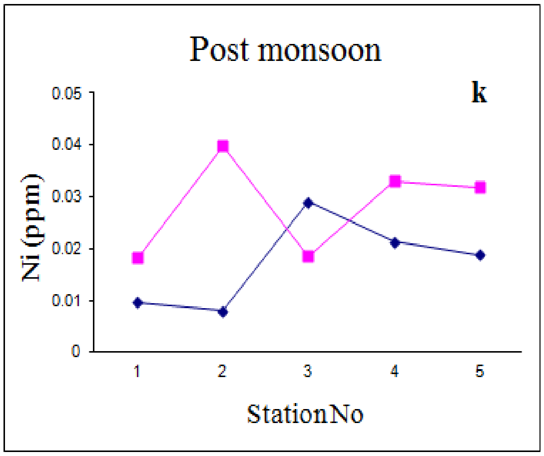

Pre monsoon
f
Mn (ppm)
3
2.5
2
1.5
1
0.5
0
1
2
3
4
5
Station No

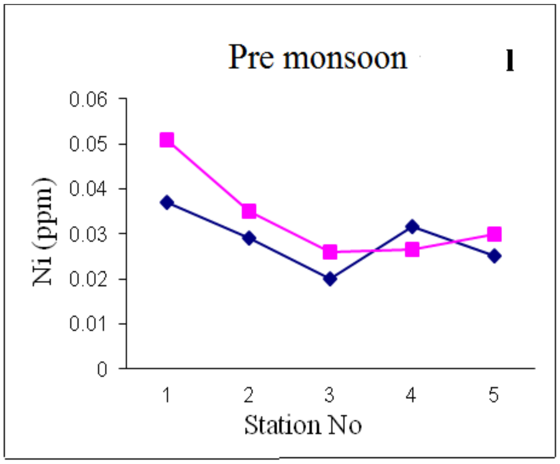

Monsoon
g
Zn (ppm)
0.45
0.4
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
1
2
3
4
5
Station No

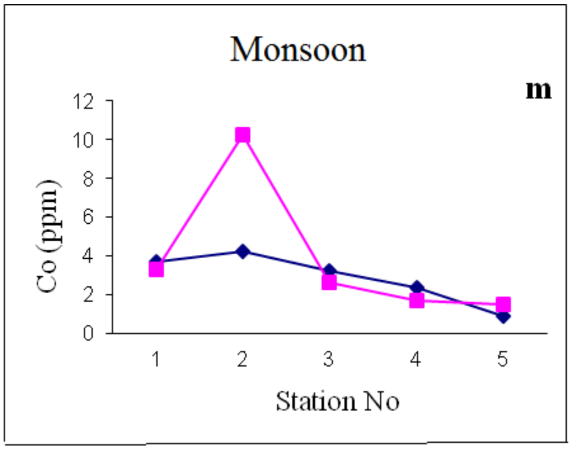

Post monsoon
h
Zn (ppm)
0.12
0.1
0.08
0.06
0.04
0.02
0
1
2
3
4
5
Station No

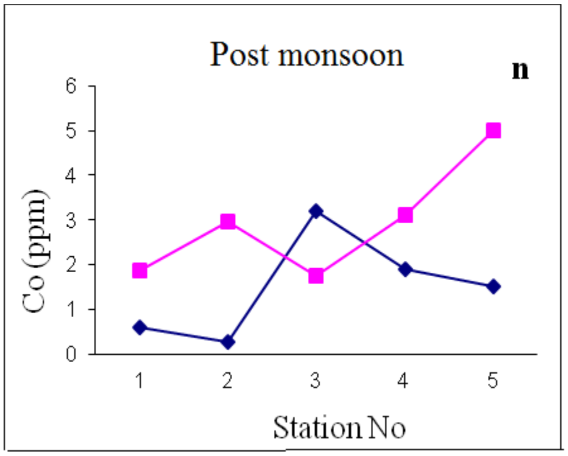

Pre monsoon
i
Zn (ppm)
0.16
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
1
2
3
4
5
Station No


Monsoon
j
Ni (ppm)
0.06
0.05
0.04
0.03
0.02
0.01
0
1
2
3
4
5
Station No


Post monsoon
k
Ni (ppm)
0.05
0.04
0.03
0.02
0.01
0
1
2
3
4
5
StationNo


Pre monsoon
l
Ni (ppm)
0.06
0.05
0.04
0.03
0.02
0.01
0
1
2
3
4
5
Station No


Monsoon
m
Co (ppm)
12
10
8
6
4
2
0
1
2
3
4
5
Station No


Post monsoon
n
Co (ppm)
6
5
4
3
2
1
0
1
2
3
4
5
Station No

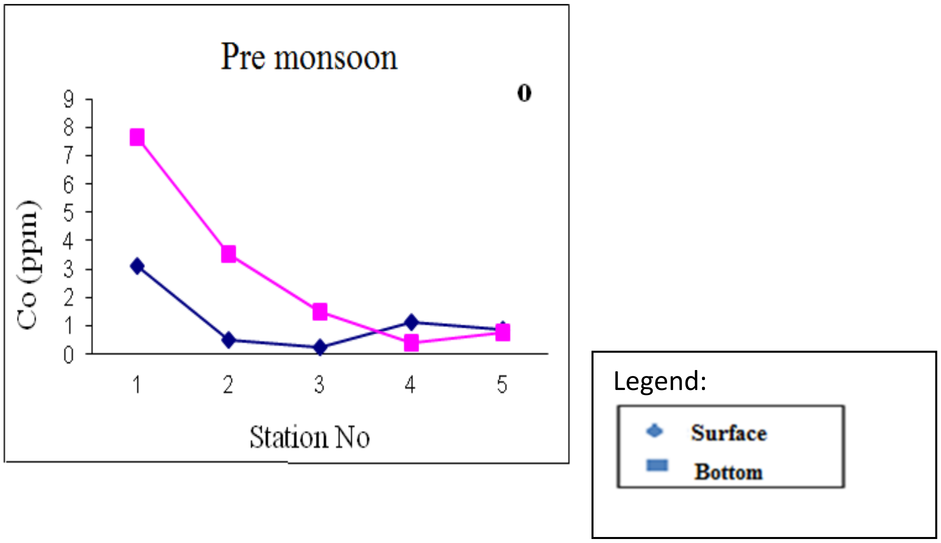


Fig 3.3. Distribution of particulate metals (Fe, Mn, Zn, Ni and Co) along the estuary from mouth (Station 1) towards the head (Station 5) in surface and bottom waters of Mandovi estuary during different seasons

3.5. TSM *Vs* Metals

Mandovi River receives considerable volume of freshwater during monsoon season, in the form of runoff from catchment area and monsoon precipitation. The salinity remains almost negligible during monsoon in the upstream regions (Station 5) and shows salt wedge feature in the downstream as can be seen from salinity distribution (Fig 3.1a). During monsoon season, Salinity and TSM show an increasing values as moved from head to mouth region in both surface and bottom waters with highest values obtained for station 2 (Fig 3.2a & b). All the elements studied also show higher concentration during monsoon at station 2 especially in bottom waters. The terrigenous particles carried by fresh water during monsoon when come in contact with high saline waters near the mouth, due to the influence of saline waters results in the double layer dynamics at the particle surface and, hence, increase the probability of flocculation and the settling velocity of the particles (Krone, 1962). Hence responsible to increase suspended particulate in bottom waters. Double layer theory implies that in waters of lower salinity, lesser ionic strength, a diffuse electrical double layer exists and the colloidal particles repel each other allowing conditions for the formation of stable suspension (Drever, 1997). An increase in salinity contributes to a constriction of the double layer and van der Waals forces dominate over repulsive forces creating agglomerations known as flocs. The important factors that influence these effects in nature are the salinity, the composition and concentration of the suspended matter, and the agitation of the water. Increasing salinity reduces the repulsion and thereby enhances the cohesion between particles. Increasing agitation of the water promotes flocculation by increasing the rate of collision. Two main factors thought to be important for the formation of a floc upon collision between two particles are: the organic coating of the particles (Van Leussen, 1994), and salinity of the surrounding water influencing the double layer dynamics. Generally elements undergoing flocculation at salinities ranging from 5 to 12 psu is well known. At higher salinity levels organic coating on particles might facilitate adsorption of elements resulting in higher suspended matter at station 2.

Alternatively, as stated earlier, violent hydrodynamic conditions prevailing in the mouth region must be responsible for transporting the particulate matter towards upstream regions during flood tide. The distribution of particulate metal profiles in Mandovi estuary with higher concentration at station 2 during the monsoon season especially in bottom waters suggests the addition of material from resuspension and remobilization from bottom sediments. The low metal concentrations in TSM toward station 1 are attributable to the mixing of fluvial suspended particles with resuspended sediments (Shynu et al., 2012). Dissolved Fe released from sediments can rapidly get oxidized in the overlying waters, producing Fe oxide coatings. This coating of Fe might also facilitate adsorption of trace metals.

Little difference in salinity is observed between surface and bottom waters during post and premonsoon. However, bottom water shows slightly higher salinity than the surface waters as mentioned earlier. The quantity of freshwater discharge from river Mandovi during postmonsoon and premonsoon period is negligible and the flow within the estuary during these seasons is regulated largely by tides of a semidiurnal nature. Thus the estuary remains partially mixed during post-monsoon and well mixed during pre-monsoon (Nayak and Bukhari, 1992). The higher TSM values observed in upstream regions during these seasons in the Mandovi estuary is contrary to the distribution of TSM in the adjacent Zuari estuary wherein decrease in TSM content from mouth to upstream during premonsoon and post-monsoon was reported by Nayak and Bukhari (1992). Higher TSM concentration observed towards the upstream especially in bottom waters during both premonsoon and postmonsoon could be due to resuspension of the bed sediments and/or input from mining activities. Biological processes, such as primary productivity and algal mats forming on the sediment surface during winter, may also contribute to TSM. However, as the area is under heavy mining activity, addition of material from mining cannot be ruled out in the upper estuary i.e. around station 5.

During postmonsoon season as the fresh water input to the estuary is reduced drastically, saline water enters the estuary during flood tide. There is considerable mixing between saline and fresh water in the middle estuarine region. Saline water moving upstream near the sediment bed surface is responsible for resuspension and remobilization of metal upstream. In the process, smaller size floccules are carried away by currents upstream where they tend to settle in calmer conditions thus increasing the TSM concentration near bottom in the upstream regions of the estuary (Postma, 1967). As stated earlier, dissolved Fe released from sediments can rapidly get oxidized in the overlying waters, producing Fe oxide coatings. Manganese, which is also a redox-sensitive element, is oxidized more slowly than Fe in natural systems (Sung and Morgan, 1980). The diagenetic mobilization of Fe and Mn in estuaries is widely recognized by Sundby et al. (1986); Owens et al. (1997); Zwolsman and Van Eck (1999). Most of the metals analyzed for the Mandovi estuary show higher concentration in the upper estuary during postmonsoon season. Based on geochemical considerations, higher trace metal concentrations can be explained on TSM in the bottom waters during postmonsoon with respect to higher Fe and Mn concentration.

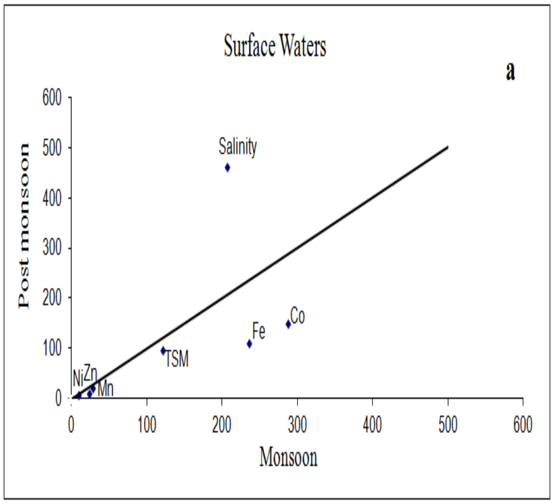
During the premonsoon season the Mandovi estuary is fully controlled and regulated by tidal influence and saline waters, and the estuary shows a well-mixed nature. During this season Fe, Ni, Co and, to some extent Zn, show higher concentration near the mouth. This indicates that the distribution of these metals in TSM is completely influenced and controlled by the highly dynamic environment near the mouth through resuspension and remobilization.

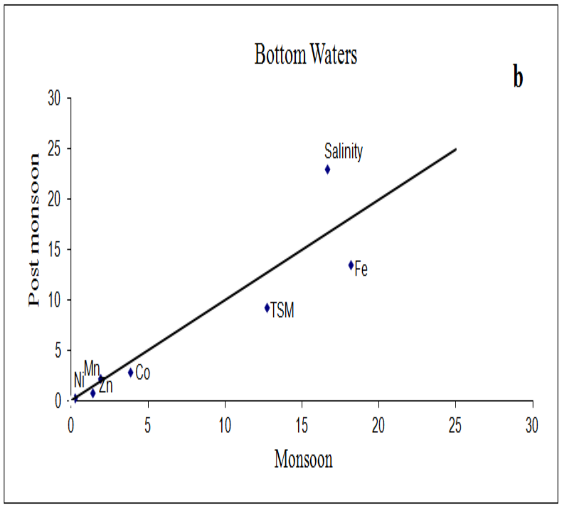
3.6. Inter- seasonal Variation

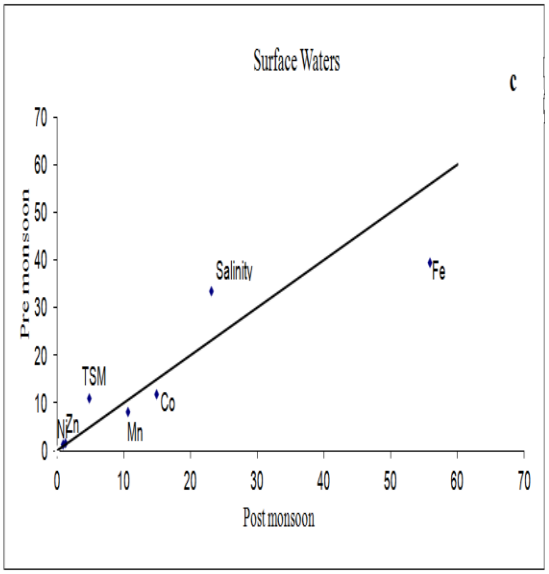
3.6. a. Isocon diagram

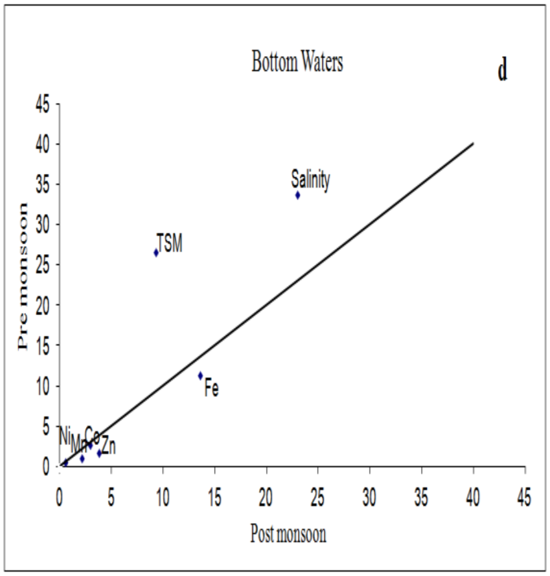
In order to understand the seasonal variations of different elements associated with TSM, data was plotted on isocon diagram. Isocon plots allow an easy visual comparison of average composition of each parameter studied in every season (Grant, 1986, Cundy et al., 1997, Rosales- Hoz et al., 2003). Average concentration of metals in surface and bottom TSM are plotted separately along with respective salinity and TSM concentration.

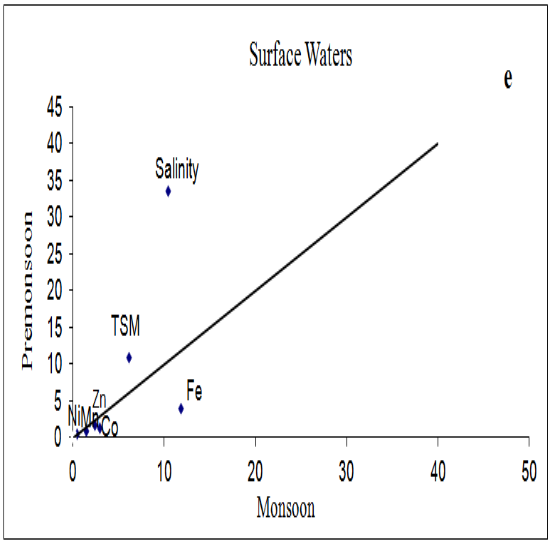
When the data of monsoon and postmonsoon are compared it is seen that Ni and Mn fall on or near the isocon line in both surface and bottom TSM indicating lesser inter-seasonal variation (Fig 3.4a & b). Salinity is more pronounced in postmonsoon as compared to monsoon in both surface and bottom waters. TSM along with metals such as Zn, Co and Fe deviate away from the isocon line and maintain higher concentration in monsoon in surface and bottom waters, indicating that these metals adsorb on to the surface of suspended matter during the monsoon season probably from terrigenous source. Comparison of data of postmonsoon and premonsoon (Fig 3.4c & d) indicates that Ni and Zn fall on or near the isocon line in surface waters and Ni, Co and Mn in bottom waters indicating not much variation between the parameters in the two seasons. In surface and bottom waters salinity and TSM concentrations are higher in premonsoon compared to postmonsoon whereas Fe, Mn and Co are more pronounced in surface and Fe and Zn are more pronounced in bottom waters during postmonsoon. This indicates that Fe and Mn hydroxides play a crucial role in distribution of Co in surface TSM. Higher concentration of Fe in bottom TSM indicates that particles sink slowly towards the bottom upon adsorption by Fe. When the data of monsoon and premonsoon (Fig 3.4e & f) are compared Ni, Mn, Zn and Co fall on or near the isocon line in both surface and bottom TSM indicating little seasonal variation. TSM along with salinity is more pronounced in premonsoon in both surface and bottom waters as compared to monsoon. High saline waters facilitate flocculation which allows the particles to move to deeper levels resulting in increase in concentration in bottom TSM.


Surface Waters
a
Post monsoon
Monsoon
0
100
200
300
400
500
600
Salinity
Co
Fe
TSM
Ni
Zn
Mn


Bottom Waters
b
Post monsoon
Monsoon
0
5
10
15
20
25
30
Salinity
Fe
TSM
Co
Ni
Mn
Zn


Surface Waters
c
Pre monsoon
Post monsoon
0
10
20
30
40
50
60
70
Salinity
Fe
TSM
Co
Mn
Ni
Zn


Bottom Waters
d
Pre monsoon
Post monsoon
0
5
10
15
20
25
30
35
40
45
Salinity
TSM
Fe
Ni
Mn
Co
Zn


Surface Waters
e
Premonsoon
Monsoon
0
5
10
15
20
25
30
35
40
45
50
Salinity
TSM
Fe
Ni
Mn
Zn
Co

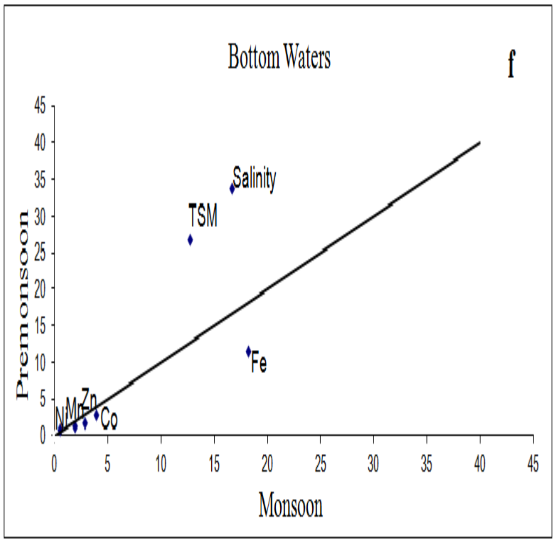

Bottom Waters
f
Premonsoon
Monsoon
0
5
10
15
20
25
30
35
40
45
Salinity
TSM
Fe
Ni
Mn
Zn
Co

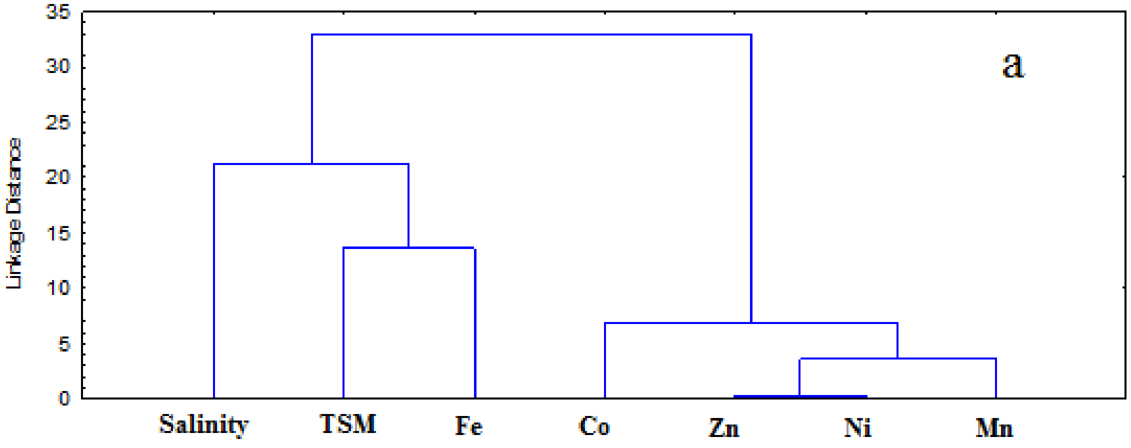
Fig 3.4. Isocon diagram (Grant, 1986) between seasons where, individual points represent average value of salinity, TSM and particulate metals (Fe, Mn, Zn, Ni, Co) in surface and bottom waters

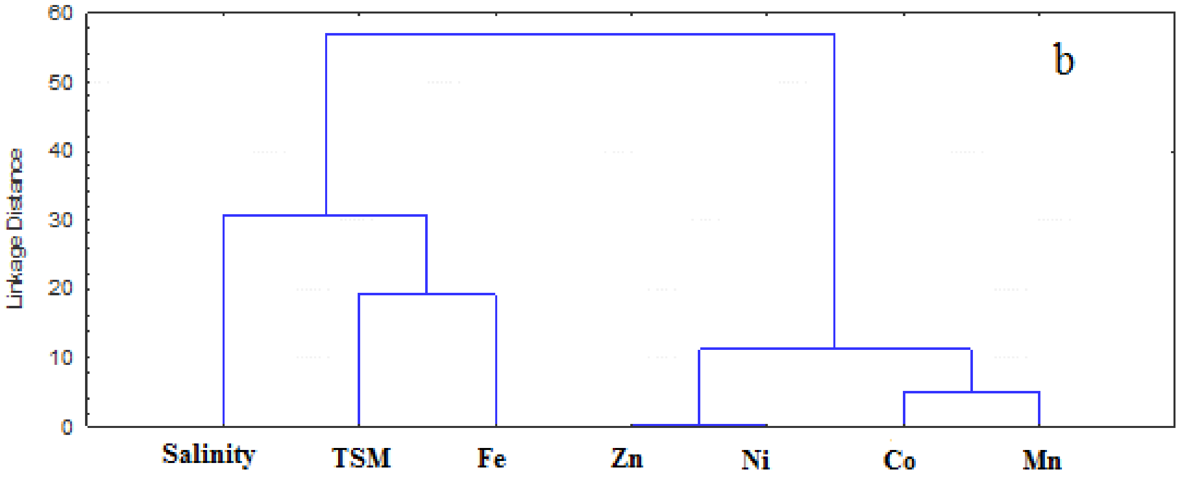
3.6. b. Cluster Analysis

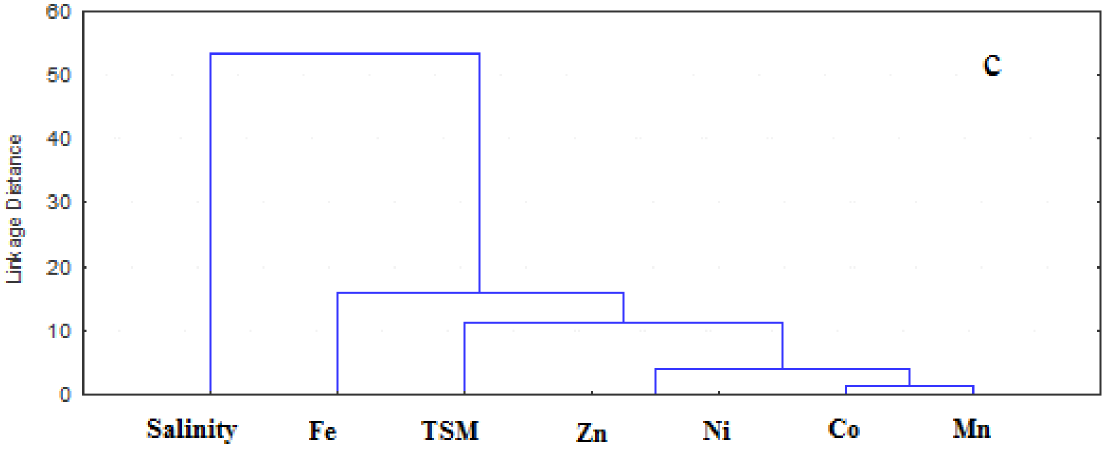
Further, cluster analysis was applied using complete linkage method to the data obtained, with Euclidean distances as criterion for forming clusters to understand the factors responsible for the association. During monsoon two clusters formed (Fig 3.5 a & b) out of which one includes salinity, TSM and Fe and the other comprises of Co, Zn, Ni and Mn in both surface and bottom waters. Martino et al. (2002) reported that Fe oxyhydroxide acts as a host phase on suspended matter for particulate trace metals. Zhaung et al. (2002) found that Fe coatings seem to be more effective in binding metals. In the present case Fe coating on TSM seems to act as adsorbing surface for other metals.

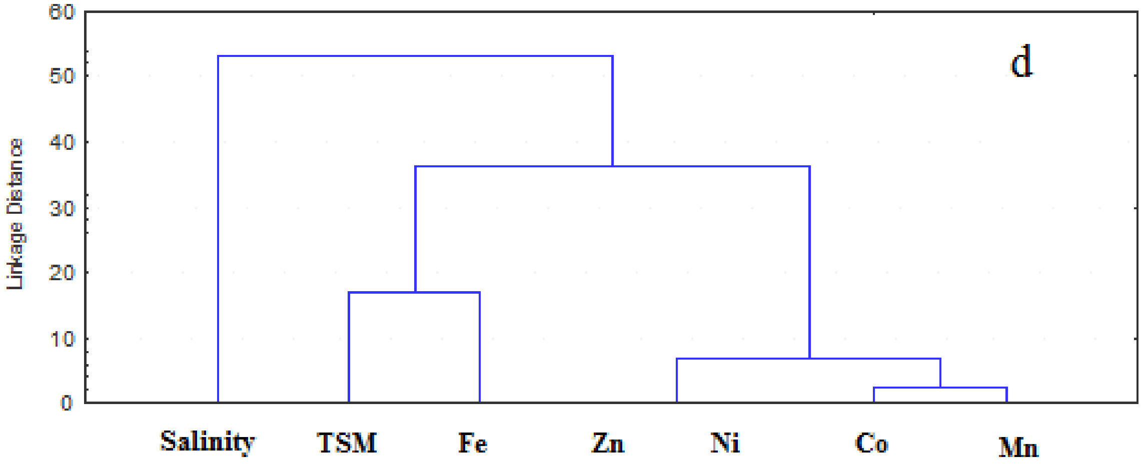
During postmonsoon in surface waters (Fig 3.5 c) salinity seems to have a direct control on particulate Fe. Distribution of other elements viz. Zn, Ni, Co and Mn seems to be not directly regulated by Fe. In the bottom waters (Fig 3.5 d) Fe and TSM form a single group. During premonsoon in bottom waters (Fig 3.5 f), salinity seems to control distribution of TSM and metals.

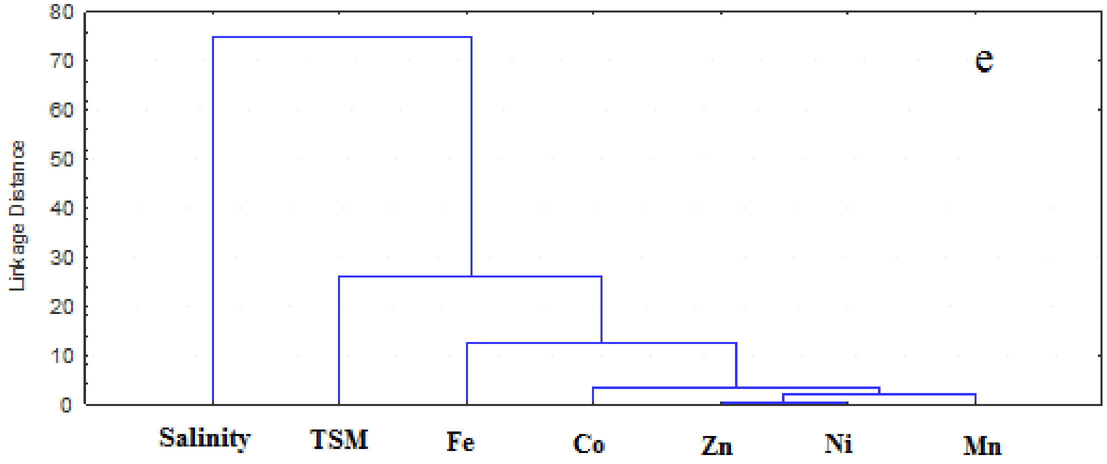
Cluster groups are similar during monsoon in both surface and bottom waters. Cluster groups during postmonsoon and premonsoon are similar in surface waters (Fig 6c & e) but are different in bottom waters indicating the role of salinity in the concentration of trace metals in TSM. Salinity and higher concentration of suspended matter are important factors that promote the aggregation of Fe and presence of Fe results in adsorption of other particulate metals.


a
Linkage Distance
35
30
25
20
15
10
5
0
Salinity
TSM
Fe
Co
Zn
Ni
Mn


b
Linkage Distance
60
50
40
30
20
10
0
Salinity
TSM
Fe
Zn
Ni
Co
Mn


c
Linkage Distance
0
10
20
30
40
50
60
Salinity
Fe
TSM
Zn
Ni
Co
Mn


d
Linkage Distance
0
10
20
30
40
50
60
Salinity
TSM
Fe
Zn
Ni
Co
Mn



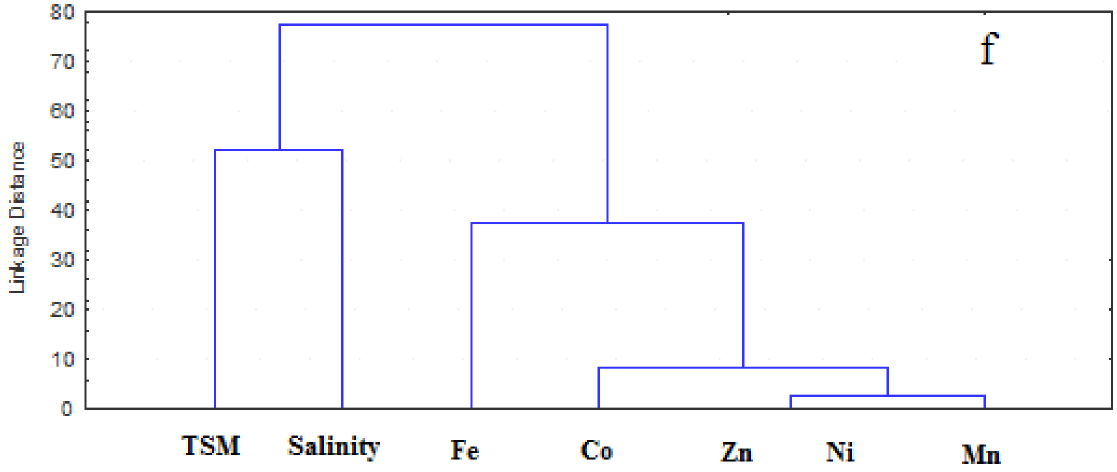


Fig 3.5. Dendograms of average values of variables - salinity, TSM, particulate metals (Fe, Mn, Zn, Ni and Co) in surface and bottom waters

3.6.c. Pearsons Correlation

Further, to understand the association of metals and possible factors responsible for the association Pearsons correlation was computed. In surface and bottom waters of the Mandovi estuary, Salinity and TSM shows a significant correlation during monsoon season. TSM exhibits significant correlation with almost all the particulate metals (Table 3.1a & 3.1b). In surface waters, Fe and Mn exhibits significant correlation with Ni and Co. In bottom waters, Fe and Mn show significant association with most of the metals. Significant association of particulate Fe and Mn with TSM and other particulate metals (Ni, Zn and Co) suggests their association with aluminosilicate and MnO_2 phases produced during weathering of rocks. Significant positive correlation of Ni, Zn and Co with TSM, Fe and Mn in bottom waters during monsoon suggests the high adsorptive capacities of clays (dominated by kaolinite, gibbsite and goethite - Kessarkar et al., 2010) and low degree of crystallinity of Fe, Mn-colloids and hydroxides (Chao, 1984; Waren and Zimmerman, 1994; Zhou et al., 2003).

During postmonsoon in surface waters, TSM and salinity shows negative correlation with most of the elements (Table 3.2a). Negative correlation obtained between TSM concentration and metals especially during this season can be due to desorption of metals in high salinity region. Also, low metal content in the high TSM can be due to the dominance of detrital fractions which are generally depleted in metals and hence contribute more mass but less metals to TSM (Jain and Sharma, 2001). However, Mn and Fe show significant correlation with Ni, Zn and Co.

Salinity exhibits negative correlation (Table 3.2b) in bottom waters during postmonsoon season. During post monsoon, the flow is primarily tidal and intrusion of salt water takes place gradually several kilometers upstream of the river (Shetye et al., 2007). It is likely that the salt water intrusion suppressed turbulence and also effectively removed fine grained particles in the estuary leading to very low concentrations. Saline water intrusion suppresses turbulence and thus probably results in not much of significant correlation between TSM and salinity. However, TSM shows significant correlation with Mn, Fe, Zn and Co (Table 3.2b) in bottom waters. Bottom water TSM generally shows significant correlation with metal concentrations than surface waters which is also noted in monsoon season (Table 3.1b). Several factors such as mixing of riverine suspended matter and particles of marine origin (Nolting et al., 1990); resuspension of sediments (Feely et al., 1986); flocculation of colloidal material (Turner and

Millward, 2002); production of organic matter and industrial and urban waste water discharges (Nolting et al., 1999) contribute to the distribution of elements within an estuary. Fe and Mn also show positive correlation with Zn and Co, indicating that either hydroxides of Fe or Mn or both must be also controlling the distribution of other elements. Inter-elemental association is observed in the case of Ni with both Zn and Co. Zn shows positive correlation with Co in both surface and bottom waters. High inter-elemental relationship among trace metals suggests a common source.

Monsoon

Table 3.1a. Pearsons correlation in surface waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	**0.65**	1.00					
Ni (ppm)	**0.97**	**0.76**	1.00				
Zn (ppm)	0.17	0.31	0.06	1.00			
Co (ppm)	**0.98**	**0.60**	**0.95**	0.12	1.00		
SALINITY	**0.72**	0.09	**0.68**	-0.41	**0.79**	1.00	
TSM	**0.93**	**0.73**	**0.97**	-0.02	**0.86**	**0.63**	1.00

Table 3.1b. Pearsons correlation in bottom waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	**0.99**	1.00					
Ni (ppm)	**1.00**	1.00	1.00				
Zn (ppm)	**0.99**	**0.99**	1.00	1.00			
Co (ppm)	**0.99**	**0.97**	**0.99**	**0.97**	1.00		
SALINITY	**0.64**	**0.55**	**0.62**	**0.63**	**0.67**	1.00	
TSM	**0.97**	**0.95**	**0.96**	**0.94**	**0.99**	**0.70**	1.00

Post monsoon

Table 3.2a. Pearsons correlation in surface waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	**0.85**	1.00					
Ni (ppm)	**0.94**	**0.93**	1.00				
Zn (ppm)	**0.80**	**0.98**	**0.90**	1.00			
Co (ppm)	**0.90**	**0.89**	**0.99**	**0.86**	1.00		
SALINITY	-0.72	-0.73	-0.63	-0.61	-0.54	1.00	
TSM	-0.52	-0.67	-0.50	-0.77	-0.44	0.33	1.00

Table 3.2b. Pearsons correlation in bottom waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	**0.96**	1.00					
Ni (ppm)	0.42	0.43	1.00				
Zn (ppm)	**0.89**	**0.82**	**0.64**	1.00			
Co (ppm)	**0.97**	**0.93**	**0.61**	**0.90**	1.00		
SALINITY	-0.63	-0.80	-0.47	-0.42	-0.70	1.00	
TSM	**0.95**	**0.89**	0.47	**0.80**	**0.97**	-0.66	1.00

Pre monsoon

Table 3.3a. Pearsons correlation in surface waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	0.20	1.00					
Ni (ppm)	**0.77**	0.18	1.00				
Zn (ppm)	0.12	0.08	**0.69**	1.00			
Co (ppm)	**0.99**	0.26	**0.85**	0.26	1.00		
SALINITY	0.24	-0.90	0.17	0.02	0.17	1.00	
TSM	0.16	-0.78	0.03	-0.32	0.07	**0.77**	1.00

Table 3.3b. Pearsons correlation in bottom waters

Variables	Fe (ppm)	Mn (ppm)	Ni (ppm)	Zn (ppm)	Co (ppm)	SALINITY	TSM
Fe (ppm)	1.00						
Mn (ppm)	**0.92**	1.00					
Ni (ppm)	**0.98**	**0.96**	1.00				
Zn (ppm)	**0.87**	**0.73**	**0.82**	1.00			
Co (ppm)	**1.00**	**0.91**	**0.97**	**0.87**	1.00		
SALINITY	**0.59**	0.28	0.43	**0.67**	**0.63**	1.00	
TSM	0.03	-0.19	0.04	-0.09	-0.02	-0.03	1.00

During premonsoon in surface waters, salinity and TSM show significant correlation. No significant correlation is obtained in the case of Mn with other particulate elements as well as with salinity and TSM (Table 3.3a). Fe is significantly correlated with Ni and Co. The results show that Fe oxides help in scavenging of metals, while the role of Mn oxides is low in natural environments (Yu et al., 2006). In bottom waters, negative correlation is observed between salinity and suspended matter (Table 3.3b). Mn exhibits significant correlation with Fe, Ni, Zn and Co whereas Fe shows significant correlation with Ni, Zn and Co. Previous studies have found that Mn-oxide surfaces may be important for Zn adsorption (James, 1977; Muller and Sigg, 1990). Ni exhibits positive correlation with Zn and Co in both surface and bottom water. Zn is correlated with Co. Strong correlation between trace metals (Ni and Co) and Fe with

TSM suggests that Fe oxides act as important factor for binding both Ni and Co (Warren and Zimmerman, 1994). In bottom waters, Fe and Mn show significant correlation with all the metals. Metal oxides are responsible for adsorbing trace metals on to the surface of suspended matter and further Fe and Mn oxides influence trace metal partitioning onto the TSM (Ratasuk et al., 2003).

CHAPTER FOUR

SURFACE SEDIMENTS

(SEASONAL VARIATION)

4.1. Sediment Components

The abundance and distribution of sediment components namely sand, silt, clay, organic carbon and metals in different regions of the estuary during different seasons are discussed below.

4.1A. Mudflats

In the samples collected from lower estuary, sand shows higher concentration (~95 %) during non-monsoon seasons compared to monsoon (~92 %). Silt and organic carbon shows less seasonal fluctuations. Clay shows higher value during monsoon (Table 4A.1a) than non-monsoon seasons (Table 4A.1b and c). pH shows less variation with seasons. Among the metals, Fe shows not much difference in its concentration between monsoon and pre-monsoon seasons but shows slightly higher concentration in post-monsoon (Table 4A.2b). Mn shows higher value in pre-monsoon (Table 4A.2c) and Al during monsoon (Table 4A.2a) season. Among trace metals, Cu, Co, Zn and Pb shows higher concentration during monsoon (Fig 4A.1a) whereas, Cr shows higher value in post-monsoon season. Ni shows less variation between seasons. Most of metals show higher concentration in monsoon followed by pre-monsoon and post-monsoon season. Total heterotrophic counts shows higher values during pre-monsoon (Table 4A.3c) and monsoon (Table 4A.3a) compared to post-monsoon (Table 4A.3b) season. At lower middle estuary, higher finer sediments concentration (~95 %) is noted in all the three seasons. Clay proportion is higher in monsoon (Table 4A.1a) season which is compensated by lower silt concentration. However, concentration of mud (silt + clay) shows not much difference among seasons. Organic carbon concentration is found to be higher in all the seasons at this location with highest concentration noted in pre-monsoon (Table 4A.1c). Largely, acidic pH is noted at this location. Fe and Al are found to be higher in post-monsoon (Table 4A.2b) along with trace metals such as Cu, Co and Pb (Fig 4A.1b) whereas, Mn shows higher concentration in pre-monsoon (Table 4A.2c) along with Cr (Fig 4A.2c) and Zn in monsoon (Fig 4A.2a) season. Ni shows less variation with seasons. Total heterotrophic counts show higher values in post-monsoon (Table 4A.3b) and pre-monsoon (Table 4A.3b) compared to monsoon.

At the upper middle estuary higher finer sediment concentration (~90 %) is noted in all the seasons. However, relatively higher concentration of sand is observed compared to lower middle estuary especially in monsoon and post-monsoon seasons (Table 4A.1a). Higher silt concentration during post-monsoon (Table 4A.1b) and pre-monsoon (Table 4A.1c) is observed at this location compared to lower middle estuarine region. Higher organic carbon is noted in all the three seasons with highest value observed during post-monsoon. Largely, acidic pH is noted at this location. Among the metals Fe shows higher concentration in monsoon (Table 4A.2a) whereas, Mn and Al show high values during post-monsoon season (Table 4A.2b). Cu, Zn and Ni are found to be higher in monsoon (Fig 4A.1a) season and Cr and Pb in post-monsoon (Fig 4A.1a). Co shows less variation with respect to different seasons. The distribution of total heterotrophic counts is similar to that observed in lower middle estuary i.e. counts are found to be higher in post-monsoon (Table 4A.3b) and pre-monsoon (Table 4A.3b) seasons.

Table 4A.1. Distribution of sediment components within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

a) Monsoon			
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary
Sand (%)	92.02	4.81	6.19
Silt (%)	3.16	37.09	41.86
Clay (%)	3.98	58.09	51.93
Organic carbon (%)	0.19	2.01	2.60
pH	7.12	7.43	8.31

b) Post-monsoon			
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary
Sand (%)	95.30	2.74	9.55
Silt (%)	3.67	49.56	55.03
Clay (%)	1.01	47.69	35.40
Organic carbon (%)	0.33	2.36	2.67
pH	6.58	6.86	6.48

c) Pre-monsoon			
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary
Sand (%)	95.93	5.50	3.17
Silt (%)	3.50	49.17	55.82
Clay (%)	0.56	45.33	41
Organic carbon (%)	0.20	2.62	2.40

pH	6.89	6.81	6.49

Table 4A.2. Distribution of Fe, Mn and Al within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

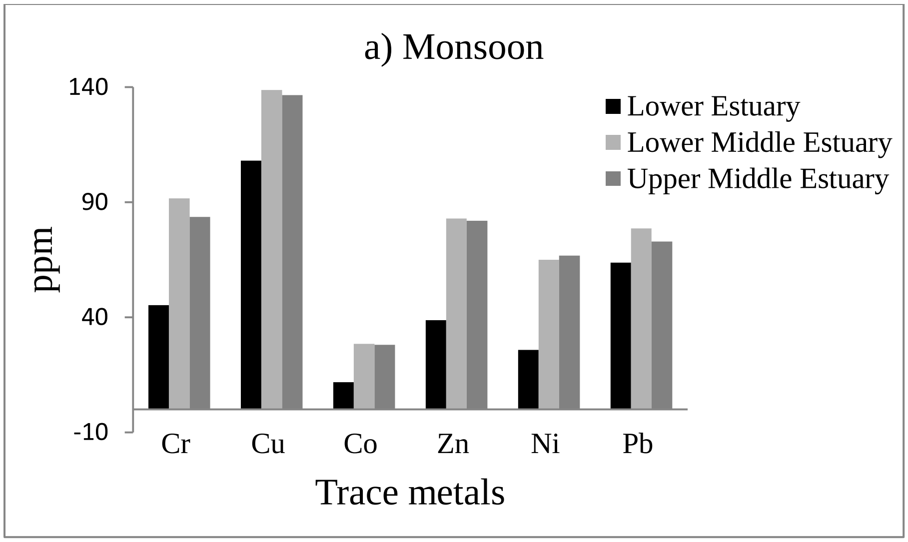
a) Monsoon			
Metals	Lower estuary	Lower middle estuary	Upper middle estuary
Fe (%)	1.67	5.99	7.70
Mn (ppm)	263.3	2585.7	1945
Al (%)	7.07	8.11	8.96

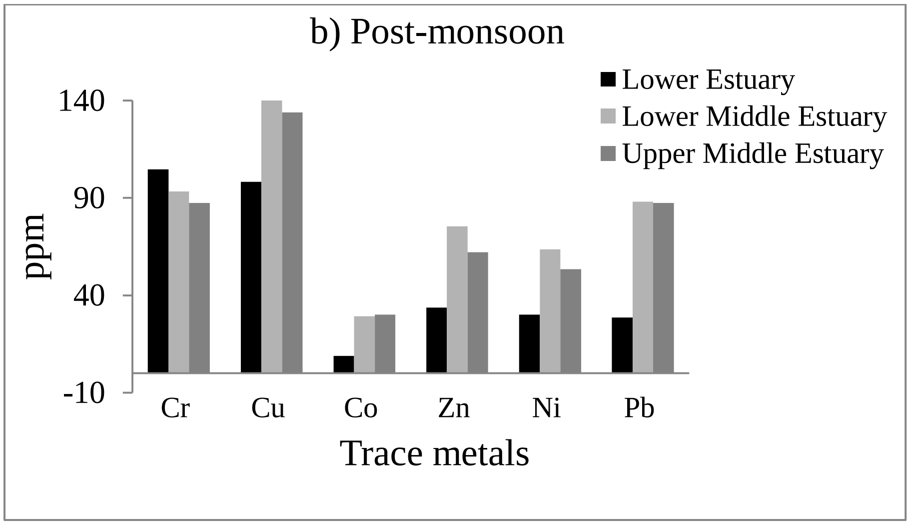
b) Post-monsoon			
Metals	Lower estuary	Lower middle estuary	Upper middle estuary
Fe (%)	1.79	6.25	6.48
Mn (ppm)	284.2	2731.6	2562.3
Al (%)	5.86	9.02	8.97

c) Pre-monsoon			
Metals	Lower estuary	Lower middle estuary	Upper middle estuary
Fe (%)	1.64	5.91	5.84
Mn (ppm)	335.3	2752.6	2266.1
Al (%)	6.26	8.21	7.45

Table 4A.3. Distribution of Total heterotrophic counts (CFU g^{-1} sediment) within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

THC			
Seasons	Lower estuary	Lower middle estuary	Upper middle estuary
Monsoon (a)	2.00E+04	2.00E+05	3.50E+05
Post-monsoon (b)	2.40E+02	3.50E+06	3.70E+06
Pre-monsoon (c)	2.60E+04	4.00E+06	3.90E+06





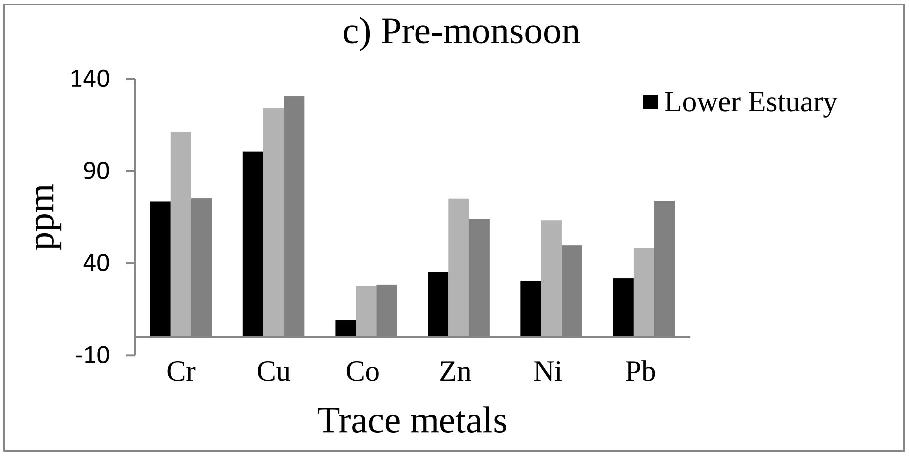


Fig 4A.1. Trace metal (Cr, Cu, Co, Zn, Ni and Pb) distribution within mudflats in lower, lower middle and upper middle regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

From the results detailed above, it is seen that higher concentration of coarser sediments (above 90 %) in lower estuary and higher concentration finer sediments (silt + clay) in middle estuary are observed in all the three seasons (Table 4A.1) in Mandovi estuary. Relatively higher wave activity at the lower estuary, irrespective of seasons, results from complex and high energy strong hydrodynamic conditions which facilitates deposition of coarser sediments carrying finer sediments to areas of lesser energy environments. Further, abrupt change in sand percentage from lower estuary to middle estuary suggests a transition from a relatively high-energy depositional environment to lower-energy environment (Fox et al., 1999). Higher total suspended matter (TSM) concentration in bottom waters obtained for the middle estuary (chapter 3) strongly supports deposition of higher finer sediments at this location. Slightly higher concentration of sand found in upper middle estuary than observed in lower middle estuary could be due to the influence of land drainage. It is important to note here that sample location of upper middle estuary is part of main estuarine channel whereas, lower middle estuary is from a sub channel of Mandovi estuary.

Addition of coarser material at upper middle estuary therefore may be river brought material from catchment area of Mandovi, which has large number of open cast mines. Further, loadings of large number of barges do takes place in upstream of Mandovi estuary. Slightly less sand percentage noted during monsoon at lower estuary is possibly because of the river brought mixed sediments. From the results, it is observed that organic carbon is found to be less in lower estuary. Organic carbon is generally associated with finer sediments (Poppe and Commeau, 1992) as C-org /surface is higher for finer sediments than coarser sediments like sand particles (Wakeham, 2002). Due to this there is higher organic matter in middle estuary. Further, variation in organic carbon content is noted between lower middle and upper middle estuary with respect to seasons. Organic matter is found to be highest in monsoon and post-monsoon in upper middle estuary whereas; during pre-monsoon in lower middle estuary. Large amount of organic debris brought by land runoff (Krupadam and Anjaneyulu, 2002) or addition of organic matter through agricultural practices in the catchment area might have resulted in higher concentration of organic carbon in monsoon and post-monsoon seasons at upper middle estuary. Highest organic matter noted in lower middle estuary in pre-monsoon season, may be due to accumulation and lower degradation rate of organic matter, which controls concentration of organic carbon in sediments during this season. Higher organic carbon concentration in surface sediments is attributed to influx of organic matter from flora and fauna communities from surrounding (Man et al., 2004) mangrove environment. During pre-monsoon, the consequence of more algae lysis, microbial biomass decomposition (Dellwig et al., 2007a) results in production of sticky carbohydrate which colligate suspended organic and inorganic particles from the water column thereby forming larger aggregates (Chen et al., 2005). Organic-rich aggregates are later incorporated into the surface sediments by tidal forces (Billerbeck et al., 2006). Higher organic matters in surface sediments indicate adsorption and incorporation of organic materials from overlying water column with accumulation of fine grained inorganic materials (Janaki- Raman et al., 2007).

As seen from the results, lower estuary shows higher concentration of metals viz Al, Cu, Co, Zn and Pb in monsoon followed by pre-monsoon and post-monsoon season. In lower middle estuary, higher concentration of many metals viz. Al, Fe, Cu, Co and Pb is observed in post-monsoon and pre-monsoon months whereas, in upper middle estuary, higher metal concentration is noted in monsoon and post-monsoon seasons. Higher metal concentration in monsoon season obtained for lower estuary and upper middle estuary, which was collected from the main channel, might be probably due to high fresh water runoff which must have brought weathered natural products along with anthropogenic material from the catchment area. During monsoon, the mixing of fresh and saline waters results in formation of floccules either by dissolved organic matter or salts. The flocs scavenge other metals from the water column while settling, thereby increasing the metal deposition. The larger aggregated particles formed tend to have a higher settling velocity and gets deposited more rapidly than smaller discrete particles (Palanques et al., 1995). However, the lower estuary has high bottom stress due to the local high hydrodynamic conditions and therefore the sediment is continuously reworked (Loomb, 2001). Higher metals concentrations along with Al in lower estuary indicate their presence in coarse grained particles of natural origin. Fe along with Cr is high in post-monsoon and Mn is high in pre-monsoon indicating role of Fe and Mn oxides in redistribution of trace metals in surface sediments at this location. Higher concentration of some of metals during monsoon in upper middle estuary might be due to the material brought from land drainage which includes mining activities, agricultural practices in addition to natural weathering products. Aggregation of clay particles by marine organic matter is a widespread phenomenon, which is responsible for the rapid sinking of land-derived materials (Chamley, 1989). Differential discharge of untreated effluents originating from industries and agriculture as well as from domestic sewage along with Fe ore carrying barges, fishing and boating activities (Flory et al., 2012) must be also contributing to metals. Mixing of material from different sources during transportation, flocculation and differential settling processes are the main mechanisms for the concentration of metals (Griffin et al., 1968). Higher concentration of metals found in non-monsoon seasons might be regulated by factors like mixing of less fresh water with more saline water, physico-chemical and geo-chemical process variables. For example, behaviour of Mn with reference to salinity within the estuary is well established. Mn remains in solution phase in the lower salinity values up to 10 (Kerdijk and Salomons, 1981). Further as the salinity increases Mn slowly flocculates and finally gets precipitates (Balachandran et al., 2005) at salinity around 18 and gets incorporated in the surface sediments. During pre-monsoon salinity higher than 18 is available at this location (Chapter 3). Higher concentration of Mn in pre-monsoon observed in lower middle estuary therefore seems to be controlled by salinity. At lower middle estuary deposition of higher finer size sediments indicates presence of calmer hydrodynamic conditions. This in turn facilitates entrapment and accumulation of metals as metals are generally associated with smaller grain size particles (Martincic et al., 1990; Biksham et al., 1991). Higher concentration of Al with metals Fe, Cu, Co, Pb in post-monsoon season indicates role of clay and clay minerals (hydrous Al- silicates) in trapping the metals. Further, Fe-oxide must also be regulating the distribution of trace metals in surface sediments. Metals viz. Ni and Co show less variation with seasons i.e. Ni in lower middle and Co in upper middle. This indicates that Ni and Co might be strongly bound with the crystal structure of minerals, thus dilution due to run off brought material has not largely affected Ni concentrations in sediment (Tavakoly Sany et al., 2011).

In summary, results obtained indicated that Al, Fe and Mn are found to be higher in middle estuary compared to lower estuary (Table 4A.2) indicating their association with finer sediments and organic matter due to larger surface area of smaller particles (Mikulic et al., 2008). Further, Al is part of clay mineral chemistry. Cr, Cu, Co, Ni, Zn and Pb are also found to be higher in middle estuary. It is inferred from the above observations that grain size of sediment play crucial role in governing the metal concentrations in the sediments of mudflats in Mandovi estuary. Decrease of metal concentration in lower estuary may be due to dilution by coarse grained sediments which generally constitute quartz and feldspar (Horowitz, 1991). The sediments in middle estuary contain higher organic matter. The metals concentration is also high in these sediments indicating the role played by organic matter in their distribution.

Total heterotrophic counts are found to be higher in pre-monsoon season in lower estuary. In lower middle and upper middle estuary, the counts are higher in post-monsoon and pre-monsoon season compared to monsoon. During monsoon, the higher fresh water influx must have diluted the microbial populations and also partly carried to offshore regions. Higher organic matter concentration must have acted as important substrate for increasing total heterotrophic counts during non-monsoon months. This could also imply that the substrate dependence or coupling of the total heterotrophic bacteria with metals will occur during the non- monsoon seasons. Alternatively, counts are diluted by sediments brought from land during monsoon season. Heterotrophic counts within sediments depends on various parameters namely temperature, salinity, abundance of organic nutrients (Fong et al., 1993) and metals act as essential micronutrients for life processes within certain limits (Bruins et al., 2000). However, it has been recognized that bacterial population may be considerably modified by interactions with biotic factors. The distributions of total heterotrophic counts (THC) are higher in middle estuary as compared to the counts at lower estuary (Table 4A.3). The distribution of heterotrophic counts can therefore be related to the nature and size of the sediment. Bacterial abundance can also be related to the organic matter concentration which is comparatively higher in middle estuary as compared to lower estuary (Bianchi and Garcin, 1993). THC values are also higher during non-monsoon seasons than monsoon season. Riemann et al. (2000) reported that, seasonal variation could result from community succession with inherently different metabolic capabilities.

4.1B. Mangroves

In the sample collected from lower estuary, sand is relatively high (32.80%) in monsoon (Table 4B.1a) compared to post monsoon (Table 4B.1b) and pre-monsoon (Table 4B.1c) seasons. Silt is higher in pre-monsoon and clay in post-monsoon compared to other seasons. The concentration of organic carbon is higher during pre-monsoon followed by post-monsoon and then monsoon. pH is acidic in all the three seasons (Table 4B.1). Fe and Al are found to be higher in monsoon (Table 4B.2a) season whereas, Mn is higher in pre-monsoon (Table 4B.2c). Among the trace metals, Zn is higher in monsoon (Fig 4B.1a) season and Cu, Co and Pb in post-monsoon (Fig

4B.1b) season. Cr and Ni are found to be higher in pre-monsoon (Fig 4B.1c) season. Total heterotrophic counts are higher during non-monsoon seasons especially in pre-monsoon (Table 4B.3c) season as compared to monsoon (Table 4B.3a) season.

At lower middle estuary, the sediments show less (<6.5 %) sand and higher finer sediments. Maximum sand content is noted during post-monsoon (Table 4B.1b) season. Although relatively silt is higher in monsoon season and clay is maximum in pre-monsoon season but when the data of both silt and clay is considered together it is noted that value of mud concentration is higher in pre-monsoon (98.2 %) season followed by monsoon (94.40 %) and post-monsoon (93.83 %) seasons. The organic carbon shows relatively lesser value during pre-monsoon (Table 4B.1c) with comparatively higher concentration in monsoon (Table 4B.1a) and post-monsoon (Table 4B.1b) seasons. pH shows lesser variations. Fe and Al are noted to be higher in monsoon (Table 4B.2a) season whereas, Mn is observed to be higher in post-monsoon (Table 4B.2b) season. Metals viz Cr, Cu, Zn and Ni are higher in monsoon season (Fig 4B.1a) whereas, Co and Pb are higher during pre-monsoon (Fig 4B.1c) season. In lower middle estuary, heterotrophic counts are higher in post-monsoon (Table 4B.3b) season.

Higher concentration of coarser sediments (21.95 %) is observed in monsoon season compared to post-monsoon and pre-monsoon season at the upper middle estuary. Silt is found to be maximum in pre-monsoon (Table 4B.1b) and clay in post-monsoon (Table 4B.1c) season. Organic carbon is noted to be maximum in monsoon (Table 4B.1a) season. Fe, Mn and Al concentration are maximum in monsoon (Table 4B.2a) season. Among trace elements, it is noted that Cu and Zn are higher in monsoon (Fig 4B.1a) season. Co and Pb are higher in post-monsoon (Fig 4B.1b) season and Cr in pre-monsoon (Fig 4B.1c) season with lesser variation noted in distribution of Ni in all the three seasons. Heterotrophic counts are observed to be higher in non–monsoon seasons i.e. pre-monsoon (Table 4B.3c) followed by post-monsoon (Table 4B.3b) and then monsoon (Table 4B.3a) season.

In the sample collected from upper estuary, distribution of sand fractions in different seasons shows less variation. The silt concentration is found to be highest in monsoon (Table 4B.1a) whereas, clay in pre-monsoon (Table 4B.1c) season. Overall, higher organic carbon is observed in non-monsoon seasons. pH shows less variation. Fe, Mn and Al are noted to be highest in monsoon (Table 4B.2a) season. Most of the metals namely Cu, Co, Ni, Zn are observed to be highest in monsoon (Fig 4B.1a) season with exception of Pb and Cr which are highest in post-monsoon (Fig 4B.1b) and pre-monsoon (Fig 4B.1c) respectively. Distribution of total heterotrophic counts shows similar trend to that observed in upper middle estuary i.e. heterotrophic counts are found to be maximum in pre-monsoon followed by post-monsoon and then monsoon.

Table 4B.1. Distribution of sediment components within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

a) Monsoon				
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Sand (%)	32.80	5.60	21.95	4.63
Silt (%)	39.47	52.29	47.43	92.30
Clay (%)	27.73	42.11	30.61	3.07
Organic carbon (%)	1.98	2.95	3.21	1.40
pH	5.92	5.89	6.77	7.40

b) Post-monsoon				
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Sand (%)	24.10	6.16	14.03	5.04
Silt (%)	45.09	45.16	42.63	69.69
Clay (%)	30.80	48.66	43.33	25.26
Organic carbon (%)	2.16	2.97	2.29	2.74
pH	6.54	6.40	6.86	6.54

c) Pre-monsoon				
Sediment parameters	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Sand (%)	25.07	1.80	14.73	4.73
Silt (%)	55.60	46.87	52.60	57.27
Clay (%)	19.33	51.33	32.67	38.00
Organic carbon (%)	2.29	2.54	2.95	2.24
pH	6.74	6.26	6.34	6.23

Table 4B.2. Distribution of Fe, Mn and Al within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

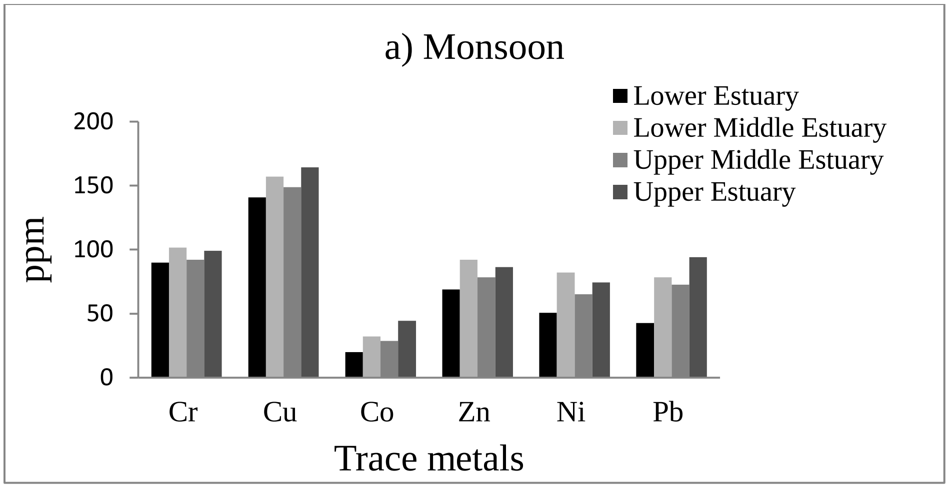
a) Monsoon				
Metals	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Fe (%)	6.1	7.1	8.3	9.7
Mn (ppm)	1141.3	2745.3	2435.6	3245.0
Al (%)	7.0	10.1	7.7	8.3

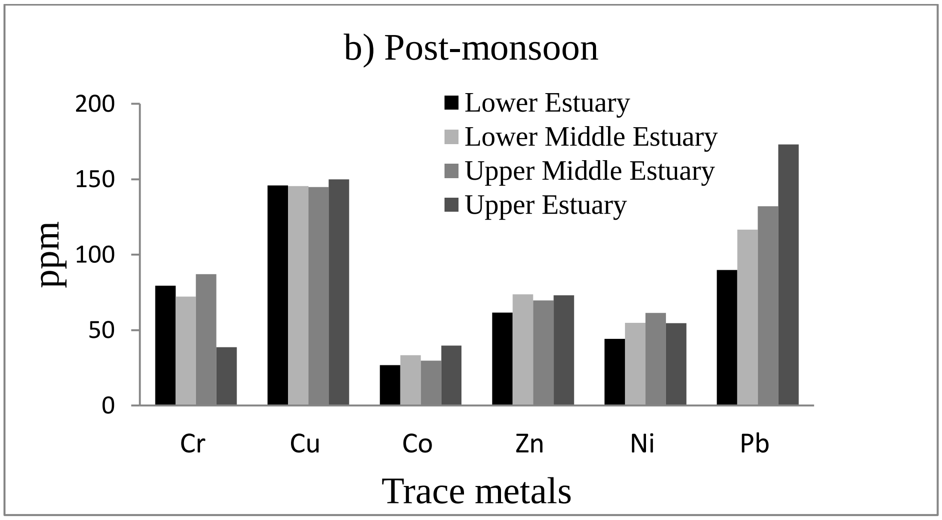
b) Post-monsoon				
Metals	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Fe (%)	5.4	5.7	7.4	7.7
Mn (ppm)	1229.9	3736.4	1724.1	2091.9
Al (%)	6.8	8.6	6.7	7.5

c) Pre-monsoon				
Metals	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
Fe (%)	5.3	5.2	6.4	7.0
Mn (ppm)	1317.5	2980.8	1860.3	2916.5
Al (%)	6.1	8.3	6.7	7.4

Fig 4B.3. Total heterotrophic bacterial distribution (CFU g^{-1} sediment) within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

THC				
Seasons	Lower estuary	Lower middle estuary	Upper middle estuary	Upper Estuary
Monsoon (a)	2.50E+05	3.00E+05	3.20E+05	3.00E+05
Post-monsoon (b)	4.00E+07	4.40E+07	3.00E+06	2.80E+07
Pre-monsoon (c)	2.00E+08	3.00E+05	4.50E+08	3.20E+07


a) Monsoon
Lower Estuary
Lower Middle Estuary
Upper Middle Estuary
Upper Estuary
ppm
200
150
100
50
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals


b) Post-monsoon
Lower Estuary
Lower Middle Estuary
Upper Middle Estuary
Upper Estuary
ppm
200
150
100
50
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals

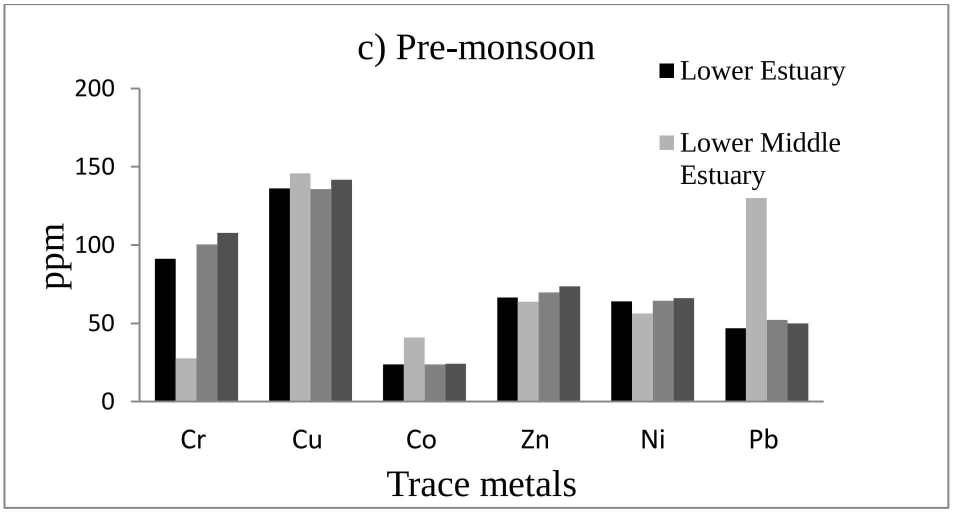

c) Pre-monsoon
Lower Estuary
Lower Middle Estuary
ppm
200
150
100
50
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals

Fig 4B.1. Trace metal (Cr, Cu, Co, Zn, Ni and Pb) distribution within mangroves in lower, lower middle, upper middle and upper estuarine regions of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons

From the results described above, it is clear that finer sediments are predominant in mangrove environment irrespective of locations and seasons. The mangrove vegetation is associated with a unique horizontal root network (Kumaran et al., 2005) that enhances the deposition of fine grained suspended sediments and prevents re-suspension and erosion of fine sediments (Soto-Jimenez and Paez-Osuna, 2001). The sediment accretion capacity of mangroves roots (Wolanski et al., 1992) increases friction and that in turn reduces tidal current velocities and as the tide progresses through the forest, the current slows and fine sediment get deposited. Entrapment of finer sediments by mangrove roots might have also act as additional factor for higher sediment deposition. Calmer hydrodynamic conditions within mangroves act as favourable environment for finer sediments deposition. Therefore, higher concentration of finer sediments is observed at all the locations and in all the three seasons within mangrove sediments.

However, sand content is higher in monsoon season compared to other seasons at lower estuary. Higher sand in monsoon season could be attributed to predominance of terrestrial over tidal sediments (Ukpong, 1997). In addition, stronger hydrodynamic activity towards lower estuary facilitates deposition of coarser sediments within unvegetated mudflats. Part of this must have transported and settled in mangroves.

In lower estuary, mud (silt+clay) and organic carbon are higher in post-monsoon and pre-monsoon indicating role of tide and tidal currents during non-monsoon seasons in distribution and deposition of finer sediments. The sampling location of lower middle estuary is part of sub-channel of Mandovi estuary. Calmer hydrodynamic conditions within the sub channel facilitate deposition of finer sediments and higher organic matter. In addition to terrestrial detritus (Jagtap, 1987), mangrove litter provides source of organic matter in mangroves. Deposition of finer sediments along with high organic matter is regulated largely by tide and tidal currents within the sub-channel. Upper middle estuary and upper estuary samples represent main river channel and therefore contain more sand and silt during monsoon respectively compared to other seasons. Upper estuarine mangrove sediments in all the three seasons contain appreciable quantity of silt possibly representing terrigenous source. Also includes organic matter from different sources like plant debris, agricultural, industrial, sewage wastes. Similarly, inorganic material will include in addition to weathering products, loose mine particles, industrial waste etc. These materials brought from the river during high run off and flood may get incorporated possibly in mangrove sediments. During non-monsoon season, especially spring high tide, when mangroves are flooded, more finer sediments transported by tidal currents will get incorporated in mangrove sediments. Therefore, Mangrove sediments are known to retain higher finer sediments along with organic matter. The root system enhances the sediment deposition of finer sediments and organic carbon (Furukawa and Wolanski, 1996).

According to Muzuka and Shaghude (2000), muddier sites contain higher content of organic carbon relative to sandier sites. Although organic carbon concentration generally depends upon the grain size and is enriched in fine grained sediments (Falco et al., 2004), factors such as balance between accumulation and degradation rate of organic matter and the source and type of organic material control the concentration of organic carbon in sediments (Schorer, 1997). Sources may be allochthonous organic production by benthic or epiphytic micro or macro algae, and local water production by phytoplankton (Bouillon et al., 2004) within water bodies. Litter from trees (leaves, propagules and twigs) and subsurface root growth also provide significant inputs of organic carbon to mangrove sediments (Alongi, 1998).

The increase in fine particles, results from the baffle effect of mangrove roots and trunks, which enhance fine particle settling and stabilization (Alongi, 2005). The trapping efficiency by root systems that retards forces of erosion seems is a key factor in the burial process. The rate of burial in mangrove forests is, highly variable and less dependent on deposition of detritus. High organic carbon obtained may be due to faster accretion resulting in extensive organic carbon burial into sediment layers where degradation is low due to anaerobic (Gonneea et al., 2004) condition. Organic carbon is less, especially in lower estuary and upper estuary during monsoon season which might be due dilution by coarser particles during this season.

In lower estuary, highest concentration of Fe and Al is noted in monsoon season and Mn in pre-monsoon season. In lower middle, upper middle estuary and upper estuary metals concentration is highest in monsoon except Mn, which is highest in post-monsoon at lower middle estuary. Further, in lower middle, upper middle and upper estuary, most of metals decrease in the concentration from monsoon to post-monsoon and then to pre-monsoon. This indicates that land derived material brought during monsoon is the major source for these metals. The process of flocculation favours the deposition of various metals during monsoon season which is also observed in mudflat environment described earlier. In general, most of the studied elements (Mn, Cr and Ni) in lower estuary; (Co and Pb) in lower middle estuary; Cr in upper middle and upper estuary shows higher concentration during pre-monsoon indicating favorable conditions for incorporations of these metals in the sediments during this season. During this season, estuary is largely controlled by saline water influx and therefore salinity and associated parameters largely regulates the deposition of metals. The deposited metals get incorporated into the sediments and remain in the sediments depending on the sediment size, organic matter and oxic/anoxic conditions. During monsoon, considerable quantity of fresh water is added to the estuary from the catchment area of Mandovi, which is responsible for diluting the salinity and thus decreasing these metal concentrations at lower estuary.

As mentioned earlier, the catchment area of Mandovi estuary is known for considerable anthropogenic activity especially open cast mining of Mn and Fe ores. The higher concentration obtained of these metals during monsoon can be directly related to the material input from the catchment area. Further, the concentration of Fe and Mn within the estuarine sediments can be related to various natural and anthropogenic activities. All along the banks of especially Mandovi estuary, ore processing units to enrich the percent of iron ore are operating and it is expected that these industries are forced to release associated elements to the waters of the estuary. It is also important to understand the processes involved in the distribution and concentration of Fe and Mn within the estuarine system in addition to the source from where they have been received. In Mandovi estuary higher salinity is maintained from the mouth of estuary up to slightly upstream end as discussed in Chapter 3. Higher concentration of Mn obtained during non-monsoon months explains possible availability of higher salinity in this part of the estuary which might have supported Mn precipitation. In lower estuary, among trace metals, except for Zn all other elements analysed are observed to show higher concentration in non-monsoon seasons. Higher Zn concentration obtained in the aquatic environment can be from industrial source as well as from sewage effluents (Alagarsamy, 2006) in addition to land runoff. Finer sediments (silt and clay) and organic carbon maintains relatively higher concentration of Zn during non-monsoon seasons, supporting that the organic carbon acts as a trap for most of the metals within the mangrove sediments. It is a known fact that the organic matter decides the site of deposition of metals due to its ability of complexation and adsorption, which leads to strong correlation between it and metal elements (Gonzalez et al., 1991; Ergin et al., 1996).

Higher concentration of Al, Fe, Mn in upper and upper middle estuary and Al, Fe in lower middle and lower estuary during monsoon indicate their source as land origin and associated with clay minerals. Further, higher trace metals Cu, Co and Ni and Zn during monsoon reveals their association with Al, Fe and Mn. There is some similarity in trace metal distribution in lower estuary and upper middle estuary they being from the main channel mangrove sediments.

When four locations are compared, higher metal concentrations are observed in lower middle estuary (Table 4B.2) and (Fig 4B.1) and to some extent in upper estuary. Based on the distribution of sediment components and metals it can be stated that lower middle estuary harbours higher proportion of finer sediments and organic carbon and acts as a sink for metals. High wave activities towards the mouth region during monsoon results in churning due to mixing of saline waters with fresh water and regulate transport of finer sediments towards calmer environments. Estuarine mixing, flocculation and primary particle properties are major factors which control and regulate the deposition of fine grained sediments (Edzwald et al., 1975). Thus, Suspended sediments acts as carrier for metals and finally settle down and get incorporated in finer sediment or “locked up” in the sediments and organic matter from several sources **(**Beavis et al., 2006). It is also important to note that the width of the river gets reduced to less than half as moved towards the upstream region which causes tidal turbulence resulting resuspension. Due to this total suspended matter is maintained always high in bottom waters (Chapter 3). Higher concentration of metals, observed in the present study, along upper estuary therefore is explained with reference to geomorphology as well as tidal surge. Metals brought by high velocity river currents, on settling within the estuary, get incorporated in the sediments. Resuspension of bed load sediments due to tidal surge and geomorphological constriction involves drag and lift force resulting removal of less denser material from the surface sediments, which in turn helps in concentration of denser particles as well as the particles with adsorbed metals and therefore responsible for higher concentration of some of the metals along this region.

Higher heterotrophic counts are noted in non-monsoon seasons at all the estuarine regions. In lower estuary, upper middle and upper estuary, higher counts are noted in pre-monsoon season. In lower middle estuary, higher heterotrophic counts are noted in post-monsoon. Alongi et al. (2000) noted that bacterial density in mangrove sediments are related to several factors one of which is organic carbon which has major effect on bacterial populations. However, higher organic matter in monsoon season has not supported higher bacterial counts. Heterotrophic bacterial counts are found to be higher in middle and upper estuarine regions while lesser counts are noted towards the lower estuary. The occurrence of higher finer sediment concentration accompanied with higher organic matter seems to determine the heterotrophic counts in Mandovi estuary.

4.2. Pollution level based on bulk sediment chemistry

Index of Geo-accumulation (Igeo) has been used widely to evaluate the degree of metal contamination or pollution in terrestrial, aquatic and marine environment (Tijani and Onodera, 2009). Pollution Load Index **(**PLI) also provides a simple, comparative means for assessing a site or estuarine quality: a value of zero indicates perfection, a value of one indicates only baseline levels of pollutants present and values above one would indicate progressive deterioration of the site and estuarine sediment quality. Igeo and PLI are computed to mudflat and mangrove sediments collected at different regions of Mandovi estuary to understand level of metal pollution.

4.2A. Mudflats

The Igeo values of the studied metals for the sediments collected from mudflats revealed that the sediments of lower middle and upper middle estuary are moderately enriched with Mn, Cu, and Pb in all the three seasons (Table 4A.4). The sediments collected from the lower estuary shows values which fall under Igeo class zero indicating unpolluted to moderately polluted for these metals. However, in lower estuary, though the metals show unpolluted to moderately polluted, Igeo values are higher in monsoon season than obtained for post-monsoon and pre-monsoon seasons. In lower middle estuary, post-monsoon season shows higher metal (Mn, Cu and Pb) contamination whereas, in upper middle estuary Mn and Pb show higher Igeo value during post-monsoon and Cu during monsoon season. When data of lower middle and upper middle estuarine regions are compared, lower middle estuarine sediments exhibit slightly higher enrichment with respect to Mn, Cu and Pb especially in monsoon and post-monsoon seasons. Cu and Pb, however, show higher values in upper middle estuary during pre-monsoon season.

Further, pollution load index was also computed for the sediments and the data indicated values greater than 1 for lower middle and upper middle estuarine sediments indicating deterioration of sediment quality with metal contamination at these locations. Sediments of lower middle and upper middle estuarine regions show higher value in monsoon and post-monsoon compared to pre-monsoon season (Table 4A.5).

4.2A.a. *Geoaccumulation Index (Igeo)*

Table 4A.4. Geoaccumulation Index (Igeo) values for lower, lower middle and upper middle regions of mudflat sediments of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

a) Monsoon			
Metals	Lower Estuary	Lower Middle Estuary	Upper Middle Estuary
Fe	-2.09	-0.24	0.12
Mn	-1.77	**1.52**	**1.11**
Cr	-1.47	-0.44	-0.58
Cu	**0.67**	**1.04**	**1.01**
Co	-1.28	0.00	-0.03
Zn	-1.89	-0.78	-0.80
Ni	-2.02	-0.66	-0.61
Pb	**0.95**	**1.38**	**1.27**

b) Post-monsoon			
Metals	Lower Estuary	Lower Middle Estuary	Upper Middle Estuary
Fe	-1.98	-0.18	-0.13

Mn	-1.78	**1.60**	**1.44**
Cr	-0.25	-0.13	-0.51
Cu	**0.54**	**1.07**	**0.99**
Co	-1.68	0.03	0.07
Zn	-2.08	-0.92	-1.22
Ni	-1.77	-0.68	-0.94

c) Pre-monsoon			
Metals	Lower Estuary	Lower Middle Estuary	Upper Middle Estuary
Fe	-2.11	-0.26	-0.28
Mn	-1.44	**1.61**	**1.32**
Cr	-0.77	-0.16	-0.73
Cu	**0.58**	**0.88**	**0.95**
Co	-1.67	-0.05	-0.01
Zn	-2.02	-0.93	-1.16
Ni	-1.75	-0.69	-1.04
Pb	**0.08**	**0.68**	**1.30**

Pb	-0.08	**1.55**	**1.54**

4.2A.b. *Pollution Load Index (PLI)*

Table 4A.5. Pollution Load Index (PLI) values for lower, lower middle and upper middle regions of mudflat sediments of Mandovi estuary in monsoon (a), post-monsoon (b) and pre-monsoon (c) seasons

Estuarine locations	Monsoon	Post-monsoon	Pre-monsoon
Lower Estuary	0.69	0.68	0.68
Lower Middle Estuary	1.75	1.79	1.27
Upper Middle Estuary	1.70	1.67	1.54

From the results, it is clear that middle estuary shows higher level of metal enrichment than lower estuary. Lower enrichment of metals at the lower estuary might be due to strong tidal flushing especially during flood tide, transporting most of the contaminants along with finer sediments towards the middle estuarine regions. In addition factors such as an abundance of coarser particles, mineral composition and physico- chemical conditions (Praveena et al., 2007) must have played significant role in lower PLI in lower estuary. Metals transported finally settled in relatively quiet environment in middle estuary, along with finer sediments and organic matter as discussed earlier. Source of the sediments may be terrigenous, marine or even atmospheric. Additional factors responsible for enrichment of metals in middle estuary are the movement of barges carrying ores and mechanized fishing boats in Mandovi estuary.

As seen from the results, metals show varying level of contamination in different regions of estuary in different seasons, indicating difference in source of supply of sediments and metals and difference in factor variables controlling the processes. Igeo data shows contamination of Mn, Cu and Pb in middle estuary in different seasons with varying values. Seasonal processes have a large role in introducing metals from various sources. Higher metal values observed in non-monsoon seasons in lower middle and upper middle estuary except for Cu, which is slightly higher in upper middle estuary during monsoon, indicate redistribution of metals regulated by tides and tidal currents. However, higher Igeo values obtained especially for Cu and Pb in monsoon at lower estuary indicates fresh water brought material as high rainfall and associated runoff must have brought large land derived material, which reaches lower estuary as most of the saline water is flushed out of the estuary by fresh water during this season.

High PLI values are obtained for lower middle and upper middle estuary. Within middle estuary, the values are relatively higher for lower middle estuary during monsoon and post-monsoon. Lower middle estuary sample location being a sub-channel, facilitates relatively calmer conditions compared to main channel during monsoon for the deposition of finer sediments, organic matter and enrichment of metals as stated earlier. Higher contamination noted in pre-monsoon at the upper middle estuary is regulated by tidal currents. In addition, anthropogenic activities like loading of iron ores and movement of barges must have contributed at this location.

4.2B. Mangroves

In order to understand the level of contamination of metals (Fe, Mn, Cr, Cu, Co, Zn, Ni and Pb) in mangrove sediments, Igeo is computed. It is observed that sediments of lower, lower middle, upper middle and upper estuary are moderately enriched with Mn, Cu, and Pb in all the three seasons with exceptions for Mn and Pb in lower estuary (Table 4B.4). In lower estuary, Mn falls in unpolluted- moderately polluted class in all the three seasons. Similar observation is also noted in monsoon and pre-monsoon seasons in case of Pb.

Igeo value of Mn in lower middle estuary during post-monsoon falls in class 3 indicating moderately – strongly polluted class whereas, it falls in moderately polluted class in other two seasons. Igeo values of Cu falls in moderately polluted class in all the three seasons. Pb falls in moderately polluted class in monsoon and post-monsoon whereas, in moderately-strongly polluted class in pre-monsoon season. In upper middle estuary, Mn and Cu fall in moderately polluted class in all the three seasons with exception noted during post-monsoon where Mn falls in unpolluted- moderately polluted class. Pb falls in moderately polluted class, moderately-strongly polluted, unpolluted- moderately polluted in monsoon, post-monsoon and pre-monsoon respectively. In the upper estuary, Mn, Cu and Pb fall in moderately polluted class except for Pb which falls in unpolluted- moderately polluted class and moderately- strongly polluted class in pre-monsoon and post-monsoon respectively.

4.2B.a. *Geoaccumulation Index (Igeo)*

Table 4B.4. Geoaccumulation Index (Igeo) values in lower, lower middle, upper middle and upper estuarine regions of mangrove sediments of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons

a) Monsoon				
Metals	Lower Estuary	Lower Middle Estuary	Upper Middle Estuary	Upper Estuary
Fe	-0.22	-0.01	0.22	0.44
Mn	**0.34**	**1.61**	**1.44**	**1.81**
Cr	-0.47	-0.30	-0.44	-0.33
Cu	**1.06**	**1.22**	**1.14**	**1.28**
Co	-0.52	0.17	0.01	0.63
Zn	-1.05	-0.63	-0.86	-0.73
Ni	-1.01	-0.31	-0.65	-0.46
Pb	**0.51**	**1.35**	**1.28**	**1.65**

b) Post-monsoon				
Metals	Lower	Lower	Upper	Upper Estuary

	Estuary	Middle Estuary	Middle Estuary	
Fe	-0.39	-0.32	0.06	0.14
Mn	**0.45**	**2.03**	**0.91**	**1.13**
Cr	-0.65	-0.79	-0.52	-1.68
Cu	**1.11**	**1.11**	**1.10**	**1.15**
Co	-0.09	0.23	2.29	0.48
Zn	-1.21	-0.95	2.61	-0.96
Ni	-1.21	-0.89	2.57	-0.90
Pb	**1.58**	**1.96**	**2.81**	**2.52**

c)Pre-monsoon				
Metals	Lower Estuary	Lower Middle Estuary	Upper Middle Estuary	Upper Estuary
Fe	-0.42	-0.45	-0.15	-0.02
Mn	**0.55**	**1.72**	**1.05**	**1.70**
Cr	-0.45	-2.17	-0.31	-0.21
Cu	**1.01**	**1.11**	**1.01**	**1.07**
Co	-0.27	0.52	-0.27	-0.23
Zn	-1.10	-1.17	-1.03	-0.95
Ni	-0.70	-0.86	-0.66	-0.62
Pb	**0.64**	**2.81**	**0.80**	**0.74**

4.2B.b. *Pollution Load Index (PLI)*

Table 4B.5. Pollution Load Index (PLI) values in lower, lower middle, upper middle and upper estuarine regions of mangrove sediments of Mandovi estuary in monsoon (a); post-monsoon (b) and pre-monsoon (c) seasons.

Estuarine locations	Monsoon	Post-monsoon	Pre-monsoon
Lower Estuary	0.78	1.44	1.40
Lower Middle Estuary	1.97	1.84	1.70
Upper Middle Estuary	1.81	1.77	1.55
Upper Estuary	2.18	1.76	1.70

From the results, it is clear that upper estuary shows higher Igeo values for Mn, Cu and Pb in monsoon season compared to other estuarine regions indicating the input from land source. In post-monsoon, Mn is higher in lower middle estuary whereas, Cu in upper estuary and Pb in upper middle estuary. In pre-monsoon, higher Mn, Cu and Pb is noted in lower middle estuary. As mentioned earlier, the catchment area of Mandovi River is known for large open cast mechanized iron and ferro-manganese mining activity. Mn addition therefore is expected from land source, and redistribution by tidal currents. Important anthropogenic inputs of Cu in estuarine and coastal waters include sewage sludge dump sites and antifouling paints (Kennish, 1998; Abu-Hilal et al., 1988; Laxen, 1983), boat exhaust, spillage of oil, and other petroleum products from mechanized boats employed for fishing and barges used for transport of ores. The municipal and/or industrial wastewater discharges into estuaries are the other important sources for contamination of water and sediment with metals (Gonzalez and Brugmann, 1991). Pb must be also adding through atmosphere source.

PLI values show deterioration of sediments with metal contaminants in monsoon season in lower middle, upper middle and upper estuary and during non-monsoon seasons in all the four regions (Table 4B.5). Among the four locations, upper estuarine sediments show highest PLI value followed by lower middle and upper middle estuary in monsoon indicating severe deterioration of sediments by metals.

Geoaccumulation Index shows that mangroves are more contaminated by metals compared to mudflats in all the three seasons in different estuarine regions of Mandovi estuary. The root controlled low hydrodynamic conditions in mangroves favours the accumulation of finer sediment, organic matter and metals. Mangrove areas are sheltered from strong tidal currents and thus subject to lesser erosion and therefore known as sinks for sediments (Rees et al., 1996), contaminated by metals.

The mudflats when compared to mangroves in Mandovi estuary, coarser sediments are found to be higher in the mudflat environment in all the seasons, in lower estuary. Finer sediments are higher in both mangrove and mudflat regions of middle estuary with variations noted in different seasons. During monsoon and post-monsoon, finer sediments are higher in mudflat sediments and during pre-monsoon they are higher in mangrove sediments at lower middle estuary. In upper middle estuary, higher finer sediments are seen in mudflat sediments than that of mangroves. Organic carbon is higher in mangrove areas than mudflats in all the three seasons except for higher organic carbon in mudflats of upper middle estuary during post-monsoon and in lower middle estuary during pre-monsoon.

During monsoon, mangroves of lower estuary show higher concentration of metals except for Al and Pb. Similarly, in post-monsoon season all the elements except Cr are higher in mangroves of lower estuary. In pre-monsoon, similar to monsoon the concentration of metals is higher in mangroves except for Al which is found to be higher in mudflats. In lower middle estuary, almost all the metals are higher in mangroves except Pb which show less variability between the two in monsoon season. In post-monsoon, Fe, Al, Cr, Zn and Ni show higher concentration in mudflats whereas, Mn, Co, Cu and Pb are found to be higher in mangroves. In pre-monsoon season, Fe, Cr, Zn and Ni are comparatively higher in mudflat sediments whereas, other elements are higher in mangroves. In upper middle estuary, Al and Zn are higher in mudflats whereas, Pb and Ni show less variability between the two sub-environments during monsoon season. The other elements are found to be higher in mangroves at upper middle estuary in monsoon season. In post-monsoon, Al and Mn are found to be higher in mudflats and others in mangroves of upper middle estuary. Cr and Co, however show less variation between the sub-environments. In pre-monsoon, Al, Mn, Co and Pb are noted to be higher in mudflats and other elements in mangroves. Overall, most of the metals show higher concentration in mangrove sediments than mudflat sediments of lower estuary in all the three seasons and to some extent in lower middle estuary. However, in middle estuary, some of the metals show higher concentration in mudflats in different seasons. Within mangroves the reducing conditions develop, because of high content of organic matter, microbial decomposition of organic matter and relatively higher fine sediment particles, result in potentially high accumulation of trace metals (Thamdrup et al., 1994). Fine particles such as clay along with colloidal materials, organic matter as well as Fe- Mn oxide surface coatings, play an important role in regulating the deposition of trace metals to these sediments (Harikumar and Jisha, 2010). The clay particles are flexible because of their sheet structure and have large surface area which is chemically reactive to attract and hold positively charged metal ions (Hiller, 2003). Therefore, mangroves in the sheltered boggy hydro environment facilitate deposition of fine sediments normally enriched with nutrients, metals and minerals (Srivastava et al., 2012). Migration of metals from mangroves to adjacent mudflats is regulated by freshwater influx and tidal currents. Seasonal variation in metal concentration between the two sub-environments is also affected by addition of metals from different sources. Mangroves of lower estuary and lower middle estuary show higher heterotrophic counts than adjacent mudflats in monsoon and post-monsoon seasons. However, in upper middle estuary, mudflats show higher heterotrophic counts than mangroves during these seasons.

4.3. Speciation- Mudflats

The sequential extraction procedure was applied for mudflat sediments collected from lower, lower middle and upper middle estuaries in different seasons in order to understand the distribution of metals in different phases. Percentage of metals (Fe, Mn, Co, Zn and Pb) are computed for exchangeable (F1); carbonate (F2); Fe-Mn oxide (F3); organic (F4) and residual (F5) fractions. Pie charts are employed to represent different fractions of a given elements. The concentration of Fe is high in residual fractions compared to non-residual fractions. Zn shows similar behaviour as that of Fe with some exceptions. Other, metals viz. Mn, Co and Pb show higher values in bioavailable fractions. Mn and Pb also show higher levels of pollution as indicated in values of pollution indices. Pie chart was plotted for Mn, Co, Pb and Zn.

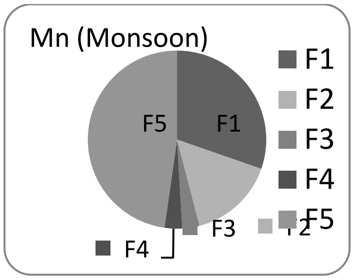
In lower estuarine sediments, high concentration of Fe is observed in residual fraction with average value of 99.36 %, 99.67 % and 98.95 % in monsoon, post-monsoon and pre-monsoon seasons respectively. The concentration of Fe bound to other fractions is observed to be less. Similar results are also observed in sediments of lower middle and upper middle estuary i.e. Fe is associated with mainly residual fraction and least concentration is found in the bioavailable fractions in all the seasons. In the lower middle estuary, the average concentration of Fe in residual fraction is 99.12 %, 96.33 % and 92.75 % in monsoon, post-monsoon and pre-monsoon seasons respectively. In the upper middle estuary, Fe concentration shows average values of 99.52 %, 97.82 % and 79.47 % in residual fraction in monsoon; post-monsoon and pre-monsoon seasons respectively. Among the four bioavailable fractions, Fe shows higher concentration in organic phase compared to other bioavailable fractions. Distribution of different fractions of metals viz. Mn, Co, Pb and Zn from sediments of lower, lower middle and upper middle estuary in different seasons is presented in figures 4A.2, 4A.3 and 4A.4 respectively. In lower estuary, significant concentration of Mn is available in F1 fraction (Avg. 30.32 %) followed by F2 fraction (Avg. 15.62 %) and less concentration is found in F3 fraction (Avg. 3.10 %) and F4 fraction (Avg. 3.19 %) in monsoon season (Fig 4A.2). Mn bound to residual fraction shows average value of 47.74 % in monsoon season. During post-monsoon, Mn concentration is 46.14 % and 25.25 % in carbonate and Fe-Mn oxide fractions respectively (Fig 4A.2). Mn bound to F1, F4 and F5 fractions show average value of 6.51 %, 4.99 % and 17.09 % respectively. Significant concentration of Mn is available in F2 fraction (Avg. 46.24 %) and least concentration is found in F1, F3 and F4 fractions with average value of 8.16 %, 4.05 % and 3.79 % respectively during pre-monsoon (Fig 4A.2). The concentration is found to be higher in bioavailable (F1+F2+F3+F4) fractions with less concentration found in residual fraction (Avg. 37.74 %) during pre-monsoon. When the data of all the first four fractions are added in order to compare the bioavailability in different seasons it is observed that, Mn is bioavailable in post-monsoon (82.90 %) followed by pre-monsoon (62.26 %) and monsoon (52.25 %). Mn is found to be higher in exchangeable fraction in monsoon season. In both post-monsoon and pre-monsoon seasons, Mn is significantly bound to carbonate fraction. Towards lower middle estuary, Mn concentration shows Avg. 25.57 % in exchangeable, Avg. 52.63 % in carbonate, Avg. 2.47 % in Fe-Mn oxide, Avg. 6.30 % in organic and Avg. 13.01 % in residual in monsoon season. In monsoon at this location, Mn is found to be higher in carbonate fraction (Fig 4A.3). During post monsoon, Mn is more associated with carbonate phase (Avg. 47.31 %). Next to carbonate Mn is higher is exchangeable with average value of 26.98 %. Mn bound to residual fraction shows value of Avg. 18.36 % and is less available in Fe-Mn oxide (2.00 %) and organic (5.33 %) fractions (Fig 4A.3). In pre-monsoon season, significant amount of Mn is available in the bioavailable phases showing Avg. 20.21 % in exchangeable, Avg. 28.05 % in carbonate, Avg. 2.23 % in Fe- Mn oxide, Avg. 40.83 % in organic and Avg. 8.65 % in residual fraction (Fig 4A.2). Mn is found to be higher in organic> carbonate> exchangeable > residual > Fe-Mn oxide in pre-monsoon season. When the data of all the three seasons are compared, Mn shows higher bioavailability within the sediments during pre-monsoon followed by monsoon and post-monsoon at this location. In upper middle estuary, the concentration of Mn in different phases shows Avg. 24.33 % in exchangeable, Avg. 27.54 % in carbonate, Avg. 1.81 % in Fe-Mn oxide, Avg. 4.95 % in organic and Avg. 41.36 % in residual fraction in monsoon season (Fig 4A.4). Considering the average values it is observed that Mn within the bioavailable fractions is associated as carbonate > exchangeable > organic > Fe-Mn oxide. During post-monsoon, Mn shows higher concentration in exchangeable fraction (Avg. 48.89 %) followed by carbonate (Avg. 22.15 %) and residual (Avg. 12.99 %). The concentration of Fe-Mn oxide is less with Avg. 12.49 % and in organic fraction it is Avg. 3.47 % (Fig 4A.2). In pre-monsoon, the concentration of Mn is found to be higher in residual fraction (Avg. 66.44 %). The concentration within the bioavailable fractions is higher in exchangeable fraction (Avg. 14.76 %). The concentration of Mn shows Avg. 6.72 % in carbonate; Avg. 3.70 % Fe-Mn oxide and Avg. 8.35 % in organic. Mn shows higher concentration in exchangeable> organic > carbonate > Fe-Mn oxide in bioavailable phases (Fig 4A.2). The bioavailability of Mn is highest in post-monsoon followed by monsoon and pre-monsoon.

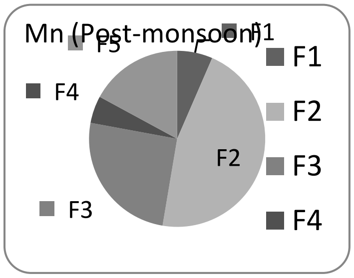
The distribution of Co in all the three seasons in different fractions is described below. In lower estuary, Co shows 53.98 %, 50.60 % and 40.06 % concentrations in Fe-Mn oxide fraction in monsoon, post-monsoon and pre-monsoon seasons (Fig 4A.2) respectively. The next dominant fraction after Fe- Mn oxide is exchangeable fraction where Co shows 29.37 %, 15.35 % and 16.43 % in monsoon, post-monsoon and pre-monsoon seasons. The concentration of Co in carbonate fraction are as follows i.e. 7.58 %, 11.23 % and 16.05 % in monsoon, post-monsoon and pre-monsoon respectively whereas, in organic fraction Co shows 2.55 %, 1.66 % and 6.72 % in monsoon, post-monsoon and pre-monsoon seasons. In residual fraction, Co attributed 6.48 %, 21.13 % and 20.17 % in monsoon, post-monsoon and pre-monsoon. Co is observed to be higher in Fe-Mn oxide fraction followed by exchangeable and carbonate phases in all the three seasons in sediments of lower estuary. The bioavailability of Co is found to be higher in monsoon followed by pre-monsoon and post-monsoon. In lower middle estuary, Co shows 11.54 % in exchangeable, 5.12 % in carbonate, 27.27 % in Fe-Mn oxide, 6.00 in organic and 50.04 % in residual during monsoon (Fig 4A.3) season whereas, 18.53 % in F1, 12.94 % in F2, 29.71 % in F3, 5.48 % in F4 and 33.32 % in F5 fraction in post-monsoon (Fig 4A.3). During pre-monsoon, higher Co is associated with residual fraction (41.83 %) although concentration in all the bioavailable fractions is considerably higher (58.16 %). The concentrations of Co in exchangeable, carbonate, Fe-Mn oxide and organic fractions are 12.55 %, 14.36 %, 20.67 % and 10.56 % respectively (Fig 4A.3). Similar results are also observed in lower estuary i.e. concentration of Co is higher in Fe-Mn oxide fraction in all the three seasons (Fig 4A.3).

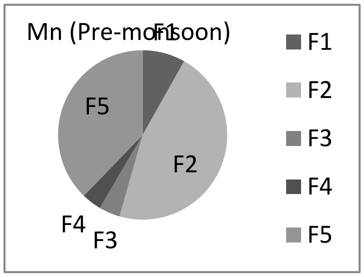
Bioavailability of Co is observed to be higher in post-monsoon followed by pre-monsoon and monsoon season. In upper middle estuary, Co bound to different fractions in monsoon season is of the order Fe - Mn oxide (39.01 %) > residual (24.32 %) > exchangeable (13.92 %) > carbonate (12.91 %) > organic (9.81 %) (Fig 4A.4). In post-monsoon, the concentration of Co is found to be higher in residual fraction (56.31 %) followed by Fe-Mn oxide (15.41 %). In exchangeable, carbonate and organic fractions, the concentration of Co is less with concentration of 11.08 %, 9.03 % and 8.14 % respectively (Fig 4A.4). In pre-monsoon, Co shows 9.82 %, 9.47 %, 19.65 %, 10.53 %, 50.50 % in exchangeable, carbonate, Fe-Mn oxide, organic and residual fractions respectively (Fig 4A.4). The concentration of Co is also found to be higher in Fe-Mn oxide as observed in both the estuarine areas i.e. lower and lower middle estuary. Bioavailability of Co is found to be higher in pre-monsoon followed by post-monsoon and monsoon season.

In lower estuary, the concentration of Pb in monsoon is 6.07 %, 11.54 %, 25.87 %, 9.60 % and 46.89 % in different fractions i.e. exchangeable, carbonate, Fe-Mn oxide, organic and residual fractions respectively (Fig 4A.2). Pb in bioavailable fractions is observed to be higher in Fe-Mn oxide > carbonate > organic > exchangeable fractions. In post-monsoon, Pb shows 4.87 % in exchangeable, 15.45 % in carbonate, 17.83 % in Fe-Mn oxide, 22.06 % in organic and 39.77 % in residual fraction (Fig 4A.2). In pre-monsoon, Pb shows 21.49 %, 7.78 %, 28.62 %, 8.68 % and 33.41 % in exchangeable, carbonate, Fe-Mn oxide, organic and residual fraction respectively (Fig 4A.2). Pb is found to be higher in Fe-Mn oxide in monsoon and pre-monsoon whereas, it is higher in organic fraction during post-monsoon within bioavailable fractions. Bioavailability of Pb is found to be higher in pre-monsoon and post-monsoon followed by monsoon season. In lower middle estuary, Pb bound to exchangeable, carbonate, Fe – Mn oxyhydroxides, organic and residual fraction show 9.64 %, 13.55 %, 32.42 %, 3.56 % and 40.81 % respectively in monsoon season (Fig 4A.3). Among different bioavailable phases, Pb is strongly associated with Fe-Mn oxide fraction followed by carbonate phase whereas, concentration in exchangeable and organic fractions is considerably less during monsoon season. During post-monsoon among bioavailable fractions, the concentration of Pb is higher in Fe-Mn oxide (34.96 %) followed by exchangeable fraction (16.26 %) with considerably less concentration in carbonate (4.60 %) and organic (6.81 %) (Fig 4A.3). The concentration of Pb in F5 fraction shows 37.35 %. In pre-monsoon season, Pb is found to be higher in residual fraction whereas, among the bioavailable phases Pb is found to be higher in exchangeable fraction (28.35 %). The concentration of Pb in other three bioavailable fractions i.e. carbonates, Fe-Mn oxide and organic is 13.80 %; 14.83 % and 13.14 % respectively (Fig 4A.3). The concentration of Pb is 29.85 % in residual fraction. The dominant bioavailable phase of Pb is Fe-Mn oxide during monsoon and post-monsoon whereas, exchangeable is the main during pre-monsoon. Bioavailability of Pb is found to be higher in pre-monsoon followed by post-monsoon and monsoon season. In upper middle estuary, Pb is associated with residual (67.26 %) followed by exchangeable (13.02 %) fraction during monsoon. The Pb concentration in carbonate, Fe- Mn oxide and organic is 5.95 %; 9.05 % and 4.70 % respectively (Fig 4A.4). In post monsoon, higher concentration of Pb is associated with Fe-Mn oxide (39.64 %). Pb in other fractions shows 17.03 %, 7.89 %, 3.16 % and 32.26 % in exchangeable, carbonate, organic and residual fraction respectively (Fig 4A.4). In pre monsoon, Pb shows higher concentration in residual (54.10 %) fraction followed by exchangeable (15.23 %) and carbonate (13.79 %) fractions (Fig 4A.4). Considerably less concentration is noted in Fe-Mn oxide (7.84 %) and organic (9.02 %) fractions. In monsoon and pre-monsoon, Pb is found to be dominant in residual fraction with values of 67.26 % and 54.1 % respectively whereas, during post-monsoon it is higher in Fe-Mn oxide fraction (39.64 %). Bioavailability is found to be higher in post-monsoon season whereas it is comparatively less in monsoon and post-monsoon seasons.

From sediments of lower estuary, Zn bound to exchangeable, carbonate, Fe-Mn oxide and organic fractions show 3.06 %, 3.02 %, 22.99 % and 4.47 % respectively in monsoon (Fig 4A.2). In post-monsoon, Zn shows 12.32 %, 0.99 %, 14.21 %, 9.40 % in exchangeable, carbonate, Fe-Mn oxide and organic fractions respectively (Fig 4A.2). The concentration of Zn in pre-monsoon season shows 3.65 %, 11.56 %, 9.63 %, 7.11 % in exchangeable, carbonate, Fe-Mn oxide and organic fractions respectively (Fig 4A.2). In lower estuary, higher Zn concentration is found to be associated with residual fraction i.e. 66.43 %; 63.06 % and 68.03 % in monsoon, post-monsoon and pre-monsoon. Within bioavailable fractions, Zn is found to be more associated with Fe-Mn oxide fraction in monsoon and post-monsoon seasons whereas, with carbonate in pre-monsoon season. The bioavailability of Zn is found to be higher in post-monsoon followed by monsoon and pre-monsoon in lower estuary. Towards lower middle estuary, the concentration of Zn shows 3.46 %, 20.59 %, 15.21 %, 8.72 % and 52.01 % in exchangeable, carbonate, Fe-Mn oxide, organic and residual fractions in monsoon season (Fig 4A.3). In post-monsoon, the concentration of Zn shows 1.78 %, 24.34 %, 35.06 %, 7.91 %, 30.89 % whereas, in pre-monsoon, the concentration of Zn shows 1.16 %, 6.19 %, 8.89 %, 19.45 % and 64.30 % in exchangeable, carbonate, Fe-Mn oxide, organic and residual fractions (Fig 4A.3). Zn shows higher concentration in residual fractions in monsoon and pre-monsoon (Fig 4A.3) whereas, in post-monsoon the concentration of Zn is higher (69.10 %) in bioavailable phases. Therefore, bioavailability of Zn is observed to be high in post-monsoon followed by monsoon and pre-monsoon seasons. Significant concentration of Zn is found to be available in carbonate, Fe-Mn oxide and organic fractions in monsoon, post-monsoon and pre-monsoon seasons. In upper middle estuary, the concentration of Zn in exchangeable is 0.09 %, 8.76 % and 1.98 % in monsoon, post-monsoon and pre-monsoon seasons (Fig 4A.4). Concentration of Zn in carbonate fraction shows 16.77 %, 11.50 % and 5.61 % whereas, organic fraction shows 7.11 %, 7.15 % and 8.62 % in monsoon; post-monsoon (Fig 4A.4) and pre-monsoon (Fig 4A.4). The concentration of Zn in residual fraction is 53.92 %, 54.26 % and 65.96 % in monsoon, post-monsoon and pre-monsoon season respectively. The concentration of Zn is found to be higher in residual fraction and less in bioavailable fractions during monsoon, post-monsoon and pre-monsoon seasons. Among the bioavailable forms Zn is mainly associated with Fe-Mn oxide fraction with values of 19.79 %; 18.31 % and 17.81 % in monsoon, post-monsoon and pre-monsoon seasons. Although, the concentration of Zn is higher in residual fraction it is clear that the concentration of bioavailable Zn is higher in post-monsoon followed by monsoon and pre-monsoon.







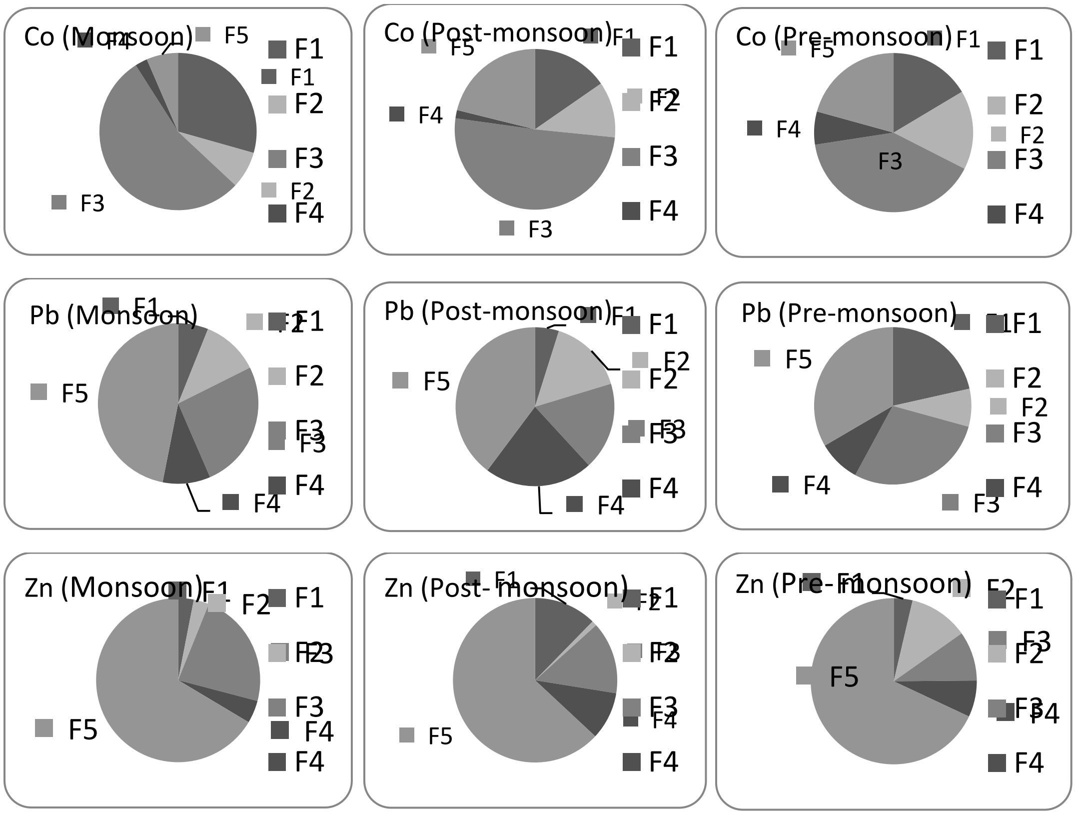
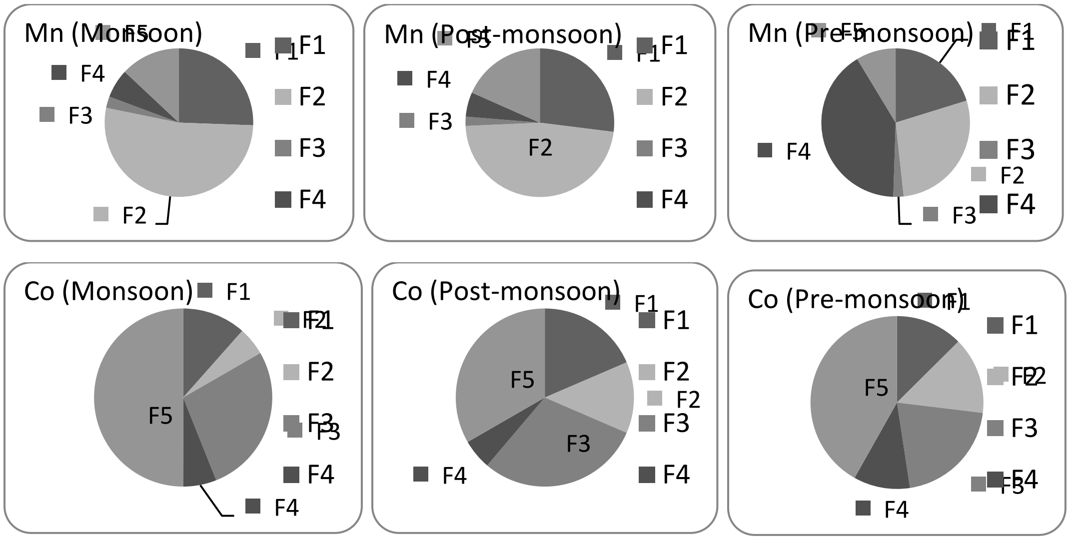


Fig 4A.2. Distribution of Mn, Co, Pb and Zn in mudflat sediments of lower estuary in different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons



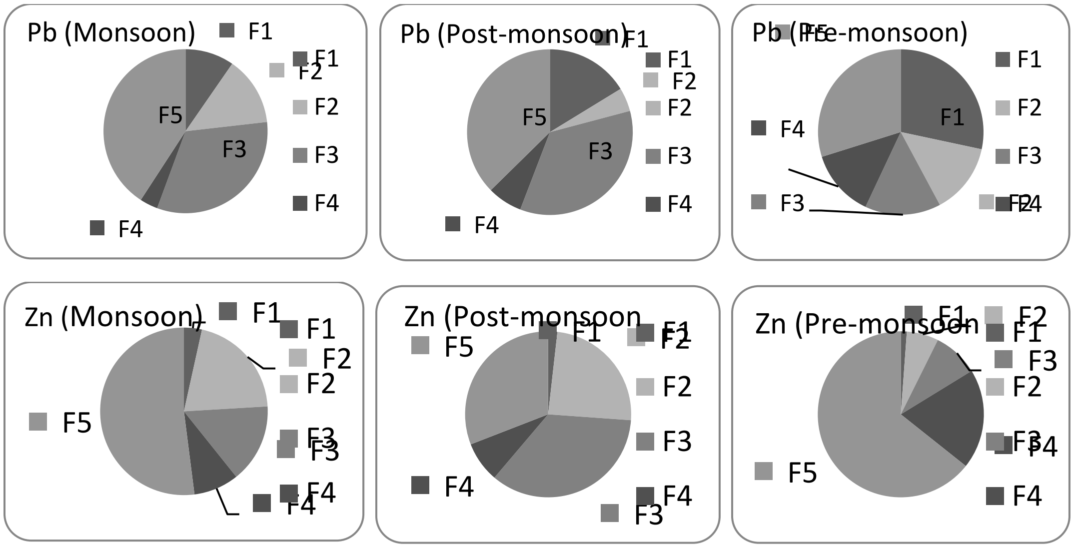
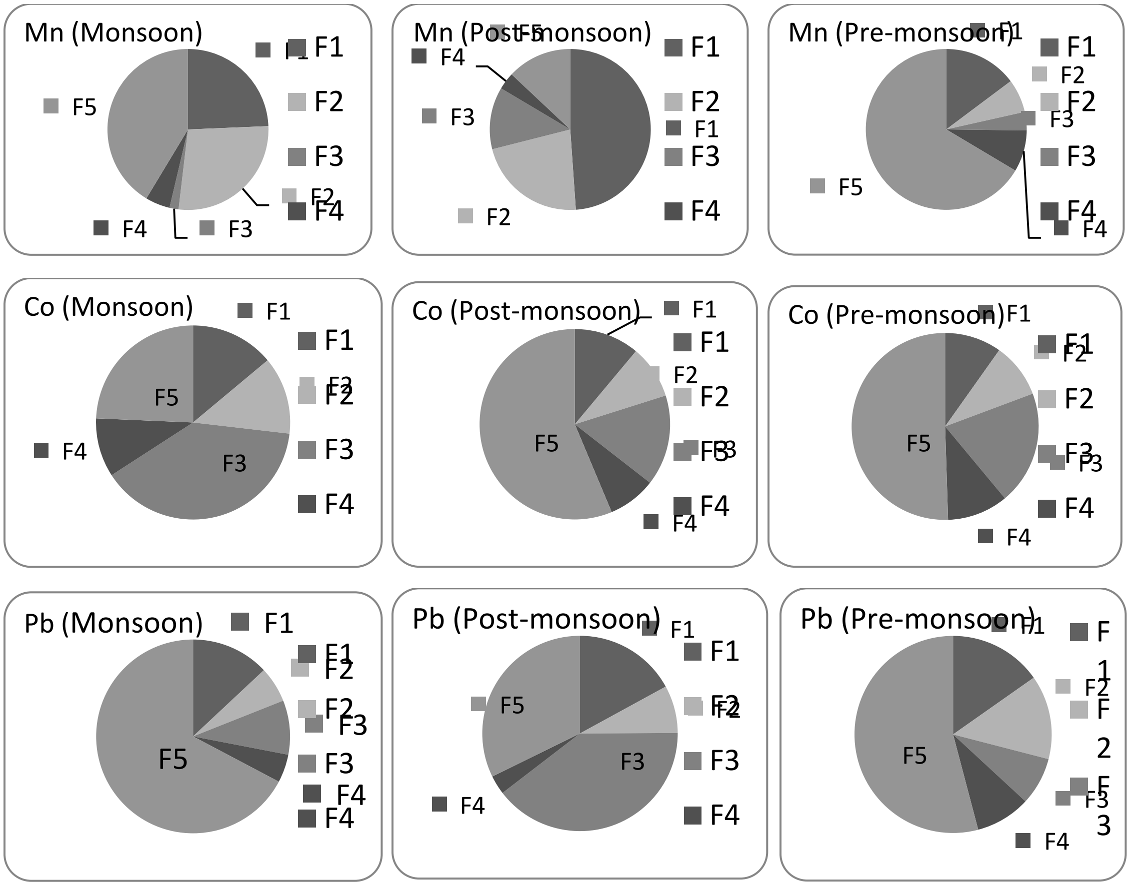


Fig 4A.3. Distribution of Mn, Co, Pb and Zn in mudflat sediments of lower middle estuarine region in different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons



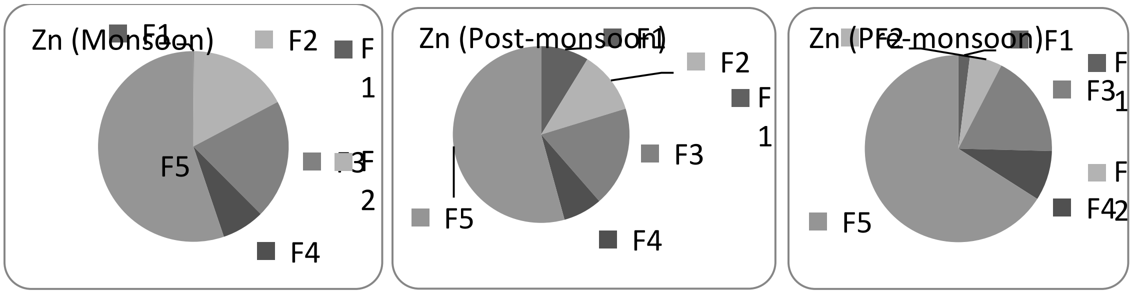


Fig 4A.4. Distribution of Mn, Co Pb and Zn in mudflat sediments of upper estuarine region in different fractions i.e F1 (exchangeable), F2 (carbonate), F3 (Fe-Mn oxides), F4 (Organic) and F5 (Residual) in monsoon, post-monsoon and pre-monsoon seasons

From the above results it is clear that Fe is associated with residual fraction in all the three estuarine areas. The residual fraction is considered the most stable, less reactive and less bioavailable since it is occluded within the crystal lattice layer of silicates and well crystallized oxide minerals (Abeh et al., 2007). Metals in this phase mostly remain stable and do not react during sedimentation and digenesis, and therefore have less potential for bioavailability. This phase could be considered as inert phase corresponding to the part of metal that cannot be mobilized (Tessier et al., 1979). To some extent Zn is also observed to be higher in residual fraction. Among the bioavailable fractions, Zn is mostly associated Fe-Mn oxide indicating Fe - Mn oxides are main carriers of Zn. The association of Zn with the Fe -Mn oxides of soils and sediments has been widely recognized by Kuo et al. (1983) and Gonzalez et al. (1994) showing that the Zn adsorption onto the oxides has high stability constants. From the results, within bioavailable fractions it is clear that Mn is higher in carbonate fraction in all the three estuarine regions with some exceptions. Mn is also higher in exchangeable fraction in monsoon season in lower estuary, in all three seasons in lower middle estuary and in monsoon and post-monsoon seasons in upper middle estuary. Further, Mn is also high in Fe-Mn oxide phase in post-monsoon in lower estuary and organic phase in pre-monsoon in lower middle estuary. The high level of Mn in carbonate fraction is due to the similarity in ionic radii of Mn to that of calcium which allows Mn to substitute for Ca in carbonate phase (Pedersen and Price, 1982; Zhang et al., 1988). The association of Mn with carbonate fraction is also related to its abundance in earth's crust (Tokalioglu et al., 2002). Similar results having carbonate fraction as dominant phase of Mn is reported by Horsfall and Spiff (2005), Iwegbue (2007) and Abeh et al. (2007) in Diobu River in Port Harcourt; Niger Delta and Plateau state of Nigeria respectively.

The Mn extracted in acid soluble fraction could come from the dissolution of Mg/Ca carbonates, as Mn^{+2} can substitute for Mg^{+2} as well as Ca^{+2} in carbonate minerals, the more so for Mg in dolomite than for Ca in calcite (Shan and Chen, 1993; Arunachalam et al.,1996). Specific adsorption of Mn at the surface of calcite is also possible (Gleyzes et al., 2002). The exchangeable and carbonate fractions are generally believed to have the greatest tendency towards remobilization from the sediment phase to the more bioavailable pore water phase, and consequently, they pose the most easily induced ecological risk (Fernandes, 1997; Li et al., 2007). It is known that Mn has greater mobilisation and thus could be toxic to the biota. The contribution of Mn with organic phase in pre-monsoon at lower middle estuary might be due to adsorption of Mn to organic matter. It can be the result of continuous accumulation of organic matter from domestic sewage. Mn in lower estuarine sediments shows higher concentration in exchangeable fraction whereas in lower middle and upper middle region it show high values in carbonate fraction during monsoon. In post-monsoon, Mn shows higher concentration in carbonate fraction in lower and lower middle estuary whereas, exchangeable and carbonate fractions in upper middle region. In pre-monsoon, Mn is dominant in carbonate phase in lower estuary whereas, organic, carbonate and exchangeable in lower middle and to some extent in upper middle estuary respectively. The study conducted by Akcay et al. (2003) has demonstrated that Mn exhibits exceptional behaviour by distributing mainly in the unstable form bonded to exchangeable, carbonate and Fe/Mn oxide. The different chemical forms of Mn can affect the solubility and mobility of the metal from sediments. The results indicate that considerable amounts of Mn may be released into the water if conditions become more acidic and /or if the sediments are subjected to more reducing conditions (Yuan et al., 2004). Mn shows highest concentration in bioavailable phase i.e. in carbonate fraction indicating its high availability to biota in post-monsoon season at lower middle estuary. Therefore, bioavailable Mn in the Mandovi estuarine sediments at lower middle estuary is of concern with respect to toxicity. In the present study it is observed that Co is more associated with Fe-Mn oxide fraction in all the seasons and in all the estuarine areas indicating role of the oxides in holding Co. The availability of Co in Fe – Mn oxide fraction have been also reported elsewhere by Calmano and Forstner (1983) and Jones and Turki (1997). This phase accumulates metals from the aqueous system by the mechanism of adsorption and co-precipitation. These processes are very sensitive to change in redox potential and minor variations may greatly affect the concentration within the fraction. This fraction is considered relatively stable, slowly mobile and poorly available but could change with variations in redox conditions. It may become more soluble under reducing conditions and less so under oxidizing ones (Horsfall and Spiff, 2005). Co shows higher bioavailability in lower estuary. Coarser sediments present at this location may be facilitating formation of oxides of Fe-Mn and then accumulation of trace metals.

From the results it is clear that among bioavailable phases, Pb is found to be higher in Fe-Mn oxide fraction in both lower estuary and lower middle estuary whereas, in exchangeable fraction in upper middle estuary in monsoon. In post-monsoon, Pb is found to be higher in Fe-Mn oxide fraction in lower middle estuary and upper middle estuary whereas, bound more to organic fraction in lower estuary. In pre-monsoon Pb is found to be higher in Fe-Mn oxide and exchangeable in lower estuary whereas, in exchangeable in both lower middle and upper middle regions. Pb generally forms stable complexes with Fe – Mn oxide (Ramos et al., 1994; Lopez – Sanchez et al., 1996; Jones and Turki, 1997). The trace metals associated with Fe-Mn fraction include metals weakly adsorbed on sediments or on their essential components namely clays, Fe and Mn hydrated oxides, humic acids, metals that can be released by ion-exchange processes and metals that can be coprecipitated with carbonates present in sediment (Marin et al., 1997; Tokalioglu et al., 2000). Pb received from anthropogenic source must be adsorbed to Fe – Mn oxides in the sediments of Mandovi estuary.

Among the bioavailable fractions Fe-Mn oxide is the main geochemical phase in mudflat sediments of Mandovi estuary. Fe and Mn oxy-hydroxide generally constitutes a sink for trace metals in the aquatic system. This phase accumulates metals from the aqueous system by the mechanism of adsorption and co-precipitation (Bordas and Bourg, 2001). The fact that Pb can form stable complexes with Fe and Mn oxide (Ramos et al., 1994) may be the reason why reducible Pb is more abundant. The metal associated with Fe –Mn oxides may be mobilized under anoxic condition in sediment and subsequently taken up by benthic organisms (Tessier and Campbell, 1987). Changes in the ionic composition, influencing adsorption– desorption reactions, or lowering of pH could also cause remobilisation of metals from this fraction (Marin et al., 1997). According to Rubio et al. (1991), metals with an anthropogenic origin are mainly associated with the exchangeable phase in sequential extraction procedure, and this is the most labile and bioavailable fraction that is most important in determining the metal toxicity of sediment. The high percentage of Pb in exchangeable fraction obtained in the present study therefore signifies anthropogenic source for the metal Pb especially during pre-monsoon in lower middle and upper middle estuary. High exchangeable phase at these locations indicate anthropogenic input of metals through agricultural and industrial waste. Pertsemli and Voutsa (2007) reported Pb association with exchangeable fraction in sediments elsewhere. High organic matter percentage in lower middle estuary helps in metal adsorption. The difference in bioavailability recorded among metals could probably be attributed to the ligand binding strength of each metal (Ongeri et al., 2010).

From the results, it is clear that bioavailability of Mn is higher in lower middle region in all the three seasons followed by upper middle region and lower estuary. In lower middle estuary, the bioavailability of Mn is higher in pre-monsoon with organic fraction as a major phase. In monsoon carbonate is the main phase. During monsoon, metals which are bound strongly to organic fraction transform to less stable carbonate and exchangeable fractions (Zoumis et al., 2001).

Bioavailability of Co is found to be higher in lower estuary. Lower middle estuary with high finer sediments, organic matter content and higher bulk metal concentration is found to facilitate higher bioavailability of metals. The source of metals may be both natural and anthropogenic. Processes such as oxidation of organic matter, bacterial degradation must have affected bioavailability of metals. Further, tides and tidal currents, redistribution of surface sediments must have responsible for changes in chemical properties of sediments that stimulate the mobilisation of metals.

4.4. Pollution level based on bioavailability of metals - Mudflats

4.4a. *SQUIRT*

Table 4A.6. Screening quick reference table for metals in marine sediments (Buchman, 1999)

ELEMENTS	TEL	ERL	PEL	ERM	AET
Fe	-	-	-	-	22% (Neanthes)
Mn	-	-	-	-	260 (Neanthes)
Cu	18.7	34	108	207	390 (Microtox & oyster larvey)
Zn	124	150	271	410	410 (Infaunal community)
Cr	52.3	81	160	370	62 (Neanthes)
Co	-	-	-	-	10 (Neanthes)

TEL: Threshold effect level; ERL: Effect range low; PEL: Probable effects level; ERM: Effect range median; AET: Apparent effects threshold

(Except for Fe, all values are in *u*g/g)

SQUIRT was developed by NOAA for screening purposes. Based on SQUIRT, the guideline values are categorized into five classes which are presented in the Table 4A.6 and 4A.7 (Buchman, 1999).

Table 4A.7. Sediment guidelines and terms used in SQUIRT

Sediment guidelines	
Threshold Effect Level (TEL)	Maximum concentration at which no toxic effects are observed
Effects Range Low (ERL)	10th percentile values in effects or toxicity may begin to be observed in sensitive species
Probable Effects Level (PEL)	Lower limit of concentration at which toxic effects are observed
Effects Range Median (ERM)	50th percentile value in effects
Apparent effects Threshold (AET)	Concentration above which adverse biological impacts are observed

In lower estuary, lower middle (Table 4A.8b) and upper middle (Table 4A.8c) estuary the percentage of Fe in bulk sediments is very low compared to apparent effect threshold (AET) indicating no harm to biota from this element and Fe is higher in the residual fraction in all the three seasons indicating less bioavailability of Fe.

There are no reported values for Mn and Co for first four classes. Both in bulk sediments and bioavailable, Mn shows the values above AET suggesting that Mn is potentially bioavailable and thus toxic to the environment in all the three seasons except for bioavailable phase in lower estuary. Co shows values more than AET in all the three seasons except in lower estuary during post-monsoon and pre-monsoon seasons where Co is observed to be less than AET value.

The values of Zn in total and sum of bio available fractions fall below TEL indicating not much risk to biota due to this metal (Table 4A.8a) irrespective of seasons and different estuarine regions.

Table 4A.8a. Average concentration of total metals and bioavailable fractions in lower estuary in the three different seasons

Lower estuary

Metals	Monsoon	Post monsoon	Pre monsoon	Monsoon	Post monsoon	Pre monsoon
	Total metals			Bio available metals (ppm)		
Fe (%)	1.67	1.79	1.64	285.72	253.9	1111.8
Mn (ppm)	263.33	284.25	335.3	110.3	207.21	208.2
Co (ppm)	11.77	8.91	9.00	10.83	7.98	8.72
Zn (ppm)	38.78	33.75	35.33	12.94	15.13	16.34
Pb (ppm)	63.75	28.58	31.75	31.70	22.19	28.74

Table 4A.8b. Average concentration of total metals and bioavailable fractions in lower middle estuary in the three different seasons

Lower middle estuary

Metals	Monsoon	Post monsoon	Pre monsoon	Monsoon	Post monsoon	Pre monsoon
	Total metals			Bio available metals (ppm)		
Fe (%)	5.99	6.25	5.91	1848	6384	16113
Mn (ppm)	2585.7	2731.6	2752.6	2518	2421	1836
Co (ppm)	28.46	29.25	27.58	17.1	24.99	21.54
Zn (ppm)	83.00	75.41	75.00	36.25	47.77	32.47
Pb (ppm)	78.69	88.08	48.08	54.20	58.91	38.94

Table 4A.8c. Average concentration of total metals and bioavailable fractions in upper middle estuary in the three different seasons

Upper middle estuary

Metals	Monsoon	Post monsoon	Pre monsoon	Monsoon	Post monsoon	Pre monsoon
	Total metals			Bio available metals (ppm)		
Fe (%)	7.70	6.48	5.84	1729	5683	9768
Mn (ppm)	1945.0	2562.3	2266.1	1208	2267	1713
Co (ppm)	28.0	30.08	28.33	22.82	17.61	25.24
Zn (ppm)	81.92	62.08	63.92	42.28	37.08	33.64
Pb (ppm)	72.92	87.41	73.75	25.59	57.40	38.2

The results of the speciation of metals in the mudflat sediments of Mandovi estuary indicate that the concentration of metals associated with fractions 1 to 4 differ considerably between the locations. It is known that chemical elements introduced due to human activity show greater mobility and are associated with sediment phases, viz carbonates, oxides, hydroxides and sulphides (Heltai et al., 2005). Among all the elements studied, Mn and Co are more bioavailable and higher in middle estuarine regions. Further, bioavailability of Mn is higher in lower middle estuary in all the three seasons compared to upper middle region. Bioavailability of Co is found to be relatively higher in upper middle region during monsoon and pre-monsoon, whereas it is higher in lower middle region in post-monsoon season. Bioavailability of metals depends on the nature of sediment particles associated, the mechanisms of metal release from sediments and variation of exposure routes (Chapman et al., 1998). Higher bioavailability of Mn and Co may affect benthic fauna as they have ability to accumulate metals (Burgos and Rainbow, 2001). Bioavailability of other studied metals is less, however, their value varies between locations and seasons indicating the role played by associated factors like sediment size, organic matter, salinity, pH and Eh.

4.4b. *Risk Assessment Code (RAC)*

Risk Assessment Code (RAC) has been used to assess the potential mobility and hazard of metal based on the percentage of exchangeable and carbonate fractions of metals within the sediment (Perin et al., 1985; Jain, 2004; Ghrefat and Yusuf, 2006). The metals in the sediments are bound with different strengths to the different fractions. The metals in acid soluble fraction are considered to be the weakest bonded metals in sediments which may equilibrate with the aqueous phase, and thus become more easily bioavailable (Pardo et al., 1990). The risk assessment code (RAC) proposed by Perin et al. (1985), mainly applies to the sum of exchangeable and carbonate bound fractions for assessing the availability of metals in sediments. If sediment samples can release in these fractions less than 1% of the total metal it will be considered safe for the environment. On the contrary, sediment releasing in the same fractions is more than 50% of the total metal then it is considered highly dangerous as it can easily enter into the food chain (Table 4A.9). Thus, metal speciation is of critical importance to their potential toxicity and mobility (Maiz et al., 2000).

Table 4A.9. Criteria of Risk Assessment code (Perin et al., 1985)

Grade	Sum of exchangeable and carbonate in % of the total	RAC
I	<1	No risk
II	1- 10	Low risk
III	11 – 30	Medium risk
IV	31 – 50	High risk
V	>50	Very high risk

RAC applied to the present study revealed that Mn in lower estuary (Table 4A.10a) shows grade IV during monsoon and V during post-monsoon and pre-monsoon seasons indicating high risk to very high risk with respect to this metal to biota. In lower middle estuary (Table 4A.10b) Mn is under grade V during monsoon and post-monsoon and IV in pre monsoon whereas, in upper middle (Table 4A.10c) estuary it falls in grade V in both monsoon and post-monsoon and III during pre-monsoon. Towards lower estuarine region, Co falls under grade between IV in monsoon and pre-monsoon and III in post-monsoon season. In lower middle estuarine region, Co shows medium risk (III) during monsoon and pre-monsoon and high risk (IV) during post-monsoon whereas, in upper middle estuary Co falls under grade III indicating medium risk. Pb exhibits grade III in lower estuary irrespective of seasons. In lower middle estuary, Pb exhibits grade III in both monsoon and post-monsoon and IV in pre-monsoon season. Pb exhibits grade III in upper middle estuary in all the three seasons.

Table 4A.10a. Sum of exchangeable and carbonate fractions in percentage

a) Lower estuary			
Metals	Monsoon	Post monsoon	Pre monsoon
Mn	45.95 (Grade IV)	52.65 (Grade V)	54.41 (Grade V)
Co	36.96 (Grade IV)	26.59 (Grade III)	32.49 (Grade IV)
Pb	17.62 (Grade III)	20.32 (Grade III)	29.27 (Grade III)

Table 4A.10b. Sum of exchangeable and carbonate fractions in percentage

a) Lower middle estuary			
Metals	Monsoon	Post monsoon	Pre monsoon
Mn	78.20 (Grade V)	74.30 (Grade V)	48.27 (Grade IV)
Co	16.67 (Grade III)	31.48 (Grade IV)	26.92 (Grade III)
Pb	23.19 (Grade III)	20.87 (Grade III)	42.16 (Grade IV)

Table 4A.10c. Sum of exchangeable and carbonate fractions in percentage

b) Upper middle estuary			
Metals	Monsoon	Post monsoon	Pre monsoon
Mn	51.85 (Grade V)	71.04 (Grade V)	21.49 (Grade III)
Co	26.84 (Grade III)	20.12 (Grade III)	19.29 (Grade III)
Pb	18.97 (Grade III)	24.92 (Grade III)	29.03Grade III)

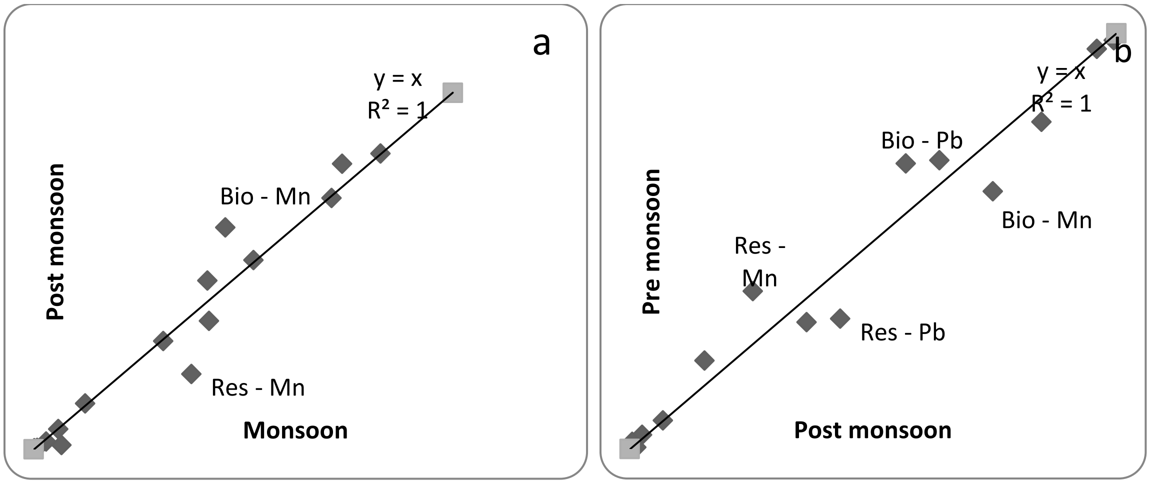
From the results detailed above, it is clear that Mn largely available in the labile fraction and therefore, falls under the very high risk to high risk grade and can easily enter the food chain.

Biota in sediments is at medium risk with respect to Pb with exception observed in lower middle region wherein high risk is noted in pre-monsoon season. In general, higher amount of metal in exchangeable form increases leaching potential. Co shows higher concentration in exchangeable and carbonate fractions and can cause deleterious effects as it is associated with labile fractions. However, it is important to note that sequential extraction results do not necessarily prove bioaccumulation of metals but merely reflect the chemical behaviour and bioavailability of metals within sediments (Coetzee, 1993). Therefore, along with understanding bioavailability of metals, determining bio-accumulation of metals is essential to know the risk of bioavailability on to the benthic biota.

4.5. Variation in bioavailability between seasons - Mudflats

In order to understand the variation in bioavailability between different seasons at different locations of Mandovi estuary within mudflats with respect to sediment parameters and metal species, the data is plotted on the Isocon diagram (Grant, 1986). Residual and bioavailable fractions of different metals are computed for which bioavailable fraction is obtained by adding first four fractions (exchangeable, carbonate, Fe- Mn oxide, organic bound metals). The parameters viz sand, silt, clay, organic carbon, pH and total bioavailable and residual fractions of metals (Fe, Mn, Co, Zn and Pb) are plotted.

Lower estuary



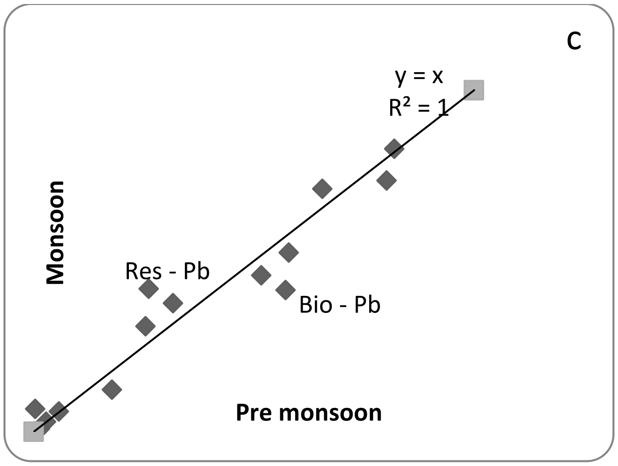


Fig 4A.5. Isocon plot for samples collected from mudflats of lower estuary in different seasons

The data points of sand, silt, clay, organic carbon, pH, bio- Fe, bio-Co, bio-Zn and bio-Pb, res – Fe, res- Co, res- Zn and res – Pb fall on or close to isocon lines indicating lesser variation (Fig 4A.5) between seasons. When the data of monsoon and post-monsoon is compared it is observed that bio – Mn is prominent in post-monsoon and res – Mn in monsoon (Fig 4A.5a). This reveals that higher amount of residual fraction is added in monsoon possibly due to lithogenic source at this location. Higher bio-Mn in post-monsoon indicates its high mobility during this season. Close association of Mn with carbonates (Dassenakis et al., 2003) is well known as endorsed by other workers (Kiratli and Ergin, 1996; Morillo et al., 2004). In this phase, weakly sorbed Mn retained on sediment surface by relatively weak electrostatic interactions is released by ion exchange processes and results in dissociation of Mn-carbonate phase (Tessier et al., 1979).The tendency towards the remobilization from the sediment phase to the more bioavailable pore water phase, could pose induced ecological risk (Li et al., 2007).

When the data of post-monsoon and pre-monsoon (Fig 4A.5b) is compared it is observed that, res – Mn and bio – Pb is higher in pre-monsoon and res – Pb and bio – Mn is prominent in post-monsoon. Fe – Mn oxide generally regulates the Pb distribution and most of anthropogenic Pb is accumulated onto Fe – Mn oxides in sediments (Li et al., 2001).

Data plotted between pre-monsoon and monsoon (Fig 4A.5c) also shows, higher bio-Pb in pre-monsoon and res – Pb in monsoon. Fe and Mn species are reduced during early diagenesis by microbially mediated redox reactions (Canfield, 1989). Bio-Pb associated with Fe-Mn oxide can be released to pore water and possibly to the overlying water column (Petersen et al., 1995) and can be available to benthic biota (Jones and Turki, 1997).

Lower middle estuary

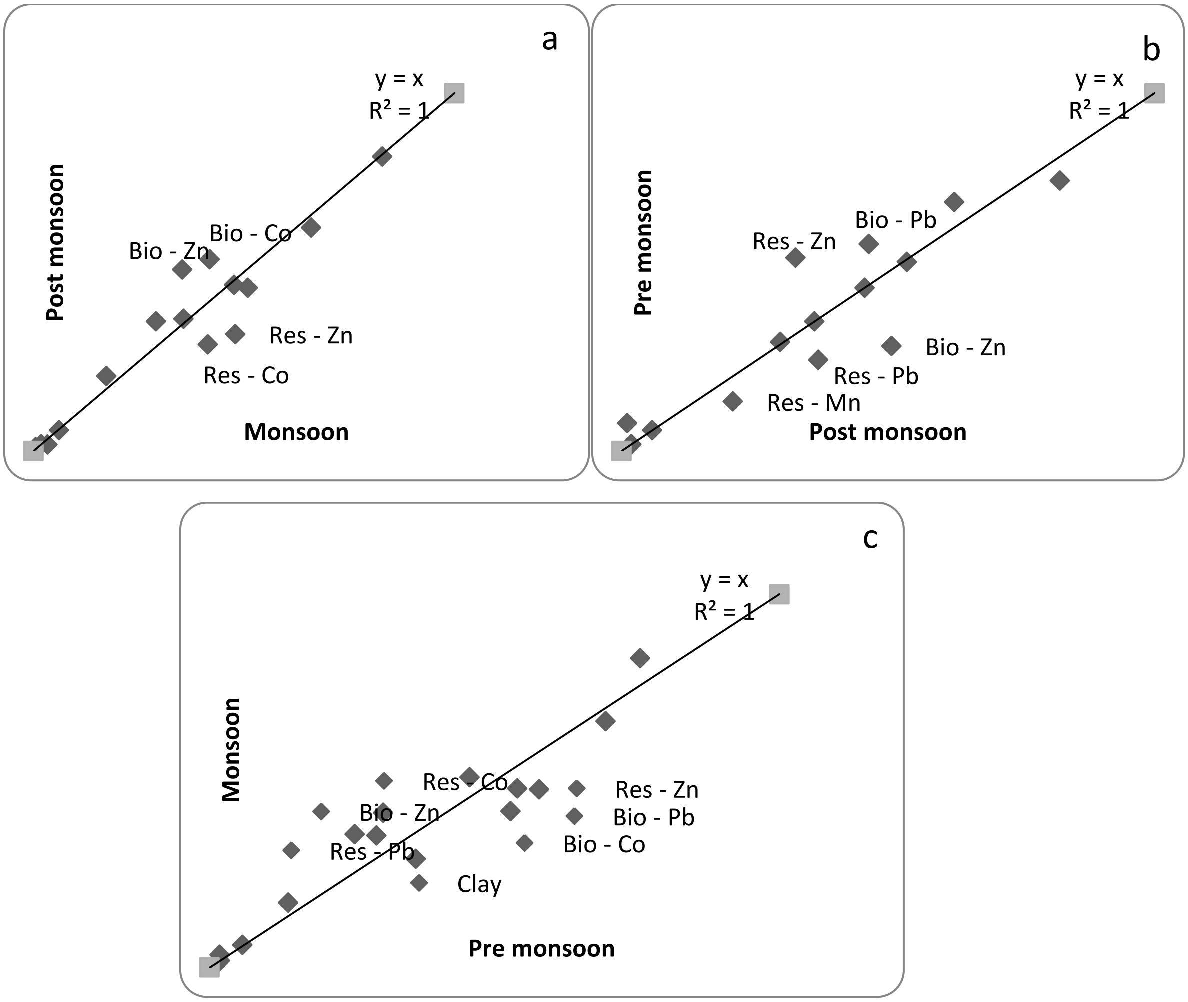
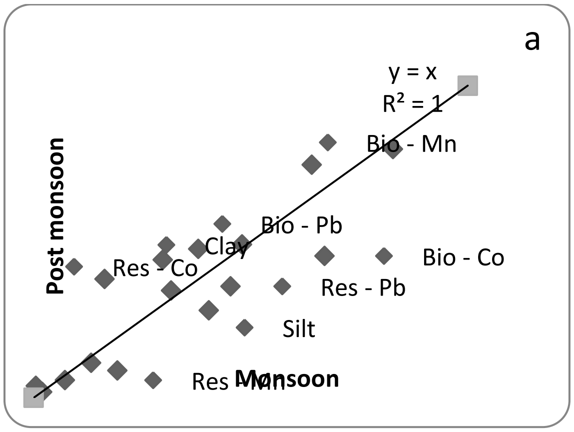


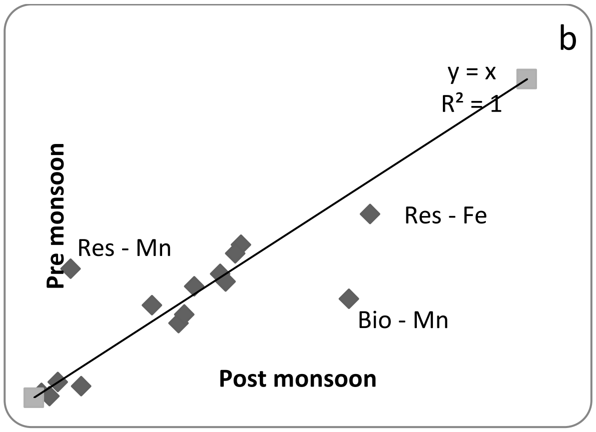
Fig 4A.6. Isocon plot for samples collected from mudflats of lower middle estuary in different seasons

In lower middle estuarine region, when sediments of monsoon and post-monsoon (Fig 4A.6a) are compared it is observed that bio – Co and bio – Zn is more pronounced in post-monsoon whereas, res – Co and res- Zn is prominent in monsoon. Residual phases of metals are generally much less toxic for organisms in aquatic environment. Mandovi estuary shows slow increase of salinity in post-monsoon from fresh water conditions of monsoon. The change in salinity and pH must have facilitated change in metal forms and their bioavailability. In general, the concentration of the free ion for all the metals increases with the decrease of the salinity values and decrease of the bioavailability of these metals at higher values of salinity in the estuary (Riba et al., 2005). Other data points lie on or close to isocon line indicating not much change. When the data of post-monsoon and pre-monsoon (Fig 4A.6b) are compared it is observed that bio – Pb and res – Zn is prominent towards the pre-monsoon and bio – Zn, residual fraction of Mn and Pb in post-monsoon season. During post-monsoon, higher bio – Zn is observed as threat to the benthic organisms as Zn can be easily mobilized and is bioavailable to the biotic community when the pH and redox conditions of the sediment changes (Li et al., 2001).

When the data of pre-monsoon and monsoon (Fig 4A.6c) season is compared it is observed that bio – Co, bio – Pb, res – Zn and clay are higher during pre-monsoon whereas, bio – Zn and res – Pb and res- Co are prominent in monsoon season. Presence of finer sediments during pre-monsoon possibly controls the deposition of anthropogenic bioavailable Pb and Co.

Upper middle estuary





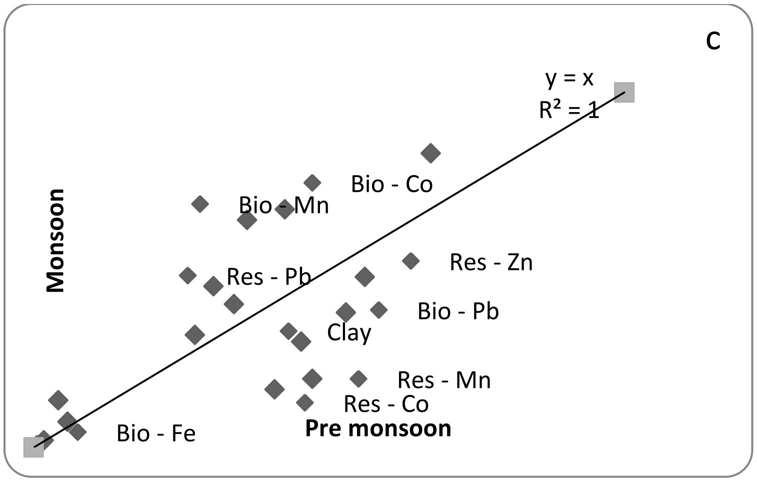


Fig 4A.7. Isocon plot for samples collected from mudflats of upper middle estuary in different seasons

In the upper middle estuarine region when the data of monsoon and post monsoon (Fig 4A.7a) is compared some of data points fall on or close to isocon line indicating less variability. However, bio – Mn, bio – Pb, res - Co along with clay re slightly higher in post monsoon whereas, res – Mn, res – Pb, bio – Co and silt are prominent in the monsoon season. Large amount of lithogenic material brought from the catchment area during monsoon, supports their mineral bound or residual phase. Mn and Pb of anthropogenic origin might be settling down slowly and are remobilized during early diagenesis.

When the sediments of post and pre-monsoon (Fig 4A.7b) are compared higher bio – Mn is noted during post monsoon. Mn present in exchangeable and carbonate phase which makes it more easily available to biota.

When the data of pre-monsoon and monsoon (Fig 4A.7c) is compared it is observed that bio – Co and bio - Mn along with residual Pb are predominant in monsoon season. Clay, res Mn, res – Co, bio – Pb are prominent in pre-monsoon. Pb must be held within clay mineral lattice and Mn and Co must be of anthropogenic origin. Mn and Co bioavailability during monsoon indicates that these metals are potentially available for exchange and/or release into the environment. This also suggested that large part of these metals introduced by human activities (Singh et al., 2005; Jain, 2004). High amounts of residual fraction of Mn and Co during pre-monsoon indicate that these metals in the sediment occur in resistant minerals and therefore are unlikely to be released to the pore waters through dissociation (Yuan et al., 2004).

CHAPTER FIVE

SUB-SURFACE SEDIMENTS

(VARIATION WITH DEPTH)

5.1. Sediment Components

The abundance and distribution of sediment components namely sand, silt, clay, organic carbon, selected metals in bulk sediments and total heterotrophic counts with depth in mudflats and mangroves in different regions of Mandovi estuary are discussed in this chapter. Also, metal species and metal tolerant microbes in sediments are discussed.

5.1A. Mudflats

Though the sediment components and metals were studied at 2 cm interval in sediment cores, to compare with heterotrophic counts and bacterial metal tolerance, results in this chapter are grouped into 0- 6 cm, 6 – 12 cm and 12 – 20 cm depths.

Results of different parameters namely sand, silt, clay, organic carbon, pH for three sections of sediments viz. 0-6 cm, 6-12 cm and 12 – 18 cm from lower estuary are shown in the (Table 5A.1a). It is observed that the percentage of sand is high (>95 %) in all the three sections with lesser values noted for finer sediments and organic carbon for this core (Table 5A.1a). Relatively silt is slightly higher in middle section (4.01 %) along with organic carbon (0.25 %). Clay concentration is less than that of silt and is found to be higher at surface (0.56 %). Fe, Mn and Al concentration for different sections with depth are presented in (Table 5A.2) and that of trace metals are presented in Figure 5A.1a. Fe and Mn concentration increases towards surface with higher values in surface sediments i.e. 0-6 cm (Table 5A.2a) compared to sediments at depths. Al however, shows higher value in the middle section. Among trace metals Cr and Cu show higher concentration followed by Zn, Ni, Pb and Co. Trace metals, Zn, Ni and Co show less variation with depth. Pb shows higher concentration in surface and middle sections whereas, Cr and Cu show higher concentration in bottom section (Fig. 5A.1a). The heterotrophic counts are found to be more in the middle and surface sections and least counts are seen (Table 5A.3a) in bottom sections.

The sediment core collected from lower middle estuary shows higher concentration (>94 %) of finer sediments i.e. silt and clay with less (<6 %) of coarser sediments (Table 5A.1b). Silt is higher (49.16 %) in surface section along with high organic carbon (2.16 %). Clay shows slightly higher value (49.39 %) in middle section. pH shows an increase towards surface. Fe, Mn and Al concentration for different sections with depth are presented in Table 5A.2b and that of trace metals are presented in Figure 5A.1b. Fe and Mn shows higher concentration in surface sediment section whereas, Al shows high value in bottom section (Table 5A. 2b). Among trace metals, as in lower estuary, Cu and Cr are relatively high in concentration, followed by Zn, Ni, Pb and Co. Cr and Cu show slightly higher concentration in surface sediments compared to middle and bottom sediment sections (Fig. 5A.1b). Zn shows slightly higher value in the middle section, whereas, Ni in bottom sediments. Not much variation noted in distribution of Co and Pb in different sediment sections. The heterotrophic counts are higher in surface sediments with least noted in bottom sediments (Table 5A.3b).

The results of different sediment sections in mudflats from upper middle estuary show predominance of finer sediments (>93 %). Silt concentration is higher (>53 %) than clay concentration (<42 %). Silt is found to be high at the surface sediment section along with high organic carbon (2.40 %) whereas, the concentration of clay shows slightly higher value in the middle sediment section (Table 5A.1c). Fe, Mn and Al concentration for different sections with depth are presented in Table 5A.2c and that of trace metals are presented in Figure 5A.1c. Fe and Mn show higher concentration in surface sediments (Table 5A.2c). Al however shows higher value in middle section. Among trace metals Cu shows highest concentration followed by Cr, Zn, Pb, Ni and Cu. Cr, Co and Zn do not show much variation with depth. Cu and Pb show increasing trend towards surface with high concentration in surface sediments. The distribution of heterotrophic counts in upper middle estuary is found to be similar to that of lower middle with highest counts in surface sediments and least counts obtained for bottom sediments (Table 5A.3c)

Table 5A.1. Distribution of sediment components within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)

Lower estuary (a)					
Sediment	Sand (%)	Silt (%)	Clay (%)	Organic	pH

sections				carbon (%)	
0-6 cm	95.93	3.50	0.56	0.20	6.89
6-12 cm	95.60	4.01	0.38	0.25	6.83
12-18 cm	96.89	2.81	0.30	0.23	6.89

Lower middle estuary (b)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	5.50	49.16	45.33	2.61	6.81
6-12 cm	5.40	45.20	49.39	2.11	6.77
12-18 cm	5.57	47.81	46.61	2.17	6.20

Upper middle estuary (c)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	3.17	55.82	41.00	2.40	6.49
6-12 cm	4.65	54.08	41.27	2.14	6.50
12-20 cm	6.18	53.56	40.26	1.90	6.37

Table 5A.2. Distribution of metals (Fe, Mn and Al) within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)

Lower estuary (a)			
Sediment	Fe (%)	Mn (ppm)	Al (%)

sections			
0-6 cm	1.63	335.3	6.26
6-12 cm	1.54	276.5	6.88
12-18 cm	1.53	223.3	5.78

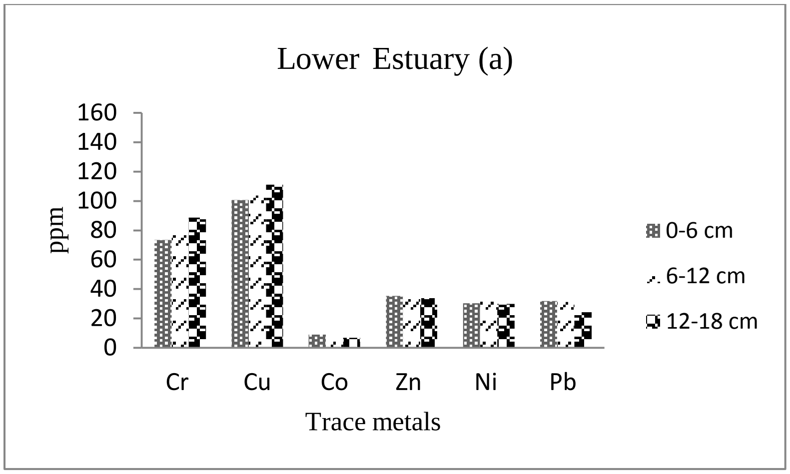
Lower middle estuary (b)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	5.91	2752.6	8.20
6-12 cm	5.55	2152.9	8.26
12-18 cm	5.61	2270.1	8.40

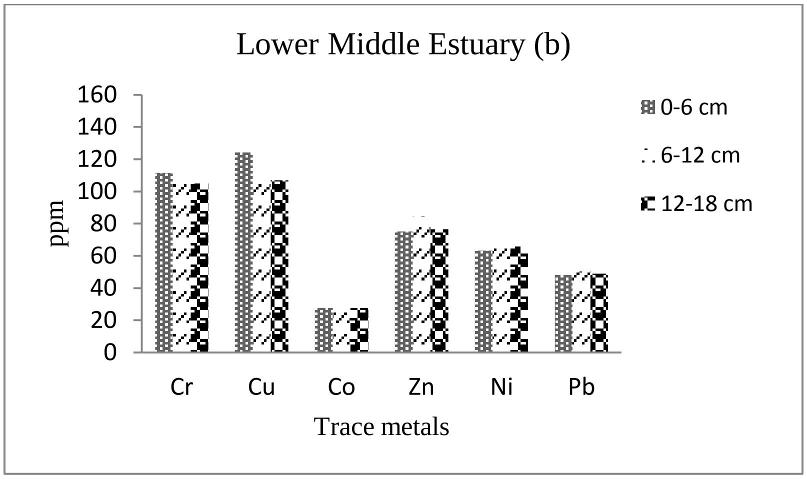
Upper middle estuary (c)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	5.84	2266.1	7.44
6-12 cm	5.77	1739.8	7.83
12-20 cm	5.68	1652.8	6.83

Table 5A.3. Total heterotrophic bacterial distribution (CFU g^{-1} sediment) within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)

Sediment	Lower	Lower middle	Upper middle

sections	estuary (a)	estuary (b)	estuary (c)
0-6 cm	2.60E+04	4.00E+06	3.90E+06
6-12 cm	2.70E+04	3.70E+06	3.70E+06
12-18 cm	1.60E+04	2.00E+06	1.90E+06





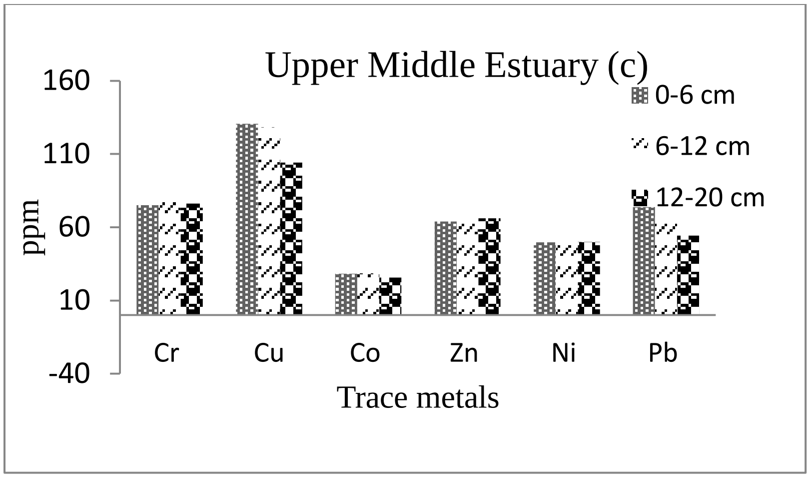


Fig. 5A.1. Distribution of metals (Cr, Cu, Co, Zn, Ni and Pb) within mudflats in different sections of the cores collected at lower estuary (a), lower middle estuary (b) and upper middle estuary (c)

From the results described above, it can be stated that sand is the predominant sediment component at different depths in lower estuary. Deposition of larger size sediments represents prevailing higher energy conditions. Relatively high tidal energy towards lower estuary results in higher hydrodynamic conditions near the mouth which facilitates the deposition of coarser sediments. Chen (1992) highlighted the role of hydrodynamics in retaining coarser sediments within intertidal mudflats near river mouth. In the middle estuary sediment distribution shows predominance of finer sediments associated with high organic matter. Presence of higher wave activity near estuarine mouth which results in complex energy conditions and facilitates deposition of coarser sediments also results in carrying finer particles to areas where wave activity is relatively less i.e. relatively calmer environment as discussed earlier in Chapter 4. In the calmer environments finer sediments slowly settle to the bottom. Deposition of organic carbon contents generally depend upon the grain size and are usually associated with fine grained sediments (Falco et al., 2004). The data of mud proportion (silt + clay) shows higher concentration in both lower middle and upper middle estuarine sediments. However, higher mud concentration is observed in surface (96.82 %) and middle (95.35 %) depth sections at upper middle estuary and bottom sediments of lower middle estuary (94.43 %). To compensate this, coarser sediments (sand) is higher in bottom sediments i.e. 12 – 20 cm in upper middle estuary and at 0 - 6 cm and 6 – 12 cm sections of lower middle estuarine region. Further, lower middle and upper middle estuarine regions show considerable difference in distribution of finer sediment components. Clay fraction is found to be higher in lower middle estuarine region in all the sediment depth sections whereas, in upper middle estuarine region silt is more. Higher organic carbon content is noted in surface sediments of both lower middle and upper middle estuarine regions. In case of upper middle it increased from bottom to surface. Increase in organic matter towards surface obtained for Pearl Estuary (Qi et al., 2010) was related to the decomposition of organic matter during the sediment diagenesis. As mentioned earlier in Chapter 4 upper middle estuary is part of main estuarine channel, whereas, location of lower middle estuary is within sub channel of Mandovi estuary. Possibly because of quiet environment prevailed in the sheltered sub-channel, lower middle estuary must be harbouring deposition of more clay and organic carbon.

Systematic variation in grain size with sand decreasing and finer sediments increasing from lower estuary to middle estuary indicate changing depositional environment from relatively higher dynamics to lesser hydrodynamics environment. Upper middle estuarine region being in the main channel is affected more by river channel processes and therefore retained higher silt percentage. Higher organic matter and clay deposition in the surface sediments in lower middle estuary is facilitated by calmer environment of the sub channel. The organic matter brought by terrestrial source including mangrove litter must have resulted in increasing of organic matter in lower middle estuarine region (Jonathan et al., 2004). Further, organic matter decrease with depth must be due to extensive degradation of organic matter through microbial oxidation of organic matter (Anawar et al., 2010).

In Mandovi estuary, both in lower middle and upper middle estuarine regions, metals especially Fe and Mn concentration are found to be higher in surface sediments and it increased from bottom section to surface section, indicating increased addition in recent years might be due to anthropogenic activities like open cast mining, land use, land cover and agricultural practices. However, in lower middle region slightly lower values of Fe and Mn concentration in middle section is noted. Increase of Fe and Mn in the surface may also be due to diffusion of these metal ions into the suboxic zone which must have facilitated precipitation of Fe and Mn as oxides (Ayyamperumal et al., 2006). Severmann et al. (2008) reported that the manganese and iron are mobilized from hypoxic sediments to more oxic settings where oxidation, precipitation and thus settling occur. In addition, activities of benthic organisms might have resulted in enhanced transport of oxygen into the sediments and consequently enhancing the rates of aerobic respiration and thus results in deposition of both Fe and Mn in oxic layers (Archer and Devol, 1992). Organisms can also alter sediment porosity, permeability, cohesion, and organic content and thus alter cycling of redox elements (Van Nugteren et al., 2009). All or some of these processes must have contributed to enrichment of Fe and Mn in surface sediments. However, distribution pattern of Fe and Mn are not agreeing with that of Al and therefore Fe and Mn must be from sources other than from mineral lattice.

Cr and Cu observed to be higher in surface sediments of lower middle estuary whereas, Pb and Cu are observed to be higher in surface section of upper middle estuary. Other trace metals studied show less variation with respect to different sediment depths. However, some metals viz Cr and Cu in lower estuary, show slightly higher concentration in bottom section of sediment core. Similarly Zn in lower middle estuarine region show higher concentration in middle section. Higher metal concentrations at bottom sediment section in lower estuary indicate their association with higher organic matter at this location. Similarly, in the lower middle estuary more clay and organic matter must be responsible for retaining high Zn.

Metals might be natural or anthropogenic source or both must be contributing to Mandovi estuary. Sediments in the middle estuary show more metal concentration than that of sediments lower estuary, attributing largely to higher finer sediments and organic matter availability which can retain more metals. Higher Cu towards surface must be the result of its association with organic matter (Jones and Turki, 1997). It is known that Pb can form stable complexes with Fe and Mn dioxide (Ramos et al., 1994). Higher Fe and Mn in the surface sediment might have facilitated Pb adsorption. Source for Pb is mainly considered as from non-point sources such as atmospheric transport and precipitation resulted from coal burning and other industrial activities. In the areas influenced by extensive human activities, a considerable proportion of Pb of anthropogenic origin is present in the environment. Distribution of other metals like Zn, Co and Ni must be regulated by process of remobilisation from sediment to water which occurs under anoxic conditions and reverses when the water phase become oxic (Petersen et al., 1995).

Middle estuarine sediments show high heterotrophic counts than observed in lower estuary. This probably could be due to availability of higher organic matter in the middle estuarine region which might have facilitated growth of the bacterial abundance. Violent hydrodynamic conditions prevailing at lower estuary must be creating an unfavourable environment for their proliferation and abundance. However, counts are relatively higher in the surface and middle sections at lower estuary. Bacterial counts are considerably high in the surface sediments in middle estuarine regions. In general, bacterial abundance and total activity decrease with increasing sediment depth. Fisher et al. (2002) stated that bacterial activity and biomass are generally highest near the sediment surface and in general decrease with depth**.** There are several factors which play an important role influencing the distribution of microbes. Sediment grain sizes, amount of organic matter, pH, Eh, metal contents in the sediment column are some of the important factors. Generally, microbial populations are more abundant in muddy sediments than in sandy ones depending on the granulometry of particles (Lakshmanaperumalsamy et al., 1986). In aquatic ecosystems, the flux of organic matter to the bottom sediments depends on primary productivity in the water column and on water depth. The number of bacterial cells is usually high where waters are not deep and where there are large numbers of organisms present. Under these conditions, leaves and other plant and animal residues decay and settle on the bottom before metabolization. This represents a good nutritional substrate for heterotrophic bacteria and favours bacterial growth. Distribution of bacteria also depends on changes in water temperature, salinity and other physicochemical parameters (Alavandi, 1990). Higher number of total heterotrophic counts in lower middle and upper middle estuarine regions might be due to stable conditions of nutrients which coincide with higher primary productivity (Santhanam, 1976).

5.1B. Mangroves

Sand, silt, clay, organic carbon and pH values are presented in Table 5B.1a. In sediment sections viz 0-6 cm, 6-12 cm and 12-20 cm of lower estuary sand and silt show higher values in 0-6 cm whereas, clay in 12 – 20 cm sediment section. Sand and silt show increasing trend from bottom to surface sediments whereas, clay show decreasing trend. Organic carbon is found to be higher in surface sediment section and lesser noted in bottom sediment sections (Table 5B.1a). Not much variation is noted in pH value between sections. Fe, Mn and Al are found to be higher in bottom sediment section (Table 5B.2a). Among trace elements, Cu and Ni and to some extent Co show slightly higher value in surface sediments whereas, Cr, Zn and Pb show higher value in bottom sediments (Fig. 5B.1a). Total heterotrophic counts are found to be higher in middle section with least counts observed at the surface (Table 5B.3a).

In lower middle estuary, sand content is less in all the three sections (Table 5B.1b). The finer sediments are higher in all the three sections with clay noted to be higher than 50 %. Not much change is noted in pH, however organic carbon shows increasing trend towards surface. Fe, Mn and Al are higher in bottom sediments (Table 5B.2b) and decrease towards surface. Among trace metals Cu, Pb and Co are higher in surface sediments. Cr is higher in bottom sediments (Fig. 5B.1b). Zn and Ni show less variability between sediment sections. Bacterial counts are found to be higher in middle section with least heterotrophic counts recorded in bottom sediment section (Table 5B.3b).

In sediments of upper middle estuary, sand shows higher concentration in bottom sediment. Silt is the predominant sediment component, which increased towards surface. Organic carbon increased from bottom section to surface. pH shows not much variation (Table 5B.1c). Fe and Al are found to be higher in middle section and Mn is higher in bottom sediments (Table 5B.2c). Cr, Cu, Co and Ni show higher concentration in middle sediment section (Fig 5B.1c). Zn and Pb show higher value in surface sediments. Distribution of total heterotrophic counts shows its dominance in surface sediments (Table 5B.3c).

In upper estuary, silt and clay show higher concentrations with silt showing highest concentration at surface sediments and clay in the bottom sediments. Sand, organic carbon and pH (Table 5B.1d) did not show much variation with depth. Organic carbon is found to be higher in all the three sediment sections with highest concentration noted in bottom sediments. Fe and Al are found to be highest in middle sediment section whereas, Mn is dominant in surface sediments (Table 5B.2d). Not much change noted in trace metal concentration with slightly high concentration noted in bottom sediments in case of Cr, Co and Ni, rest of the elements show slightly higher concentration in surface sediments (Fig 5B.1d). Heterotrophic counts are found to be higher in surface sediments with least noted in middle sediment section (Table 5B.3d).

Table 5B.1. Distribution of sediment components within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)

Lower estuary (a)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	25.06	55.6	19.33	2.29	6.74
6-12 cm	21.7	51.3	27.00	2.27	6.62
12-20 cm	16	14.96	48.64	1.90	6.78

Lower middle estuary (b)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	1.80	46.86	51.33	2.53	6.26
6-12 cm	1.82	48.17	50	2.00	6.91
12-22 cm	1.93	44.73	53.33	1.76	6.40

Upper middle estuary (c)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	14.73	52.60	32.66	2.94	6.33
6-12 cm	12.97	52.02	35.00	2.77	6.43
12-22 cm	19.26	47.40	33.33	1.67	6.61

Upper estuary (d)					
Sediment sections	Sand (%)	Silt (%)	Clay (%)	Organic carbon (%)	pH
0-6 cm	4.73	57.26	38.00	2.24	6.22
6-12 cm	6.35	49.65	44.00	2.13	6.16
12-18 cm	3.65	48.35	48.00	2.42	6.14

Table 5B.2. Distribution of metals (Fe, Mn and Al) within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)

Lower estuary (a)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	5.28	1317.5	6.14
6-12 cm	5.11	1428.5	6.33
12-20 cm	5.80	1884.7	6.74

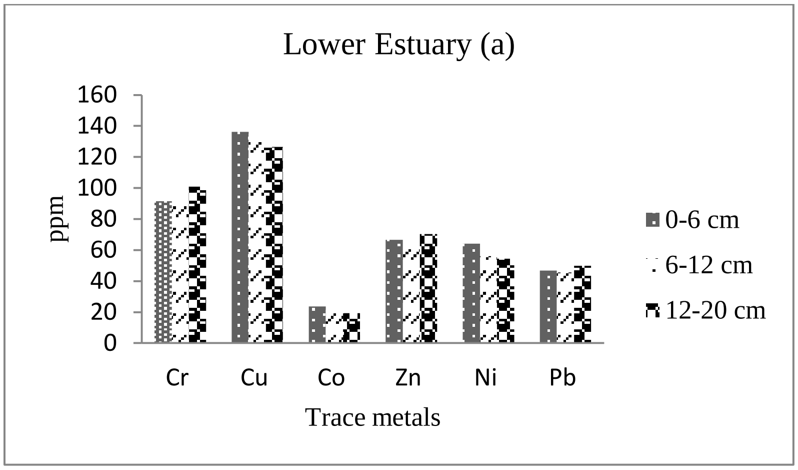
Lower middle estuary (b)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	5.20	2980.8	8.32
6-12 cm	5.73	3093.1	8.60
12-22 cm	6.98	3680.2	8.86

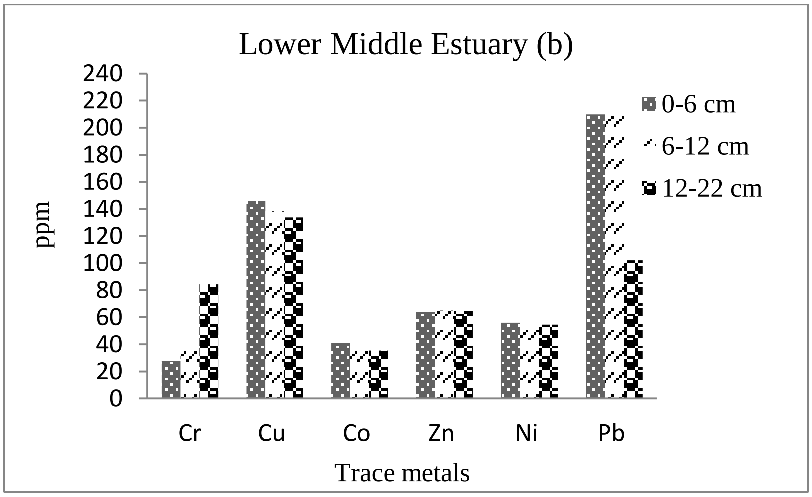
Upper middle estuary (c)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	6.36	1860.3	6.68
6-12 cm	6.43	2379.0	7.54
12-22 cm	6.22	2768.6	7.33

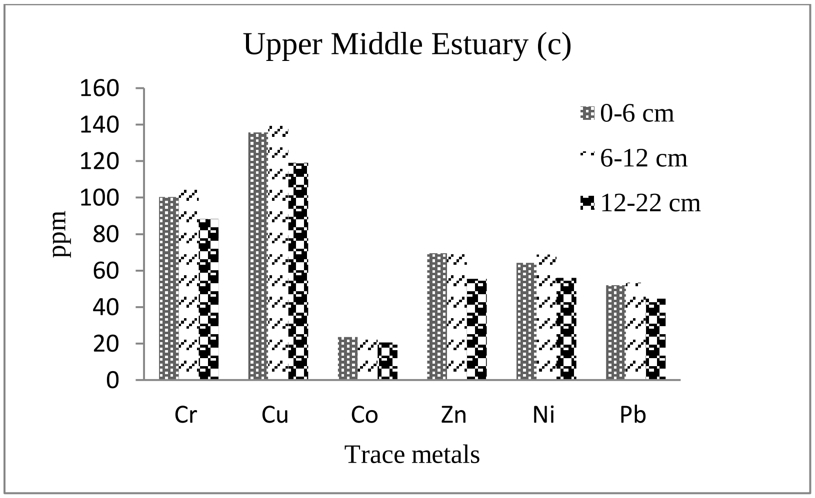
Upper estuary (d)			
Sediment sections	Fe (%)	Mn (ppm)	Al (%)
0-6 cm	6.97	2916.5	7.39
6-12 cm	7.73	2482.5	7.84
12-18 cm	7.69	2631.5	7.65

Table 5B.3. Total heterotrophic bacterial distribution (CFU g^{-1} sediment) within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)

Sediment sections	Lower estuary	Lower middle estuary	Upper middle estuary	Upper estuary
0-6 cm	2.00E+08	3.00E+08	4.50E+08	3.20E+07
6-12 cm	5.50E+08	4.20E+08	4.00E+08	1.90E+07
12-18 cm	4.20E+08	2.20E+08	3.00E+08	5.00E+06


Lower Estuary (a)
ppm
160
140
120
100
80
60
40
20
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals
0-6 cm
6-12 cm
12-20 cm


Lower Middle Estuary (b)
ppm
240
220
200
180
160
140
120
100
80
60
40
20
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals
0-6 cm
6-12 cm
12-22 cm


Upper Middle Estuary (c)
ppm
160
140
120
100
80
60
40
20
0
Cr
Cu
Co
Zn
Ni
Pb
Trace metals
0-6 cm
6-12 cm
12-22 cm

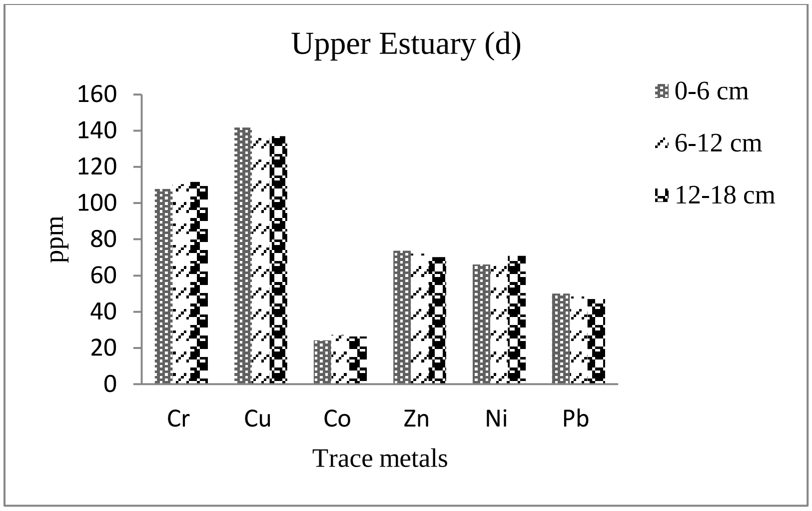


Fig. 5B.1. Distribution of metals (Cr, Cu, Co, Zn, Ni and Pb) within mangroves in different sections of the cores collected at lower estuary (a), lower middle estuary (b), upper middle estuary (c) and upper estuary (d)

Finer sediments are dominant in all the four studied locations (Table 5B.1). Mangroves have ability to dissipate surface wave energy, reduce wave heights, and decelerate flow of water, thus minimizing erosion, therefore, finer sediments are deposited within the mangrove areas. However, presence of slightly higher coarser sediments (15 to 25 %) is noted in lower estuary. Violent hydrodynamic conditions prevailing towards the lower estuary might have facilitated higher coarser sediment deposition as discussed earlier. Upper middle estuary also shows presence of slightly higher coarser sediments. Upper middle estuary is part of the main channel and must be representing river brought terrigenous material. Clay is found to be higher in lower middle estuary. As mentioned in earlier chapter, lower middle estuary sampling location is in the inlet of main channel which facilitates calm conditions which in turn favours finer sediment deposition. Silt is relatively more in surface sediments at all the locations except at the lower middle region. pH and organic carbon show less variations within the sediment sections and between the different estuarine regions. However, organic matter like silt also shows slightly higher concentration in surface sediments. High organic matter content in surface sediments indicates anthropogenic contribution in addition to mangrove litter in recent years. In upper estuary, higher concentration of organic matter is found towards the bottom sediments. The refractory organic matter received from land drainage must have mineralized (Marchand et al., 2011) faster at this location.

In lower estuary Fe, Mn and Al are higher in bottom sediments. Trace metals viz. Cu, Co and Ni are higher in surface sediments. Zn, Pb and Cr are found to be higher in bottom sediments. In lower middle estuary, Fe, Mn and Al are found to be higher in bottom sediments like that noted in lower estuary. Concentration of Zn and Ni do not show any variation with depth. Cr is found to be higher in bottom sediments whereas, Cu, Co and Pb in surface sediments. In the upper middle estuary, Fe and Al along with Cr, Cu, Co and Ni are found to be higher in middle sediment section whereas, Mn shows higher value in bottom sediments. Zn and Pb are found higher in both surface and middle sections. In upper estuary, Fe and Al are found to be higher in middle sediment sections whereas, Mn is high in surface sediments. Pb is also found to be higher in surface sediments. Cr is found to be higher in bottom sediments. Cu and Zn are found to be higher in surface and middle sediments and Co and Ni are high in middle and bottom sediments. Higher percentage of finer sediments mostly clay in bottom sediments might have facilitated the deposition of most of the metals in bottom sediments at lower estuary. Due to lack of vigorous water motion, the fine sediments tend to settle at the bottom. Mangrove roots tend to hold the finer sediments. Fine material, with a larger surface to volume (or weight) ratio, has a greater potential to scavenge both inorganic and organic pollutants from the water column (Madiseh et al., 2009). The fine grain sediments provide larger reactive surface area that can gather the trace elements. Moreover, very fine sediment grain size like clays which have a negative surface charge and cation exchange capacities that readily attract trace elements and trace element-carrying substances (Tam and Wong, 2000). Higher content of organic matter in surface sediments might have supported the metal deposition in surface sediments of lower estuary. Metals probably have been retained in the sediments bound to the clay and organic matter in bottom and surface sediments respectively. Metals such as Zn, Pb and Cr are found to be higher in bottom sediments at lower estuary. According to the studies on the chemical phase of metals in the estuarine sediments, organic/sulfide fractions and Fe/Mn oxides are important geochemical phases for heavy metals in the sediments (Liu et al., 2003a) in the sediment column. In addition, metals can be adsorbed onto the surface of minerals, like clay minerals (Dong et al., 2000).

In lower middle estuary, higher amount of clay in bottom sediments must have resulted in deposition of Fe, Mn and Al. Presence of higher Cr in bottom sediments in lower middle estuary might be due to its association with aluminosilicates. Increase of Cu, Co and Pb in surface sediment section reflects recent anthropogenic addition. Increase of organic matter content in surface sediments might be derived from the decomposition of root material and leaf litter in addition to anthropogenic source.

In upper middle estuarine region, Fe and Al are found to be higher in middle sediment section whereas, Mn is higher in bottom sediments. Release of Mn from surface sediments to the overlying water might have occurred by diffusion processes which resulted in decrease of Mn concentration in surface sediments. Finer sediments in middle sediment section with Al and Fe indicate their natural source along with trace metals Cr, Cu, Co and Ni. Similarly, in upper estuary, Fe and Al are found to be higher in middle sediment section whereas, Mn is high in surface sediments. The higher and lower contents of Mn and Fe indicate the change in redox environment and detrital input during sedimentation (Anawar et al., 2010). Anthropogenic activities which include open cast mining in the catchment area of Mandovi River could result in higher Mn in surface sediments towards upper estuarine region. Pb is found to be higher in surface sediments. Increase in Mn and Pb in surface sediments may be due to co-precipitation of Pb with Mn during Mn oxide formation (Kumar and Edward, 2009; Finney and Huy, 1989), in addition to anthropogenic origin. Ramos et al. (2006) suggested that mangrove trees can be considered as biochemical reactors due to their active role in organic matter decomposition within the sediments that greatly influence mobility of heavy metals. Cr and Ni are found to be higher in bottom sediments and are associated with increase in organic matter. Organic matter is an important factor in the distribution and concentration of trace metals (Rubio et al., 2000). Cu and Zn are found to be higher in upper and middle sediment sections and Co and Ni in middle and bottom sediments. Higher metal concentration in middle sediment section of both upper middle and upper estuarine regions might be due to their association with aluminosilicates. The lower values in the bottom section may be due to the increase of sulphate reduction while Fe content has decreased because of FeS formation (Goldhaber and Kaplan, 1974).

Total heterotrophic bacterial counts are found to be higher in middle sediment section in lower estuary and lower middle estuary which is associated with organic carbon and finer sediments. In upper middle estuary, heterotrophic counts are found to be higher in surface sediments which agree with higher organic matter concentration in surface sediments. Increase in total heterotrophic bacterial counts is also found in surface sediments of upper estuary and is associated with higher of silt and organic carbon. Sediment properties such as mineralogy, organic matter content and grain size have a major role in influencing bacterial attachment to surface and their proliferation (Johnson and Logan, 1996). Organic matter of marine origin is considered to be more easily degraded than organic matter of terrestrial origin. Microbial communities tend to sequentially use the organic substrates which can be easily decomposable. Thus labile fractions of organic matter are consumed and less utilizable components get accumulated and thus bacterial count decrease in bottom sediment sections. Thus towards upper middle and upper estuarine regions microbes are found to be associated with surface sediments. Total heterotrophic counts are higher at 6 – 12 cm in lower and lower middle regions. Organic carbon is highest in the surface sediments. Organic matter is also considerably high in middle sediment section in lower estuary and lower middle region. This further suggests that not only concentration of organic matter is important to determine the microbial counts but the presence of form i.e. either labile or refractory of organic matter is important. This might have affected the microbial population to proliferate at 6- 12 cm depth.

Within mangroves sand is higher in lower estuary followed by upper middle estuary. Concentration of finer sediments on the other hand is more in all the four locations between 0 – 6 cm sediment section compared to deeper sections. Overall when the four mangrove areas are compared, it is observed that most of the metals are observed to be higher in lower middle estuary and to some extent in upper estuarine region. Metals are found to be higher in deeper sediment sections and this must be due to the formation of metal sulphides and mobilisation of metals to deeper sections and higher metal concentration in surface sediments might be due to recent anthropogenic inputs or their remobilization during early diagenesis from deeper sections.

5.2. Speciation of metals

5.2A. Mudflats

Fe in all the three areas viz lower estuary, lower middle and upper middle estuarine regions, show maximum concentration in F5 fraction at all the three depth sections (Fig. 5A.2a). The concentration of Fe in residual fraction shows 98.95 %, 98.79 % and 97.75 % in sediment sections at 0-6 cm, 6-12 cm and 12 – 18 cm of lower estuary. In lower middle estuary, 92.75 %, 92.99 % and 87.66 % of Fe is present in residual fraction in 0-6 cm, 6-12 cm and 12- 18 cm respectively. At upper middle estuary, 79.47 %, 81.61 % and 78.42 % of Fe is present in 0-6 cm, 6-12 cm and 12- 18 cm respectively. Residual phase of Fe decreases from mouth to upstream.

In lower estuary, the concentration of Mn associated with bioavailable fractions is comparatively higher than in residual fraction (Fig. 5A.2b). The concentration of Mn in exchangeable fraction shows 8.16%, 4.37 % and 3.08 % in 0-6 cm, 6-12 cm and 12-18 cm sections respectively. In carbonate fraction, the concentration of Mn shows 46.24 % in 0-6 cm, 31.35 % in 6-12 cm and 31.68 % in 12-18 cm. In Fe-Mn oxide phase, the concentration of Mn is found to be higher in 6-12 cm and 12- 18 cm with 20.69 % and 21.01 % respectively but with 4.05 % noted in 0- 6 cm. In organic bound phase the concentration of Mn in 0-6 cm, 6-12 cm and 12- 18 cm show 3.79 %, 2.72 % and 2.49 %. In residual fraction the Mn concentration at different depths shows 37.74 %, 40.87 % and 41.79 %. Among the bioavailable fractions Mn is largely associated with carbonate fraction in all the three sediment sections. The concentration of Mn associated with different sections is in the order of carbonate > residual > exchangeable > Fe-Mn oxide and organic in 0-6 cm in lower estuary. In 6 – 12 cm and 12- 18 cm it is residual > carbonate > Fe-Mn oxide > exchangeable and organic phase in lower estuary. In lower middle estuary, the concentration of Mn in exchangeable phase show 20.21 %, 25.44 % and 26.26 % in 0-6 cm, 6- 12 cm and 12 – 18 cm respectively. In carbonate fraction 28.05 %, 22.95 % and 27.11 % whereas, in Fe-Mn oxide 2.23 %, 3.13 % and 3.13 % of Mn is present in different sediment sections. In organic phase, 40.83 %, 30.27 % and 25.26 % of Mn is present. In residual fraction the concentration of Mn is 8.66 %, 18.21 % and 18.24 % at different sediment sections from surface to deeper sections. In general Mn is associated with, organic, carbonate and exchangeable fractions with least found in Fe- Mn oxide phase among bioavailable phase in lower middle estuary. In lower middle estuary, the concentration of Mn is found to be higher in exchangeable in 0-6 and 6-12 cm sections whereas, it is higher in carbonate phase in 12 – 18 cm section. In upper middle estuary, the concentration of Mn in exchangeable fraction show 14.76 %, 21.36 % and 23.45 % in 0-6 cm, 6-12 cm and 12- 20 cm depth respectively. 6.72 %, 10.06 % and 17.64 % of Mn is available in carbonate fraction whereas, 3.70 %, 0.49 % and 0.63 % in Fe- Mn oxide fraction. In organic fraction, 8.35 %, 4.48 % and 5.46 % of Mn is present in respective depths and in residual fraction, the concentration of Mn is observed to be 66.44 %, 63.61 % and 52.82 % at 0-6 cm, 6-

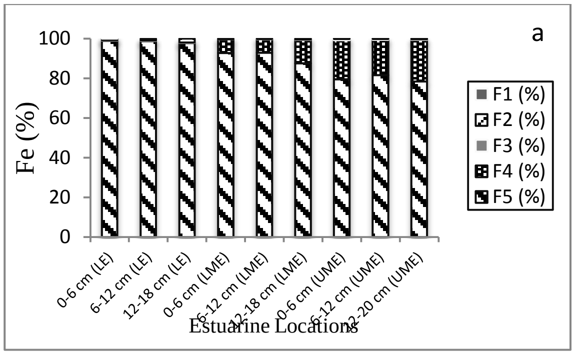
12 cm and 12- 20 cm depth sections. In the upper middle estuary, the concentration of Mn is higher in associated with residual fraction when compared to bioavailable phases. Among the bioavailable phases, F1 fraction shows higher values compared to other phases.

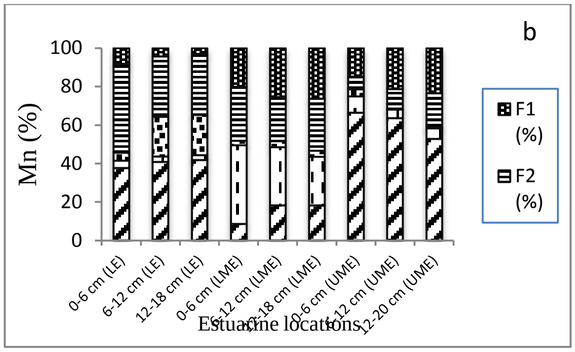
The concentration of Co is found to be 16.43 %, 14.31 % and 18.05 % in exchangeable fraction, 16.05 %, 14.83 % and 15.57 % in carbonate fraction, 40.07 %, 36.48 % and 38 % in Fe-Mn oxide fraction, 6.72 %, 6.67 % and 7.67 % in organic fraction and 20.17 %, 27.71 % and 20.71 % in residual fraction in 0-6 cm, 6-12 cm and 12- 18 cm depths in lower estuary. In lower estuary, Co concentration is higher in bioavailable fractions, of which Fe- Mn oxide is the dominant phase holding the metal in all the three sediment sections (Fig. 5A.2c). In lower middle estuary, Co is 12.55 %, 13.37 % and 16.31 %; 14.37 %, 11.97 % and 10.53 %; 20.68 %, 23.57 % and 26.11 %; 10.57 %, 11.15 % and 13.82 % in exchangeable, carbonate, Fe-Mn oxide and organic fractions in 0-6 cm, 6-12 cm and 12- 18 cm depth sections. In residual fraction the concentration of Co is 41.83 %, 39.95 % and 33.24 % at 0-6 cm, 6-12 cm and 12- 18 cm sections respectively. In lower middle estuarine region, concentration of bioavailable fractions is less than that in lower estuary. In upper middle estuary, 9.82 %, 12.34 % and 8.04 % of Co is found in exchangeable fraction whereas, 9.47 %, 10.22 % and 10.60 % is found in carbonate phase at 0-6 cm, 6-12 cm and 12- 20 cm respectively. In Fe-Mn oxide fraction, 19.65 %, 21.60 % and 17.80 % of Co is present whereas, 10.53 %, 12.38 % and 10.98 % in organic fraction at 0-6 cm, 6-12 cm and 12-20 cm respectively. In residual fraction, 50.50 %; 43.47 % and 52.58 % of Co is present. In upper middle region, bioavailability is comparatively less than observed in lower middle region. Among bioavailable phases Co is more bound mainly to Fe – Mn oxide at this location. Among the three estuarine areas, lower estuary shows higher bioavailability of 79.28 %, 72.29 % and 79.29 % at 0-6 cm, 6- 12 cm and 12 – 18 cm depth sections respectively followed by lower middle and upper middle estuarine region.

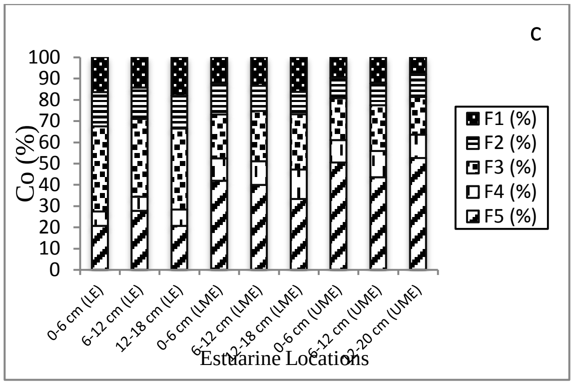
In lower estuary, the concentration of Zn in exchangeable fraction shows 3.65 %, 2.37 % and 3.13 % at 0-6 cm, 6- 12 cm and 12- 18 cm depth sections respectively. The concentration of Zn in carbonate fraction shows 11.56 %, 9.64 % and 7.39 % whereas, the concentration of Zn show 9.63 %, 10.90 % and 11.62 % in Fe-Mn oxide bound fraction. Zn in organic fraction shows 7.11 %, 7.18 % and 7.98 % at 0-6 cm, 6- 12 cm and 12 – 18 cm depth sections. The concentration of

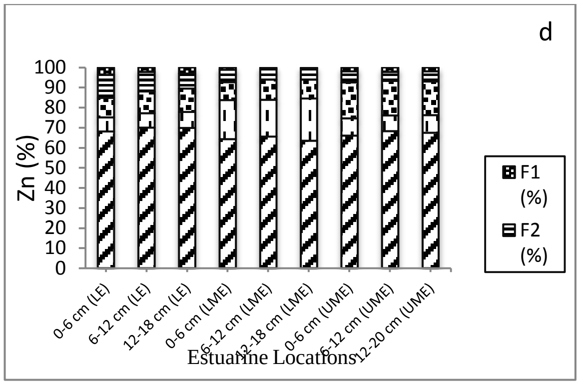
Zn in residual fraction shows 68.03 %, 69.91 % and 69.88 % at 0-6 cm, 6- 12 cm and 12- 18 cm. In lower middle estuary, the concentration of Zn in exchangeable fraction shows 1.16 %, 0.89 % and 0.55 % whereas, 6.19 %, 5.24 % and 5.60 % is present in carbonate fraction at 0-6 cm, 6-12 cm and 12- 18 cm depth sections. The concentration of Zn in Fe-Mn oxide fraction shows 8.89 %, 10.00 % and 9.37 % whereas, in organic fraction it is 19.45 %, 18.30 % and 21.10 %. The concentration of Zn in residual fraction shows 64.30 %; 65.58 % and 63.39 % at 0-6 cm, 6- 12 cm and 12 – 18 cm depth sections. In upper middle region, the concentration of Zn in exchangeable fraction shows 1.98 %, 2.36 % and 1.98 % whereas, in carbonate fraction, it is 5.61 %, 4.35 % and 5.03 % at 0-6 cm, 6-12 cm and 12- 20 cm depths respectively. In Fe-Mn oxide fraction, the concentration of Zn shows 17.81 %, 17.30 % and 16.83 % whereas, it shows in organic fraction 8.62 %, 7.81 % and 8.74 %. The concentration of Zn in residual fraction shows 65.96 %, 68.18 % and 67.43 % at 0-6 cm, 6-12 cm and 12- 20 cm depth sections. In all the three estuarine areas, the concentration of Zn in residual fraction is higher irrespective of depth sections (Fig. 5A.2d).

The concentration of Pb in lower estuary in different fractions are, 21.49 %, 20.39 % and 27.61 % in exchangeable; 7.78 %, 13.11 % and 14.35 % in carbonate; 28.62 %, 29.88 % and 17.45 % in Fe- Mn oxide; 8.68 %, 6.64 % and 8.70 % in organic fraction and 33.41 %, 29.97 % and 31.90 % in residual phase in different sediment sections i.e. 0-6 cm, 6- 12 cm and 12 – 18 cm respectively. In the lower middle region, 28.35 %, 27.27 % and 28.83 % of Pb is found in exchangeable; 13.80 %, 12.47 % and 14.20 % of Pb in carbonate; 14.83 %, 20.78 % and 11.55 % in Fe-Mn oxide; 13.14 %, 12.12 % and 12.24 % in organic fraction and 29.85 %, 27.36 % and 33.18 % in residual fraction in different sediment sections i.e. 0-6 cm, 6- 12 cm and 12 – 18 cm respectively. In the upper middle region, 15.23 %, 16.96 % and 23.85 % of Pb is available in exchangeable fraction whereas, 13.79 %, 13.54 % and 14.83 % in carbonate fraction; 7.84 %, 21.35 % and 19.21 % in Fe-Mn oxide of Pb is found at 0-6 cm, 6- 12 cm and 12-20 cm depth sections respectively. In organic fraction, 9.02 %, 10.12 % and 15.31 % and in residual phase 54.10 %, 38.02 % and 26.79 % of Pb is noted in 0-6 cm, 6- 12 cm and 12- 20 cm depth respectively. The concentration of Pb in bioavailable fractions is higher than found in residual phases in all the three estuarine regions at different depths except for 0-6 cm section in upper middle estuary (Fig. 5A.2e), where the concentration of Pb is higher in residual fraction. The dominant bioavailable phase in lower estuary is Fe- Mn oxide in 0-6 cm and 6- 12 cm whereas in 12 – 18 cm it is higher in exchangeable fraction in lower estuary. In lower middle region, Pb is higher in F1 fraction followed by F3 in 0-6 cm and 6-12 cm sections. In case of 12-20 cm F2 fraction follows F1 fraction. In upper middle region, bioavailability of Pb shows decrease from bottom to surface sections, with considerable Pb concentration in residual fraction at 0- 6 cm. Among the bioavailable fractions, Fe – Mn oxide is predominant at 6 – 12 cm and exchangeable fraction in 12 – 20 cm and also 0-6 cm.









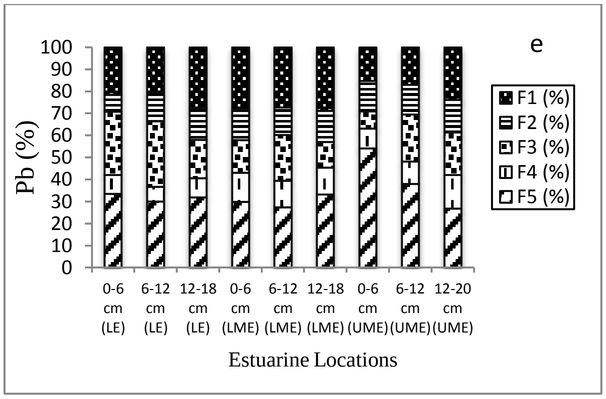


Fig. 5A.2. Geochemical fractionation of Fe, Mn, Co, Zn and Pb with depth in lower estuary (LE), Lower middle estuary (LME), Upper middle estuary (UME) of mudflat sediments in Mandovi estuary

From the results detailed above, it is clear that the residual fraction is the main carrier for Fe and Zn, which generally comprises of detrital silicates, resistant sulphides and refractory organics (Tessier et al., 1979). Metals in this phase mostly remain stable and do not react during sedimentation and digenesis, and therefore have less potential bioavailability. This phase could be considered as inert phase corresponding to the part of metal that cannot be mobilized and as the geochemical background value for the elements in the sediments (Tessier et al., 1979). The greater the percentages of the heavy metal present in this category, the smaller the bioavailability and toxicity. Exchangeable fraction of Fe accounts for <1 % of total metal content in the estuarine sediments, and this level is insignificant to cause any adverse effect on biota. Metals bound within silicate mineral lattices probably account for the bulk of the metals in this phase and concentrations are largely governed by catchment geology (Carrel, 1995).

Within the bioavailable phases, Mn is high in carbonate and Fe-Mn oxide in lower estuary; organic, carbonate and exchangeable in lower middle region and in exchangeable in upper middle estuarine region. The higher value of Mn in carbonate fraction is well understood as their ionic radius is similar to that of calcium which allows Mn to substitute for Ca in carbonate phase (Pedersen and Price, 1982; Zhang et al., 1988). Pizzaro et al. (2003) noted higher values of Mn in Fe-Mn oxyhydroxides and is related to Mn oxyhydroxide removal from water under the strong oxidizing conditions with neutral pH. The higher Mn in organic fraction in lower middle estuarine region is a result of adsorption of Mn onto organic matter and also continuous accumulation of organic matter. Generally, organic matter has higher specific surface area than Fe-Mn oxides or carbonates (Surija and Branica, 1995). However, lower concentration of Mn bound to organics is reported by (Bendell-Young and Harvey, 1992) which is explained as due to weak affinity of Mn for organics. The concentration of Mn in bioavailable fractions is found to be higher in lower middle estuary i.e. 91.34 %, 81.79 % and 81.76 % at 0-6 cm, 6-12 cm and 12-18 cm respectively. The bioavailability of Mn is found to be 62.26 %, 59.13 % and 81.76 % in lower estuary whereas, 33.56 %, 36.39 % and 47.18 % in upper middle estuary at 0-6 cm, 6-12 cm and 12- 18 cm respectively. Mn bioavailability is relatively higher in lower middle estuary followed by lower estuary and upper middle estuarine region.

Compared to Fe, a considerable portion of Mn is associated with bioavailable fractions which imply notable input of Mn from anthropogenic sources. According to Wolters et al. (1989), fertilizer runoff is one of the predominant manmade sources of Mn enrichment. Slightly lower levels of Mn bound to Fe - Mn association with organic bound fraction in lower estuary and upper middle estuarine regions could be attributed to the competition between Fe-Mn organic complexes and hydrous Fe-Mn oxide forms (Sundaray, 2007).

The low concentration of Zn in bioavailable fractions is probably due the fact that metals in this form can easily be absorbed and utilized by organisms in the aquatic environment (Campbell, 1995). Among the bioavailable fractions the Fe–Mn oxide phase is prominent than the other fractions especially in upper middle estuarine region. The association of Zn with the Fe and Mn oxides of soils and sediments has been widely recognized by Kuo et al. (1983) and Gonzalez et al. (1994) which indicate that the Zn adsorption onto these oxides has high stability constants. Zn shows higher concentration in bioavailable fractions in lower middle estuarine region followed by upper middle and lower estuary. Even though Zn is more in residual fraction, its concentration is also relatively higher in exchangeable, carbonate, Fe – Mn oxide and organic fractions put together. This might be probably due to the incorporation of Zn into these phases that precipitates when the river water, with relatively large amounts of dissolved Zn mixes with seawater (Nasrabadi et al., 2010). Rubio et al. (2000) suggested that Zn is fixed mainly to the oxy-hydroxides during deposition. The high percentage of Zn in Fe-Mn oxides/hydroxides supports the known ability of this phase to scavenge Zn from aqueous phase (Isuare et al., 2005). Zn association with organic matter and/or sulphides are low except in lower middle estuary which can be explained, not only by the relatively weak bond of Zn with the organic matter, but by the tendency of this metal to form sulphides, depending on the redox potential of the soil. Post-depositional processes may have influenced the observed distribution of Zn, since it has been suggested that Zn which is mobilized during the dissolution of Fe-Mn oxides, may be repartitioned to carbonates (Zwolsman et al., 1993).

The concentration of Pb associated with different fractions is in the order of Residual > Fe – Mn oxide > exchangeable > carbonate > organic in lower estuary and to some extent in upper middle estuary. Fernandes (1997) and Zhou et al. (1998) also reported similar distribution of Pb species elsewhere. Among bioavailable fractions, Pb is mainly associated with Fe - Mn oxide. Pb can from stable complexes with Fe – Mn oxide (Ramos et al., 1994; Lopez – Sanchez et al., 1996 and Jones and Turki, 1997). The different chemical forms of Pb can affect the solubility and mobility of the metal in the sediments. Pb is also available in considerable quantity in exchangeable, carbonate and organic forms in Mandovi estuary. When all the three estuarine areas are compared except for 0- 6 cm and 6 – 12 cm in lower estuarine region, which shows Fe – Mn fraction as dominant phase, all the sediment sections show higher concentration in exchangeable phase. Pb available in exchangeable form can easily mobilize and thus could be toxic to biota. Gallon et al. (2004) suggested that reductive dissolution of hydrous Fe-Mn oxides could release Pb from sediments to interstitial water in bottom sediment layers. The trace metals in Fe-Mn oxide phase include metals weakly adsorbed on sediments or on their essential components namely clays, Fe and Mn hydrated oxides and humic acids and these metals can be released by ion-exchange processes and further these metals are able to co-precipitate with carbonates present in sediments (Marin et al., 1997; Tokalioglu et al., 2000). Changes in the ionic composition, influencing adsorption– desorption reactions, or lowering of pH could cause remobilisation of metals from Fe-Mn oxides fraction (Marin et al., 1997). Gardolinski et al. (2002) and Gong et al. (2003) have also reported similar observations. The higher concentration of Pb in exchangeable fraction indicates input of Pb from anthropogenic activities. Metals bound to different phases will behave differently in the different sedimentary and digenetic environment. The high mobility of exchangeable phase facilitates it being taken up by benthic organisms living in sediments. Benthic invertebrates are an important link in the transfer of metals to higher trophic levels because of their close association with sediments and their ability to accumulate metals (Morillo et al., 2002). Furthermore, they are often a major component in the diet of many fish (Summers, 1980). The entry of this element into the food chain is a considerable hazard because of its high toxicity. Pb and its compounds from industrial effluents, sewage sludge, domestic waters, pigments, petrol and combustion of fossil fuels are likely to ultimately reach the aquatic environment. Jain et al. (2004) reported approximately 4 % of Pb is associated with exchangeable fraction due to anthropogenic sources, such as municipal and industrial discharges, in sediments of River Narmada. The concentration of Pb associated with organic fraction is relatively less in all the three estuarine areas. This can be attributed to the association of Pb with organic fraction which is influenced by numerous factors, such as total metal load, competition between heavy metals and sediment organic matter concentration (Chakraborty et al., 2012).

Fe- Mn oxides and organic matter plays excellent scavengers role for the removal of heavy metals from water column in aquatic environment. Organic bound fraction is a key scavenger for metals like Mn observed in lower middle estuarine region. Increased levels of exchangeable and carbonate fractions of metals obtained in the study show that these metals are originated from nonlithogenic sources for Pb and Mn respectively in sediment sections. Exchangeable fraction of heavy metals can be easily released back into water column, which may cause secondary pollution and subsequent health risk to exposed population. Numerous earlier reports have also considered exchangeable fraction to be the most mobile and bioavailable phase present in the sediments (Howari and Banat, 2001). Significant variations observed in different fractions could be attributed to numerous factors such as weathering, mineral transport, anthropogenic inputs and physicochemical components of sediments (Sundaray et al., 2011). Anthropogenically derived Co is preferentially associated with Fe - Mn oxides in estuarine sediments (Chiffoleau et al., 1994). The availability of Co in Fe – Mn oxide fraction have been also reported elsewhere by Calmano and Forstner (1983) and Jones and Turki (1997). The concentration of Co in bioavailable phase seems to be less in upper middle estuarine regions. Here, major part of the Co may have come from the parent material of geologic origin and may exist in the residual form in the sediments.

Metals bound to different phases will behave differently in the sedimentary and diagenetic environment, and thus have different potentials for remobilization and for uptake by biota. The reducible phase, consisting of Fe - Mn oxides is a major host for Co and Pb. In the study, it is observed that among different metals higher Co concentration is available in Fe- Mn oxide fraction followed by exchangeable phase especially in lower estuary and lower middle estuary. Pb is found associated with Fe-Mn oxide fraction and thereafter bound to exchangeable phase. During early diagenesis, microbially mediated redox reactions quickly result in the reduction of insoluble Fe (III) and Mn (IV) oxides and release of Fe (II) and Mn (II) species indicating that metals bound to these oxides have potential for metal mobilization from reducibly bound metals to these sediments. Thus the process will result in dissolution of these oxides and possible release of associated metals. Some of the metals originally bound to oxide phases may be fixed as pyrite as sulphidization progresses (Huerta- Diaz and Morse, 1992) effectively immobilizing them as long as conditions remain reducing as observed in case of Zn.

Organic fraction is the dominant Mn host observed in lower middle estuarine region. Continuing microbial oxidation of the organic substrate within the sediment will result in further gradual release to labile phases i.e. carbonate and exchangeable fractions. In addition, agitation of the sediment and mixing of oxygenated water, as a result of dredging of the shipping channel would enhance oxidative degradation of organic matter and could solubilise significant amounts of Mn. Within the few centimetres of sediment water-interface any remaining oxides of Fe (III) must be rapidly converted to monosulphides and eventually pyrite. Half life of common sedimentary oxide minerals in presence of dissolved sulphide is small and thus converted to sulphidal form (Canfield et al., 1992). In this condition Mn escapes and migrates towards sediment water interface. Mineralization of organic material in the oxic upper layers of the sediment can release bound metals and a significant flux of these metals from the sediment to the overlying water (Petersen et al., 1995) which is observed in lower estuary that is why the concentration of Mn is lesser in exchangeable fraction as Mn might have been lost or released during early diagenesis.

To understand relative concentration of bioavailable metals in different parts of the estuary, the data was plotted on isocon diagram. When lower estuary and lower middle estuary regions are compared (Fig. 5A.3a) it is observed that, the concentration of bio – Mn, res – Co, silt and clay are higher in lower middle and bio – Co, res – Mn and sand are higher in lower estuary.

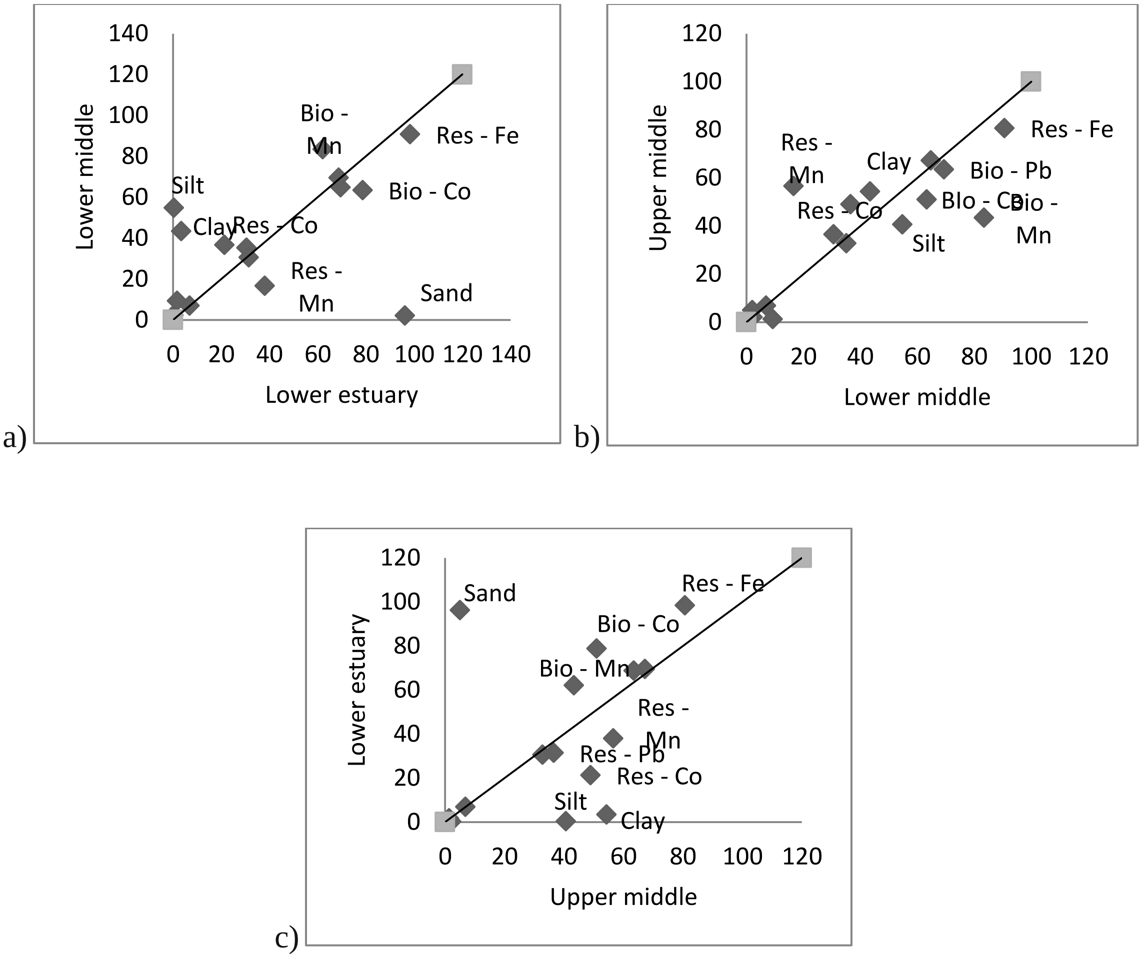


Fig 5A.3. Isocon plot for cores collected from mudflats of lower, lower middle and upper middle estuary

When lower middle and upper middle estuarine regions (Fig. 5A. 3b) are compared it is observed that res – Mn, res – Co and clay are prominent in upper middle whereas, bio – Mn, bio – Co, res – Fe and silt are higher in lower middle estuary. This indicates presence of higher concentration of bioavailable Mn in lower middle estuary. Possible factors for higher and changing bioavailability of metals are higher primary productivity which results in higher organic matter degradation which in turn changes the pH. Other data points fall on or close to isocon line indicating minor variability within the studied parameters.

Further, when of lower estuary is compared with upper middle estuary (Fig. 5A. 3c), residual form of Mn and Co along with clay and silt are prominent in upper middle estuary. Res – Fe, bio – Co, bio – Mn and sand are higher in lower estuary. This suggests the role of Fe – Mn oxides in entrapment of bioavailable metals in lower estuary. Residual form of metals present in upper middle estuary indicates their lithogenic origin.

5.2B. Mangroves

Out of four locations, for the study of speciation within mangroves only lower estuary, lower middle estuary and upper middle estuary is considered.

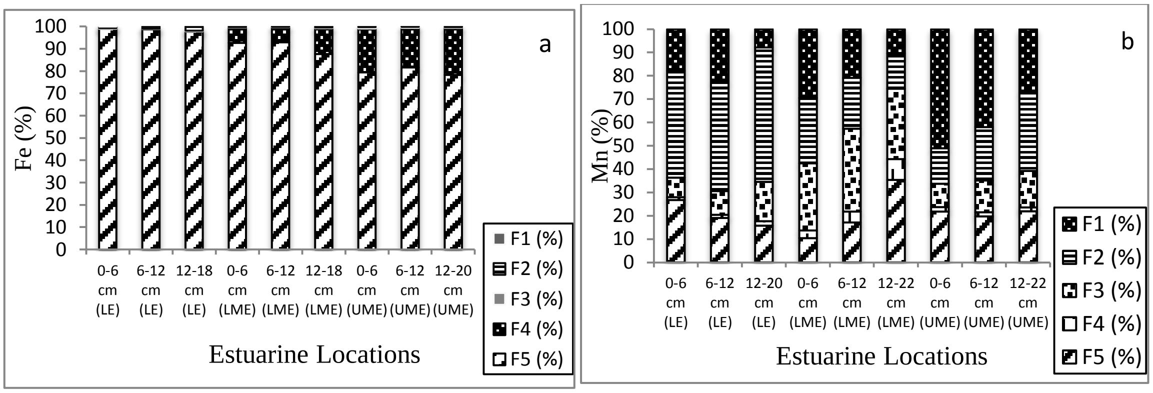
Fe in all the three estuarine regions shows maximum concentration in residual fraction in all the three sediment sections similar to that of mudflat sediments (Fig. 5B.2a).

In lower estuary, the concentration of Mn in exchangeable fraction shows 17.94 %, 22.62 % and 7.92 % whereas, in carbonate fraction the concentration is 45.73 %, 46.86 % and 57.61 % in 0-6 cm, 6- 12 cm and 12- 20 cm sections respectively. The concentration of Mn in Fe- Mn oxide shows 8.47 %, 10.19 % and 16.82 % and in organic fraction 1.20 %, 1.33 % and 1.91 % in 0-6 cm, 6- 12 cm and 12- 20 cm sediment sections respectively. The concentration of Mn in residual fraction shows 26.66 %, 19.02 % and 15.74 % in 0-6 cm, 6- 12 cm and 12- 20 cm respectively. Mn is higher in bioavailable fractions with lesser concentration noted in residual phase. Mn is dominant in carbonate phase followed by exchangeable at 0- 6 cm and 6 -12 cm. In 12-18 cm fraction higher concentration of Mn is found in carbonate fraction followed by Fe- Mn oxide in lower estuary (Fig. 5B.2b). Bioavailability shows increasing trend from surface to bottom sediment sections. In lower middle estuary, the concentration of Mn is found to be higher in Fe- Mn oxide with 28.79 %, 35.53 % and 30.20 % in 0-6 cm, 6- 12 cm and 12- 22 cm sediment sections respectively. Mn concentration in exchangeable fraction shows 29.57 %, 20.91 % and 10.44 % whereas, in carbonate phase it is 27.97 %, 21.75 % and 15.18 % at the different depths. The concentration in residual fraction shows 10.38 %, 17.02 % and 35.27 % in 0-6 cm 6- 12 cm and 12- 22 cm sections respectively. In organic fraction, it is less with 3.29 %, 4.79 % and 8.91 % in different depth sections. Mn is high in bioavailable fractions with dominant in Fe – Mn oxide and Mn shows decreasing bioavailability from surface to bottom. At upper middle region, the concentration of Mn in F1, F2, F3, F4 and F5 is 51.09 %, 42.01 % and 27.20 %; 15.25 %, 22.99 % and 33.67 %; 10.01 %, 13.54 % and 15.56 %; 1.90 %, 1.82 % and 1.69 %; 21.75 %; 19.64 % and 21.89 % at depth sections of 0-6 cm, 6- 12 cm and 12-22 cm respectively. In upper middle estuary, the concentration of Mn is high in exchangeable fraction in 0-6 cm and 6- 12 cm sections whereas, in 12- 22 cm it is high in carbonate fraction. Mn within mangroves is found to be more available in upper middle region followed by lower estuary and lower middle estuarine region.

Among bioavailable phases, Co is bound mainly to Fe-Mn oxide in all the three estuarine regions with 28.87 %, 32.94 % and 37.13 % in 0- 6 cm, 6- 12 cm and 12- 20 cm in lower estuary; 23.01 %, 21.25 % and 28.08 % in 0- 6 cm, 6- 12 cm and 12- 22 cm in lower middle region and 22.88 %, 23.18 % and 21.62 % in 0- 6 cm, 6- 12 cm and 12- 22 cm in upper middle region (Fig. 5B.2c). The concentration of Co in exchangeable, carbonate, organic and residual fractions is 2.34 %, 1.80 % and 1.94 %; 10.23 %, 11.63 % and 14.19 %; 14.72 %, 16.64 % and 13.63 %; 43.83 %, 36.99 % and 33.11 % respectively in lower estuary at different sediment sections from surface to deeper levels. Similarly, in lower middle region, 1.38 %, 0.77 % and 0.95 %; 1.58 %, 0.92 % and 0.33 %; 2.34 %, 1.23 % and 2.96 %; 71.70 %, 75.84 % and 67.67 % in exchangeable, carbonate, organic and residual fractions respectively. In upper middle region, 4.85 %, 3.09 % and 1.01 % in exchangeable; 5.20 %, 7.35 % and 8.58 % in carbonate; 11.06 %, 10.59 % and 9.24 % in organic and 56.00 %, 55.78 % and 59.54 % in residual fractions present in 0- 6 cm, 6- 12 cm and 12- 22 cm sediment section respectively. In all the three estuarine areas Zn is found to be higher in residual fraction. Among bioavailable fractions Fe- Mn oxide is most prominent followed by organic fraction (Fig. 5B.2d). Zn concentration shows 0.24 %, 0.40 % and 0.30 % in exchangeable fraction; 9.21 %, 9.08 % and 7.02 % in carbonate; 15.42 %, 19.23 % and 18.88 % in Fe- Mn oxide; 7.87 %, 8.90 % and 9.66 % in organic and 67.27 %, 62.38 % and 64.12 % in residual fraction in lower estuarine region between 0- 6cm, 6- 12 cm and 12- 20 cm sediment section respectively. In lower middle estuary, 0.18 %, 0.30 % and 0.68 %; 5.28 %, 4.33 % and 4.72 %; 14.82 %, 12.30 % and 11.80 %; 6.00 %, 4.61 % and 4.37 %; 73.72 %, 78.46 % and 78.43 % is found in exchangeable, carbonate, Fe- Mn oxide, organic and residual fractions in 0- 6 cm, 6- 12 cm and 12- 22 cm sections respectively. Similarly in upper middle region, the concentration of Zn bound to exchangeable fraction shows 1.56 %, 2.89 % and 0.97 % whereas, in carbonate it is 8.57 %, 8.77 % and 7.32 % in 0- 6 cm, 6- 12 cm and 12- 22 cm sections respectively. In Fe-Mn oxide fraction, 16.34 %, 17.36 % and 16.12 % is available and 10.51 %, 9.65 % and 7.81 % in organic phase in 0- 6 cm, 6- 12 cm and 12- 22 cm sections respectively.

The concentration is higher in residual fraction contributing 63.02 %, 61.33 % and 67.77 % between 0- 6 cm, 6- 12 cm and 12 – 22 cm respectively.

The distribution of Pb is found to be similar to that of Fe and Zn where in the metal is observed to be higher in F5 fraction (Fig. 5B.2e). The concentration of Pb found in exchangeable fraction is 9.64 %, 9.57 % and 9.07 % in lower estuary between 0- 6 cm, 6- 12 cm and 12 –20 cm sediment sections; 6.36 %, 6.18 % and 6.95 % in lower middle region in 0- 6 cm, 6- 12 cm and 12 – 22 cm and 8.69 %, 10.14 % and 8.77 % in upper middle region 0- 6 cm, 6- 12 cm and 12 – 22 cm respectively. In carbonate fraction, the concentration of Pb is found to be 0.85 %, 1.36 % and 2.43 % in lower estuary, 1.60 %, 1.83 % and 2.19 % in lower middle region whereas, 0.92 %, 1.19 % and 3.25 % in upper middle region in different sediment sections. Pb concentration in Fe- Mn oxide is 9.22 %, 14.92 % and 19.82 %, 12.67 %, 10.43 % and 14.08 % and 10.33 %, 9.46 % and 12.34 % in lower estuary, lower middle and upper middle regions in 0-6 cm, 6-12 cm and 12-22 cm sediment sections. The concentration of Pb in organic and residual fractions shows 8.31 %, 7.95 % and 10.57 % and 71.98 %, 66.20 % and 58.11 % in lower estuary; 8.77 %, 8.53 % and 8.42 % and 70.60 %, 73.02 % and 68.36 % in lower middle estuary and 10.64 %, 12.21 % and 11.93 % and 69.42 %, 67.01 % and 63.71 % in upper middle estuary respectively in 0- 6 cm, 6- 12 cm and 12 – 20 cm in lower estuary and in 0- 6 cm, 6- 12 cm and 12 – 22 cm in lower middle estuary and in 0- 6 cm, 6 – 12 cm and 12 – 22 cm in case of upper middle estuary.



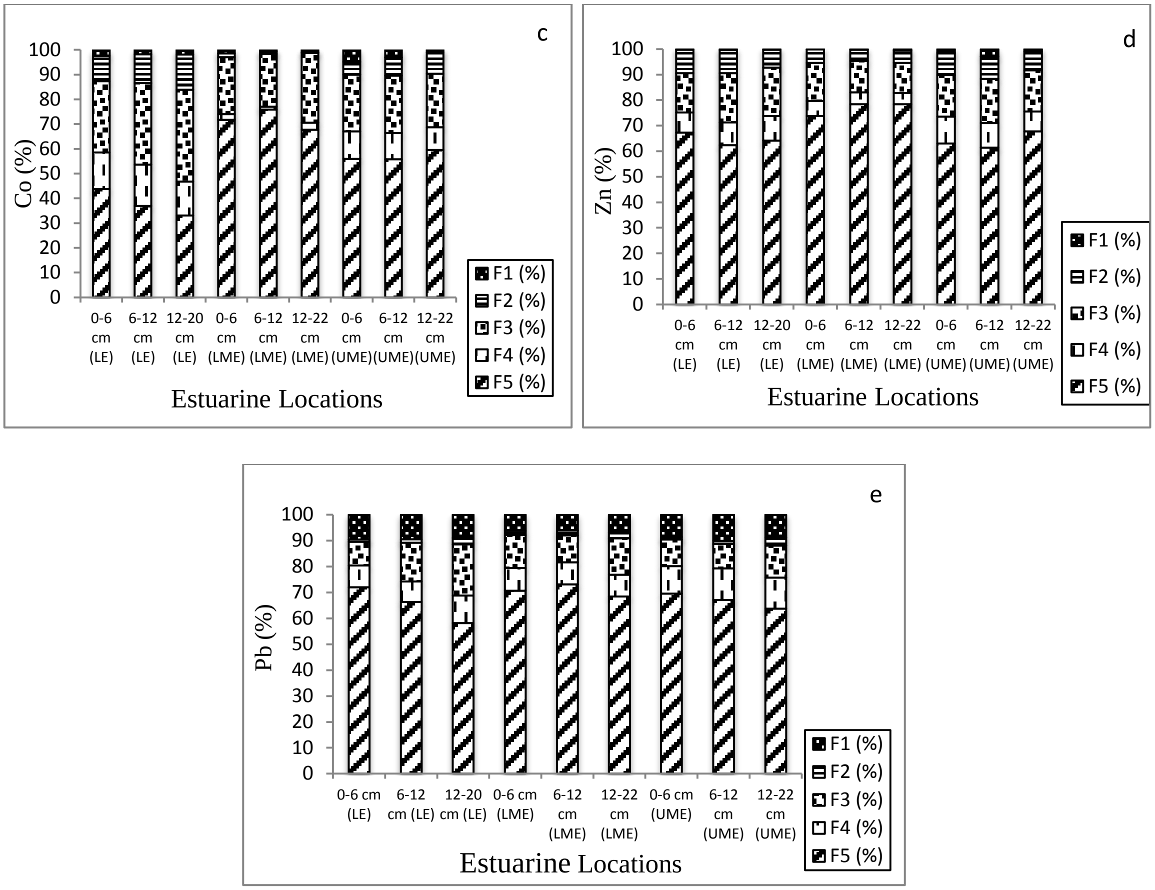


Fig. 5B.2. Geochemical fractionation profile of Fe, Mn, Co, Zn and Pb with depth in lower estuary (LE), Lower middle estuary (LME), Upper middle estuary (UME) of mangrove sediment in Mandovi estuary

The results indicate that Mn is present more in bioavailable form. Further, it is more in surface sediments in lower middle region. During early diagenesis remobilization of Mn from reducing subsurface sediment layers takes place which results in enrichment of Mn in the oxidizing surface layers (Gobeil et al., 1997). Because Mn sulfides are generally unstable in reduced sediments (Forstner and Wittman, 1979), Mn commonly presents a typical behaviour in mangrove sediments, with a higher mobility and bioavailability than most other trace metals, that form more stable metal sulfides (Lacerda, 1998). The Mn migration and accumulation at or near the sediment- water interface may make Mn available for biotic uptake (Lacerda, 1994). Mn is available in exchangeable, carbonate, oxides and also organ complexes. This characterizes the intertidal sediments as a substantial potential of Mn to living organisms inhabiting the sediments which includes microbial populations, burrowing fauna and rooting macrophytes. The oxidizing activity of mangrove rhizospheres has been demonstrated by Nedwell et al. (1994) which also results in oxic conditions within the sediments and facilitate the Mn precipitation towards the surface sediment sections as noted in lower middle and upper middle regions. Clark et al. (1998) show that the concentration and chemical speciation of many trace metals in mangrove sediments are affected by the distribution of geochemically distinct horizons, with oxidation from root activity. Mn is less bound to organic phase in lower estuary as well as middle estuary but higher in carbonate phase. The association of Mn to carbonate fraction has been related to abundance of Mn in earth crust (Tokalioglu et al., 2002). Higher concentration of Mn in lower and upper middle region in exchangeable phase is related to anthropogenic origin as it is mainly extracted in first step of sequential extraction procedure and can be easily leached as a result of acidification processes in sediments (Li et al., 2001). Higher amount of Mn in bioavailable fractions i.e. 0-6 cm in upper middle sediments depicts its availability to biota.

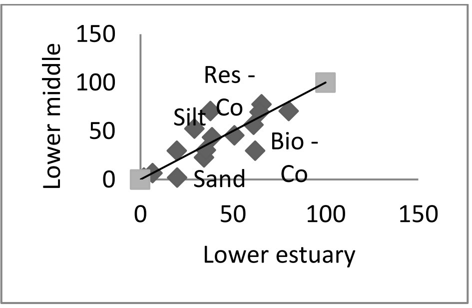
In lower estuary, Co concentration is found to be higher in bioavailable fractions as compared to residual fraction. Co is associated with Fe- Mn oxide phase and bioavailability increased towards the bottom sediment sections i.e. 12 – 20 cm (Fig. 5B.2c). In lower middle and upper middle region, Co shows higher concentration in residual fractions although Fe- Mn oxide is important phase for Co among the bioavailable fractions. Iron and Manganese oxides exist as nodules and cement between particles. These oxides hold trace metals and are mobilized under reducing and acidic conditions. Due to the large surface area, amorphous hydrous Fe- Mn oxides are one of the most important geochemical phases impacting the mobility and behaviour of trace metals (Swallow et al., 1980; Petersen et al., 1993; Turner, 2000). Under oxic conditions Co is removed from the water by co-precipitation and/or adsorption on the freshly formed hydrous ferric oxides. Remobilization of Co from sediment to water occurs under anoxic conditions (Petersen et al., 1995). Among the three areas, Co shows higher bioavailability in lower estuarine sediments followed by upper middle region and lower middle region.

Zn is generally associated with Fe – Mn oxide of sediments and adsorption of Zn on to these oxides has higher stability constants than onto carbonates. Fe and Mn oxides are reported to be main carriers of Zn from the fluvial environment (Fernandes, 1997). Zn is more associated in the residual fraction than bioavailable at all the three estuarine regions. Though higher concentration in F5 fraction it is observed that Zn is also higher in Fe- Mn fraction within non residual fraction.

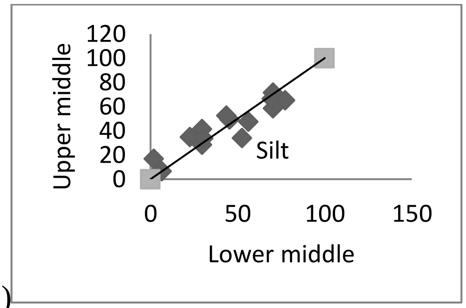
This is in agreement with the studies of Chester (1988) who found that zinc has a preference for the Fe- Mn fraction under oxidising conditions, while under reducing conditions, the most important enrichment phases for Zn in sediments, according to Salomons and Forstner (1984), are the reducible as well as the organic and sulphidic phases. However, from the results obtained it can be stated that Zn might have deposited in oxidising conditions as the concentration of Zn is found more bound to Fe- Mn oxides rather than organic phase.

Like Zn, Pb also associated with hydrous Fe-Mn oxides among bioavailable phases. This suggests that hydrous Fe-Mn oxides play a major role in controlling the fate and transport of Pb in the sediments of Mandovi estuary. It has been reported that if Pb forms stable compounds with Fe hydroxide and Mn oxide it is proved to be sensitive and anthropogenic (Modak et al., 1992). This is in agreement with results reported in Morillo et al. (2004) which shows that Fe and Mn hydrous oxides are important scavengers of Pb in sediments. Bioavailability of Pb decreases from bottom to surface sediments. Anoxic environment prevailing at greater depths creates reducing conditions and thus Pb forms sulphides and thus less concentration is available in Fe-Mn oxide form at middle and surface sediment layers. Reductive dissolution of iron oxyhydroxides and subsequent release of adsorbed Pb is a potential source of Pb in pore waters (Gallon et al., 2004).

Variation in bioavailability between estuarine locations: The data of sand, silt, clay, OC, pH, bioavailable (Bio – Fe, Bio – Mn, Bio – Co, Bio – Zn and Bio Pb) and residual (Res – Fe, Mn, Co, Pb and Zn) fractions are plotted on Isocon diagram.



a)



b)

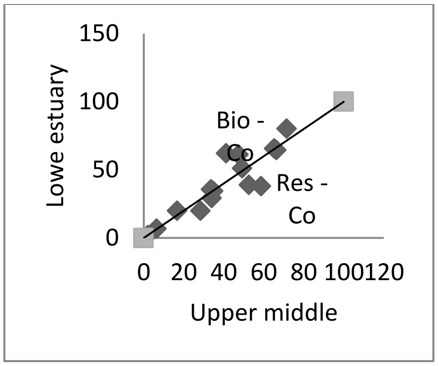
c) 

Fig 5B.3. Isocon plot for cores collected from mangroves of lower, lower middle and upper middle estuary

When the cores between lower estuary and lower middle estuarine region, are compared it is observed that silt along with res- Co is prominent towards the lower middle estuary and bio –Co and sand is higher in lower estuary (Fig. 5B.3a). Other data points lie on or close to isocon line indicating less variation. When the mangrove sediment cores collected within the lower middle and upper middle estuarine regions are compared it is noted that silt is higher in lower middle region (Fig. 5B.3b).

The data points when plotted between upper middle and lower estuary when compared show less variability except bio- Co which is relatively more in lower estuary (Fig. 5B.3c) and res – Co in the upper middle region. This indicates presence of res – Co from natural sources. Presence of bio – Co reflects anthropogenic input (Li et al., 2000) at lower estuary.

5.3. Mudflats v/s Mangroves Environments

When the mudflat and mangroves sub-environments are compared, sand is higher in mudflats at lower estuary whereas clay is comparatively higher at lower estuary and lower middle estuary of the mangroves of the Mandovi estuary. In the upper middle regions sand is around (12-19 %) in mangroves and clay is more in mudflats. The trapping efficiency by root systems in mangroves that retards forces of erosion seems to be a key factor in the burial process unlike observed in mudflats. However, in case of upper middle region river runoff brought material must be playing role in distribution of sediment particles. Organic carbon is higher in most of the sediment sections in mangrove areas except lower middle region. Within mangrove sediments leaves, propagules and twigs and litter from trees are the potential sources of carbon. Leaf litter is an important phenomenon (Rajendran and Karthiksen, 2007) occurring in mangrove areas which plays an important role in adding up the organic carbon faster than its degradation unlike mudflats areas wherein organic matter available undergoes microbial degradation. In addition higher organic carbon in mangrove sediments may be due to the association of finer sediments and sedimentation rate (Raghunath and Sreedhara Murthy, 1996). Organic matter is also transported from mangrove areas. The quantity of litter available for export from mangroves is dependent on the forest type, hydrodynamics, degree of litter retention and bioturbation. Lower middle region being a sub-channel facilitates calmer depositional conditions and therefore more organic matter is associated with mudflats at this location. Metal concentration is higher in mangrove areas than mudflat region. In mudflats the increases of metals are towards the surface sediments (0- 6 cm) whereas, increase is observed towards the bottom sediments (below 12 cm) in mangroves. This indicates the role of high density of roots, short period of flooding in the mangrove forest environment, the redox status due to presence of sulphides of vegetated sediments for metal concentration as metal sulphides (Marchand et al., 2012) in mangroves. In mudflats, coarser sediments facilitate remobilisation of metals towards surface to form Fe- Mn oxides.

Total heterotrophic counts are higher in mangrove sediments in all the three estuarine regions except for surface sediments in lower estuary and lower middle estuary. Generally, organic and inorganic nutrients come from land to the mangroves and are trapped within the mangrove ecosystems by roots. In addition leaf litter production has a significant effect on nutrient cycling in mangrove ecosystems. Litter fall is a good indicator of the mangrove productivity, and its decomposition rate is of great significance as it reflects the nutrient recycling in the estuarine ecosystems. Consequently, the mangrove environments become important sink for nutrients, organic and inorganic contaminants (Harbison, 1986). Mangrove sediments are usually characterized by waterlogged anaerobic soils (Clark et al., 1998; Harbison, 1986) and have high organic matter and sulfide contents (Machado et al., 2002). Horizontal and vertical distribution of bacterial populations in sediments is influenced by various factors important among them are the physico-chemical nature of sediments and the presence of high organic matter concentrations. Higher heterotrophic counts observed in surface sediments in lower middle estuary may be regulated by more organic matter present. Generally, microbial populations are more abundant in muddy sediments than in sandy ones. High bacterial population may indicate favourable conditions for opportunistic species, whereas the complex substrate requirements of microbial communities in later successional states may be reflected by lower viable numbers. Although mangroves are characterized by water logged anaerobic soils there are various means where in mangrove ecosystem is aerated by biological processes such as bioturbation or oxygen release from mangrove roots, resulting in oxidizing conditions (Clark et al., 1998).

The results of bioavailability shows mangroves of lower estuary and upper middle estuary show higher bioavailability of Mn in different sediment sections whereas, higher bioavailability of Mn is noted in lower middle mudflats in 0-6 cm and 12- 18 cm sediment sections. Bioavailability of Mn in 6- 12 cm shows negligible difference between the mudflat and mangrove area of lower middle estuary. Higher concentration of Mn in exchangeable fraction is noted in mangroves, in addition carbonate phase contributed for higher bioavailability of Mn. Redox conditions determine intensity of Mn dissolution as well as diffusion and precipitation of Mn. Early diagenesis is responsible for remobilization of Mn from the reducing deeper sediment layers and accumulation in oxidized surface layers and thus increases bioavailability.

The intensity of Mn diagenetic remobilization from mangrove forest to mudflat sediments varies observed, which confirms that the degree of redox conditions dominate the Mn mobility in these intertidal environments. Bioavailability of Co is very less in sediments of mangrove regions in lower middle and upper middle estuarine regions at all the depths. The percentage of Co in first four fractions is found to be higher in sediments of lower estuary, lower middle and upper middle mudflat regions and also in mangroves of lower estuary. Pb also shows higher concentrations in bioavailable fractions in the sediments of mudflats.

The mudflat environments, subject to a periodic submergence and contain coarser sediments and therefore can provide an aerobic condition and thus regulates processes leading to mobilisation of metals. In mangroves, lack of tidal flushing and good water circulation, anaerobic conditions may prevail which favours the formation of stable and insoluble metal sulphides via the action of sulfate reducing bacteria, but facilitates migration of Mn as it is more stable as oxide.

5.3.a. Mudflats *Vs* Mangroves (Sediment components)

In order to understand the difference between mudflats and mangroves the data of various parameters was plotted on the Isocon diagram (Grant, 1986; Siraswar and Nayak, 2011). The average values of parameters such as sand, silt, clay, organic carbon, pH, total heterotrophic counts (THC) and metals such as Fe, Mn, Al, Cr, Cu, Co, Zn, Ni and Pb (Fig 5.4) are plotted.

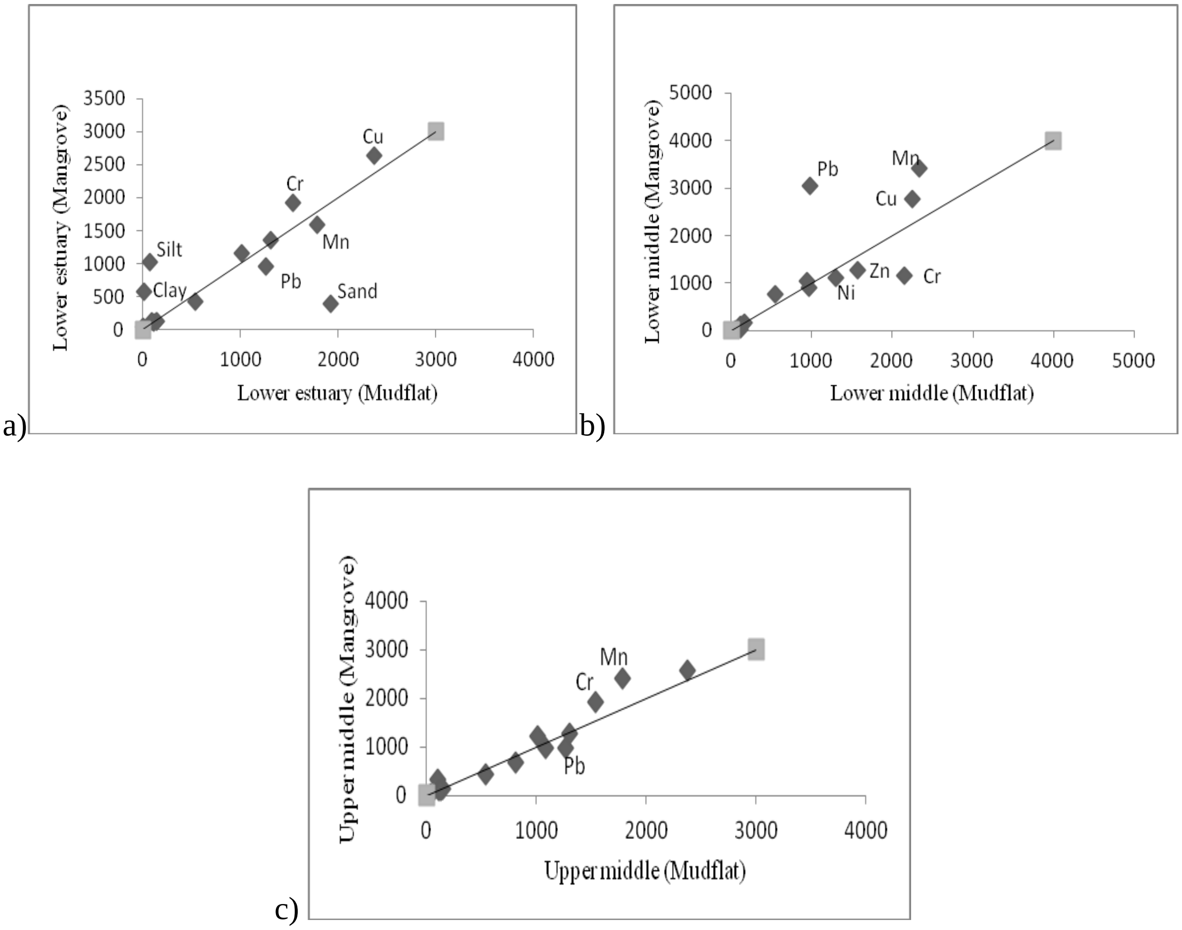


Fig. 5.4. Isocon diagram (Grant, 1986) individual points represent average value of sediment component and element of the core, mudflats vs mangroves

Sand, Mn and Pb are more pronounced in mudflats (Fig. 5.4a) whereas, silt, clay, Cu and Cr are higher in mangrove region at lower estuary. Other data points lie on or close to the isocon line indicating less variation between the two sub-environments. Mn deposition in the sediment is attributed to the formation of Mn coatings on calcite crystals (Kanellopoulos et al., 2006). Adsorption of Pb on Mn oxyhydroxides is also observed earlier (Nelson et al., 1999) in sediments.

In the lower middle region, Mn, Cu and Pb are higher in mangrove region and Zn and Cr are more in the mudflat area at lower middle estuary (Fig. 5.4b). Mangrove sediments being anaerobic and reduced, with high sulphide and organic matter content, have a high capacity to retain some metals either in sulphide form at deeper levels or as oxides in the surface sediments. Mn oxyhydroxides have a capacity to scavenge metals and thus Cu and Pb from anthropogenic origin get associated with Mn. Mn and Cr are found to be prominent in mangrove sediments and Pb in mudflat sediments towards upper middle region (Fig 5.4c).

The low hydrodynamic condition in mangroves favours the accumulation of finer sediments and organic matter. Clay minerals have a higher surface area to volume ration and can absorb material into their lattice framework (Loomb, 2001). In general, estuarine mudflat being at lower elevation is generally subjected to strong hydrodynamic conditions of tidal action compared to mangroves which are being situated at a higher elevation. Areas that are sheltered from strong tidal currents are subject to lesser amounts of erosion and are liable to be storage areas or sinks for contaminated sediments (Rees et al., 1996).

Lacking tidal flushing and good water circulation, anaerobic conditions may prevail which favours the formation of stable and insoluble metal sulphides via the action of sulfate reducing bacteria as stated earlier. In contrast, the mudflat environment, subject to a periodic immersion and emersion, is more aerobic environment, especially at the sediment-water interface (Che, 1999). Metal concentrations in the mudflat sediments and that of mangrove differ because of variations in sediment characteristics in the two sub environments. Sediments that accumulate in mangroves are potential repositories of anthropogenic pollution because of high total organic carbon content, anaerobic characteristics and rapid turnover and burial (Marchand et al., 2006).

5.3.b. Mudflats v/s Mangroves (Metal species)

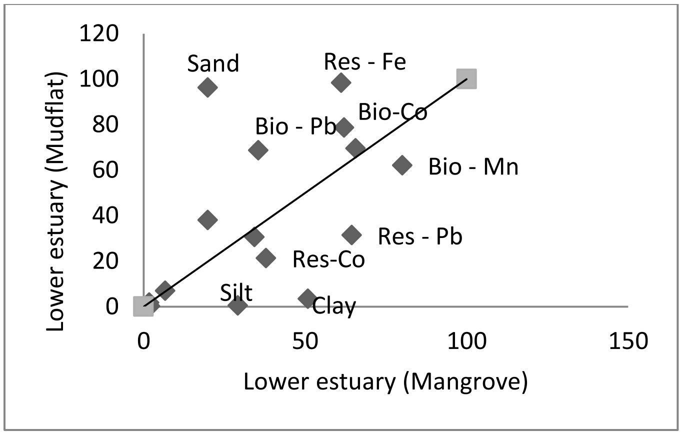


Fig 5.5a. Isocon plot for cores collected from mudflats and mangroves of lower estuary

When the cores from mudflats and mangroves from lower estuary are compared (Fig. 5.5a) bio – Pb, bio- Co and res – Fe and sand are prominent in mudflat indicating anthropogenic source for Pb and Co. Higher concentration of metals in bioavailable fractions indicate the anthropogenic inputs from industrial development and urbanisation in the surrounding areas. Bio- Mn, res- Co and res - Pb with clay and silt are higher in mangroves. The very fine sediment grain sizes provided larger reactive surface area that can gather the trace elements and hydrous oxides, which in turn may also form the trace element complexes. Moreover, very fine sediment grain sizes which have a negative surface charge and cation exchange capacities that readily attract trace elements and trace element-carrying substrates (Tam and Wong, 2000).

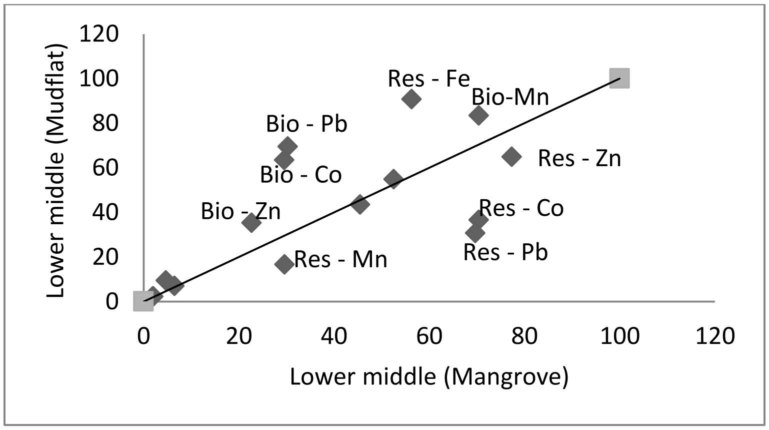


Fig 5.5b. Isocon plot for cores collected from mudflats and mangroves of lower middle estuary

In lower middle estuarine region, prominent variables which are higher in mudflats are bio – Pb, bio –Mn, bio – Zn, bio – Co along with res- Fe (Fig 5.5b) are higher in mudflats. In mangroves, residual forms are higher. The mudflat which is subject to a periodic submergence with tidal water provides aerobic conditions, may facilitate remobilisation of metals, in a calm sub-channel environment.

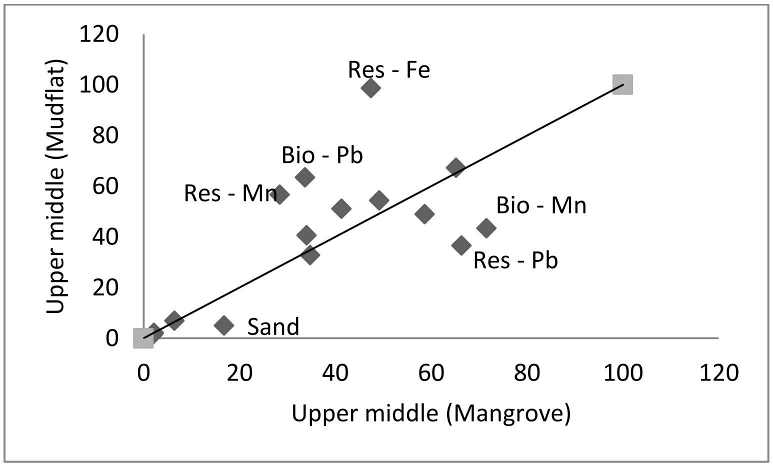


Fig 5.5c. Isocon plot for cores collected from mudflats and mangroves of upper middle estuary

In the upper middle estuarine region, most of the points fall on or close to isocon line indicating less variability (Fig 5.5c). Res – Mn, res – Fe along with bio –Pb is prominent in the upper middle mudflats. Bio-Pb must be of anthropogenic source. In mangroves bio- Mn and res –Pb along with sand is prominent. Roots present within mangroves might entraped metals carried and transported by tidal currents and may eventually incorporate into sediments.

5.4. Metal Enrichment

The Geoaccumulation index is a quantitative measure of the degree of pollution in aquatic sediments. It consists of seven grades ranging from unpolluted to very extremely polluted. Geoaccumulation index has been computed for both mudflat and mangrove sediments of Mandovi estuary to understand grade of pollution with depth. Further, pollution severity and its variation for the sites are determined with the help of pollution load index. This index is a quick tool in order to compare the pollution status of different places. The pollution load index (PLI) for core or region is calculated from the contamination factors (CFMe = concentration of metal in sediment/ base value for that metal) of each of its constituent samples (Tomlinson et al., 1980), where the metal base value represents its average concentration in shale (Li and Schoonmaker, 2005). Further, sediment quality values are used to screen the potential of contaminants within sediment and compared sediment contaminant concentration with the corresponding quality guidelines (Spencer and Macleod, 2002).

5.4.a. *Geoaccumulation Index*

Igeo values were computed following Muller (1979). Igeo is provided with pollution levels, ‘0’ as unpolluted and ‘6’ as strongly polluted (Table 2.3) as described in chapter 2.

In the core sediment collected at the lower estuary, most of the metals namely Fe, Mn, Ni, Zn, Cr, Co and Pb fall in unpolluted class (Table 5A.4) indicating sediments at this location are not contaminated by these metals. However, Igeo values of Cu falls in unpolluted to moderately polluted class at all the sediment sections. The sediments of lower middle estuary exhibits zero Igeo value indicating unpolluted class in case of metals Fe, Ni, Cr, Co and Zn. Mn falls under moderately polluted class at all the sediment sections whereas, Cu and Pb fall under unpolluted to moderately polluted class in all the sediment sections. In the sediments of upper middle estuary, metals Ni, Zn, Co and Cr exhibits negative values. However, Mn falls in moderately polluted class in 0- 6 cm whereas, unpolluted- moderately polluted class in 6- 12 cm and 12 – 20 cm. Cu falls under unpolluted to moderately polluted class in all the sediment sections. Pb falls in moderately polluted class in 0-6 cm and 6-12 cm sections whereas, unpolluted to moderately polluted class in 12- 20 cm section.

Table 5A.4. Geoaccumulation Index (Igeo) values at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of Mandovi estuary

Mudflat									
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary		
Metals	0-6	6-12	12-18	0-6	6-12	12-18	0-6	6-12	12-20
Fe	-2.11	-2.22	-2.22	-0.26	-0.35	-0.34	-0.28	-0.30	-0.33
Mn	-1.44	-1.71	-2.01	1.61	1.23	1.33	1.32	0.89	0.87
Cr	-0.77	-0.62	-0.51	-0.16	-0.23	-0.25	-0.73	-0.64	-0.71
Cu	0.58	0.62	0.72	0.88	0.68	0.66	0.95	0.92	0.63
Co	-1.67	-1.66	-2.11	-0.05	-0.08	-0.05	-0.01	-0.00	-0.71
Zn	-2.02	-1.96	-2.08	-0.93	-0.77	-0.90	-1.16	-1.09	-1.11
Ni	-1.75	-1.71	-1.78	-0.69	-0.65	-0.64	-1.04	-0.92	-1.03
Pb	0.08	0.05	-0.32	0.68	0.74	0.70	1.30	1.16	0.85

In mangroves, Fe, Ni, Zn, Cr, Co fall in zero class indicating uncontaminated sediments (Table 5B.4) at the lower estuary, however, Mn falls in unpolluted class in 0-6 cm section whereas, in 6-

12 cm and 12 – 20 cm it falls in moderately polluted class. Cu falls in moderately polluted class in 0- 6 cm whereas, it is in unpolluted to moderately polluted class in 6- 12 cm and 12- 20 cm sediment sections. Pb falls in unpolluted to moderately polluted class at all the sediment sections. Sediments of lower middle exhibits zero class in case of metals Fe, Cr, Zn and Ni. Sediments fall under moderately polluted class in case of Mn at all the sediment sections. Cu also falls in similar class as that of Mn in 0-6 cm whereas, in 6- 12 cm and 12 – 22 cm it is found to be in class 1 i.e. unpolluted to moderately polluted class. Pb falls under moderately to strongly polluted in 0- 6 cm and 6- 12 cm whereas, in 12- 22 it falls in moderately polluted class. In the upper middle estuary, Mn falls under moderately polluted class in all the sediment sections. Cu falls under moderately polluted class in 0- 6 cm and 6- 12 cm whereas, unpolluted to moderately polluted in 12- 22 cm. Pb falls in unpolluted to moderately polluted class at all sediment sections. In upper estuary, Mn and Cu fall in moderately polluted class at all depths. However, Pb values fall in unpolluted to moderately polluted class.

Table 5B.4. Geoaccumulation Index (Igeo) values at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary

Mangrove												
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary			Upper Estuary		
Metals	0-6	6-12	12-20	0-6	6-12	12-22	0-6	6-12	12-22	0-6	6-12	12-22
Fe	-0.42	-0.50	-0.29	-0.45	-0.24	-0.22	-0.15	-0.14	-0.22	-0.22	0.12	0.11
Mn	-0.55	1.03	1.07	1.72	1.68	1.99	1.05	1.38	1.57	1.70	1.44	1.49
Cr	-0.45	-0.25	-0.31	-2.17	-0.47	-0.56	-0.31	-0.22	-0.53	-0.21	-0.18	-0.16
Cu	1.01	0.95	0.90	1.11	0.98	0.98	1.01	1.05	0.78	1.07	1.06	1.02
Co	-0.27	-0.88	-0.62	0.52	0.56	0.31	-0.27	-0.20	-0.53	-0.23	-0.07	-0.12
Zn	-1.10	-1.03	-1.02	-1.17	-1.03	-1.14	-1.03	-1.02	-1.38	-0.95	-0.98	-1.03
Ni	-0.70	-0.90	-0.92	-0.86	-0.88	-0.90	-0.66	-0.57	-0.88	-0.62	-0.53	-0.53
Pb	0.64	0.58	0.73	2.81	2.67	1.69	0.80	0.83	0.54	0.74	0.69	0.65

5.4.b. *Pollution Load Index (PLI)*

In the lower estuary the value of PLI is less than 1 indicating mudflat sediments here are within unpolluted class or no deterioration is observed with respect to metal contamination (Table 5A.5). In lower middle and upper middle estuarine regions the PLI exhibits values >1, indicating sediments have been deteriorated with metal contamination at these locations in all sediment sections.

When the bulk metal data is compared with SQUIRT (Table 5.7), the value of Mn, Cr and Co is above AET in middle estuarine regions (Table 5A.6). Co is less than AET and Cr is above AET in lower estuary. Mn also shows values above AET in 0-6 cm and 6-12 cm and below AET in 12-18 cm in lower estuary. Cu values are below ERM in all the estuarine areas and all the depths. Zn shows values below TEL in all the estuarine locations at all the depths.

Table 5A.5. Pollution Load Index (PLI) values at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of Mandovi estuary

	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary		
Mudflat	0-6	6-12	12-18	0-6	6-12	12-18	0-6	6-12	12-20
PLI	0.68	0.67	0.61	1.27	1.22	1.21	1.54	1.50	1.37

Table 5A.6. Bulk metal concentration at different sediment depths (cm) of lower, lower middle and upper middle estuarine regions in mudflat sediments of Mandovi estuary

Bulk metal concentration (Mudflat regions)									
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary		
Metals	0-6	6-12	12-18	0-6	6-12	12-18	0-6	6-12	12-20
Mn	335.33	276.50	223.31	2752.6	2152.1	2270.1	2266.1	1739.8	1652.8
Cu	100.58	103.56	111.00	124.16	108.44	106.94	130.66	128.25	104.15
Zn	35.33	36.63	33.75	75.00	84.63	76.50	63.91	66.93	66.15
Cr	73.41	81.44	88.69	111.41	105.88	104.94	75.16	80.12	76.15
Co	9.00	9.06	8.16	27.58	26.94	27.63	28.33	28.43	25.55

Sediments of mangrove regions of lower, lower middle, upper middle and upper estuary exhibit PLI values greater than 1 at all sediment sections and thus indicating deterioration of sediments (Table 5B.5). Bulk metal concentration of mangrove regions are compared with SQUIRT (Table 5.7), it is observed that Mn and Co values are above AET, Cu above PEL at different sections and at all the estuarine regions of Mandovi estuary (Table 5B.6). Cr also shows values above AET in lower estuary, upper middle and upper estuary at all the sediment sections. Cr shows value below AET in 0-6 cm and 6- 12 cm whereas, above AET in 12-22 cm depth. Zn shows values below TEL in all the estuarine locations at all the depths. Thus sediments of mudflat and mangrove sub environments of Mandovi estuary are contaminated and are under the influence of anthropogenic sources.

Table 5B.5. Pollution Load Index (PLI) values at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary

	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary			Upper Estuary		
Mangrove	0-6	6-12	12-20	0-6	6-12	12-22	0-6	6-12	12-22	0-6	6-12	12-22
PLI	1.40	1.37	1.44	1.70	1.79	1.84	1.55	1.65	1.44	1.70	1.71	1.70

Table 5B.6. Bulk metal concentration at different sediment depths (cm) of lower, lower middle, upper middle and upper estuarine regions in mangrove sediments of Mandovi estuary

Bulk metal concentration (Mangrove regions)												
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary			Upper Estuary		
Metals	0-6	6-12	12-20	0-6	6-12	12-22	0-6	6-12	12-22	0-6	6-12	12-22
Mn	1317.5	1428.5	1884.7	2980.83	3093.1	3680.2	1860.3	2379.0	2768.6	2916.5	2482.5	2631.5
Cu	136.17	135	126.55	145.83	138.25	133.71	135.83	139.81	119.17	141.75	140.44	136.94
Zn	66.50	67	70.25	63.83	65	64.67	69.67	70.69	55.63	73.67	72.19	70.13
Cr	91.25	94.19	100.80	27.67	41.94	84.38	100.50	107.31	88.33	107.83	110.44	111.69
Co	23.67	21.44	19.20	40.92	41.44	35.54	23.67	24.81	119.17	24.25	27.19	26.38

5.4.c. *SQUIRT*

Table 5.7. Screening quick reference table for metals in marine sediments (Buchman 1999)

ELEMENTS	TEL	ERL	PEL	ERM	AET
Fe	-	-	-	-	22% (Neanthes)
Mn	-	-	-	-	260 (Neanthes)
Cu	18.7	34	108	207	390 (Microtox & oyster larvey)
Zn	124	150	271	410	410 (Infaunal community)
Cr	52.3	81	160	370	62 (Neanthes)
Co	-	-	-	-	10 (Neanthes)

TEL: Threshold effect level; ERL: Effect range low; PEL: Probable effects level; ERM: Effect range median; AET: Apparent effects threshold
(Except for Fe, all values are in *u*g/g)

Screening Quick Reference Table (SQUIRT) developed by (NOAA) is presented in Table 5.7. Based on SQUIRT, the guideline values are categorized into five classes which are presented in the Table 5.8 (Buchman, 1999).

Table 5.8. Sediment guidelines and terms used in SQUIRT

Sediment guidelines	
Threshold Effect Level (TEL)	Maximum concentration at which no toxic effects are observed
Effects Range Low (ERL)	10th percentile values in effects or toxicity may begin to be observed in sensitive species
Probable Effects Level (PEL)	Lower limit of concentration at which toxic effects are observed
Effects Range Median (ERM)	50th percentile value in effects
Apparent effects Threshold (AET)	Concentration above which adverse biological impacts are observed

When the speciation data is compared with SQUIRT (Table 5.7), Mn shows values above AET indicating Mn is bioavailable and toxic to biota in mudflats of lower middle estuary and upper middle estuarine regions (Table 5A.9). In mangroves, Mn values are higher than AET in lower estuary, lower middle and upper middle regions and thus bioavailable Mn is toxic to biota (Table 5B.9). Similarly, Co data is compared with SQUIRT and the results show the Co concentration is less than AET in lower estuary and higher values i.e. above AET in mudflats of lower middle and upper middle regions of the estuary at all the depths and thus is toxic to biota. Mangrove regions show less Co value i.e. below AET and thus are considered as safe. Pb is also higher in bioavailable phase within mudflats but noted higher values in residual phase within mangroves. Pb values computed for both mudflats and mangroves are presented in Tables 5A.9 and 5B.9 respectively. However, Pb values could not be compared with the SQUIRT values as published list does not include Pb.

Table 5A.9. Average concentration of metals in bioavailable fractions at different sediment depths (cm) of lower, lower middle and upper middle regions in mudflat sediments of Mandovi estuary

Metal speciation (Mudflat regions)									
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary		
Metals	0-6	6-12	12-18	0-6	6-12	12-18	0-6	6-12	12-20
Mn	208.25	176.05	140.08	1836.6	1809.18	1834.0	1713.5	1396.5	1699.6
Co	8.72	8.4	7.18	21.54	21.01	20.77	25.24	23.39	24.89
Pb	23.28	21.43	14.48	38.94	43.12	34.71	38.2	51.58	47.40

Table 5B.9. Average concentration of metals in bioavailable fractions at different sediment depths (cm) of lower, lower middle and upper middle regions in mangrove sediments of Mandovi estuary

Metal speciation (Mangrove regions)									
	Lower Estuary			Lower Middle Estuary			Upper Middle Estuary		
Metals	0-6	6-12	12-20	0-6	6-12	12-22	0-6	6-12	12-22
Mn	1068.9	1285.9	1637.4	2594.4	2520.7	2406.9	1496.4	2096.6	2756.8
Co	4.58	5.15	5.42	6.04	6.19	7.53	7.79	8.45	7.03
Pb	13.41	14.08	13.54	15.39	16.06	16.27	15.97	18.00	15.15

5.4.d. *Risk assessment code (RAC)*

The metals in acid soluble fractions are considered to be the weakest bonded metals in sediments which may equilibrate with the aqueous phase, and thus become more easily bioavailable (Pardo et al., 1990). Thus, metal speciation is of critical importance to their potential toxicity and mobility (Maiz et al., 2000). A criterion called ‘‘Risk Assessment Code (RAC)’’ has been used to assess the potential mobility and hazard of metal based on the percentage of exchangeable and bound to carbonate metal in the sediment (e.g. Perin et al., 1985; Jain, 2004; Ghrefat and Yusuf, 2006). The metals in different fractions are bound with different strengths in the sediments. If a sediment sample can release in these fractions less than 1% of the total metal, it will be considered safe for the environment. On the contrary, sediment releasing in the same fractions more than 50% of the total metal (Table 5.10) has to be considered highly dangerous and can easily enter into the food chain.

Table 5.10. Criteria of Risk Assessment code (Perin et al., 1985)

Grade	Sum of exchangeable and carbonate in % of the total	RAC
I	<1	No risk
II	2- 10	Low risk
III	11 – 30	Medium risk
IV	31 – 50	High risk
V	>50	Very high risk

RAC is applied for Mn, Co and Pb and not applied for Fe and Zn as their concentration in bioavailable fractions is comparatively less than found in residual fractions. Within mudflats in lower estuary, the percentage of Mn associated with the exchangeable and carbonate- bound fraction (F1 and F2) ranges from Grade IV to V indicating high risk to very high risk from bottom to surface sediments (Table 5A.11a). Co and Pb fall in Grade III to IV indicating medium risk to high risk in sediment sections at this location. In lower middle estuarine region (Table 5A.11b) RAC grades from IV in surface and middle section to V in the bottom section for Mn. Co is uniformly showing grade III and Pb grade IV. In the upper middle, Mn grades from IV to III from bottom to surface (Table 5A.11c). Co and Pb show grade III except Pb in bottom section where in it shows grade IV. The results indicate that the metals Mn, Co and Pb within Mandovi estuary pose medium to very high risk for biota.

Table 5A.11. Sum of exchangeable and carbonate fractions in % of the total in lower, lower middle and upper middle regions in mudflat sediments of the estuary

Table 5A.11 a. Sum of exchangeable and carbonate fractions in % of the total metal

b) Lower estuary

Metals	0-6 cm	6 – 12 cm	12 – 18 cm
Mn	54.41 (Grade V)	35.72 (Grade IV)	34.70 (Grade IV)
Co	32.49 (Grade IV)	29.14 (Grade III)	33.62 (Grade IV)
Pb	29.27 (Grade III)	33.50 (Grade IV)	41.96 (Grade IV)

Table 5A.11 b. Sum of exchangeable and carbonate fractions in % of the total metal

a) Lower middle estuary

Metals	0-6 cm	6 – 12 cm	12 – 18 cm
Mn	48.27 (Grade IV)	48.39 (Grade IV)	53.37 (Grade V)
Co	26.92 (Grade III)	25.34 (Grade III)	26.83 (Grade III)
Pb	42.16 (Grade IV)	39.75 (Grade IV)	43.03 (Grade IV)

Table 5A.11 c. Sum of exchangeable and carbonate fractions in % of the total metal

c) Upper middle estuary

Metals	0-6 cm	6 – 12 cm	12 – 20 cm
Mn	21.49 (Grade III)	31.42 (Grade IV)	41.09 (Grade IV)
Co	19.30 (Grade III)	22.55 (Grade III)	18.64 (Grade III)
Pb	29.03 (Grade III)	30.50 (Grade III)	38.69(Grade IV)

Table 5B.11. Sum of exchangeable and carbonate fractions in % of the total in lower, lower middle and upper middle regions in mangrove sediments of the estuary

Table 5B.11a. Sum of exchangeable and carbonate fractions in % of the total metal

a) Lower estuary

Metals	0-6 cm	6 – 12 cm	12 – 20 cm
Mn	63.67 (Grade V)	69.47 (Grade V)	65.53 (Grade V)
Co	12.58 (Grade III)	13.48 (Grade III)	16.13 (Grade III)
Pb	10.49 (Grade II)	10.93 (Grade II)	11.50 (Grade III)

Table 5B.11 b. Sum of exchangeable and carbonate fractions in % of the total metal

b) Lower middle estuary

Metals	0-6 cm	6 – 12 cm	12 – 22 cm
Mn	57.54 (Grade V)	42.66 (Grade IV)	25.62 (Grade III)
Co	2.96 (Grade II)	1.68 (Grade II)	1.29 (Grade II)

Pb	7.96 (Grade II)	8.02 (Grade II)	9.15 (Grade II)

Table 5B.11 c. Sum of exchangeable and carbonate fractions in % of the total metal

c) Upper middle estuary

Metals	0-6 cm	6 – 12 cm	12 – 22 cm
Mn	66.34 (Grade V)	65.00 (Grade V)	60.86 (Grade V)
Co	10.05 (Grade II)	10.44 (Grade II)	9.59 (Grade II)
Pb	9.61 (Grade II)	11.32 (Grade III)	12.01 (Grade III)

The results (Table 5B.11a, b, c) indicate that within mangroves Mn falls in grade V indicating very high risk in all the sediment sections of lower estuary and upper middle estuarine region. In lower middle estuarine region, Mn attributed very high risk in surface sediments and high risk in middle sediments (6 – 12) and medium risk in bottom sediments (12 – 18). Co shows medium risk and Pb falls between Grade II – III indicating low – medium risk in lower estuary. Co and Pb show lower risk in all the sediment sections in lower middle estuary. Co shows low risk and Pb shows medium risk in sediment sections of 6 – 12 cm and 12 – 20 cm, whereas, indicates lower risk towards the surface sediments at upper middle estuary.

RAC of Mn shows very high risk therefore, Mn input from anthropogenic source to Mandovi estuary requires constant monitoring.

5.5. Metal tolerant microbes in mudflat sediments

5.5.a. *Lower estuary (Betim)*

The results obtained show higher bacterial counts on the enriched media i.e. Zobell marine agar and the counts are lower in minimal media which is devoid of nutrients (Fig 5A.6a). Retrievable counts of metal tolerant bacteria in metal amended media are given in (Fig. 5A.7a). In general, higher counts of Co and Zn tolerant bacteria are observed in minimal media containing 25 ppm, although Fe and Mn is also prominent but are less than that of Co and Zn in lower estuary (Fig.5A.7a). Dominance of metal tolerant bacteria at 0-6 cm is noted and decreased towards the deeper sediment sections.

5.5.b. *Lower middle estuary (Karyabhat)*

The growth in Zobell and minimal media (Fig. 5A.6b) when observed, the counts in Zobell agar show similar to that in lower estuarine region, however, growth in minimal media is very low compared to the growth in Zobell media and also compared to growth in minimal media of lower estuary. In lower middle estuarine region, except for Mn tolerant bacteria which are dominant other metal tolerant bacteria are comparatively very less (Fig.5A.7b). Trend is almost similar to that observed in case of lower estuary where in metal tolerant bacteria are higher at surface sediment section.

5.5.c. *Upper middle estuary (Ribander)*

The growth in Zobell and minimal media (Fig. 5A.6c) is similar to that in lower middle estuarine region. In upper middle estuarine region, except for Zn tolerant bacteria which show comparatively less counts, rest all the metal tolerant bacteria show higher and prominent counts (Fig.5A.7c). Trend is similar to that observed at other locations wherein the counts decrease towards the deeper sediment sections. Among the metal tolerant bacteria, Co tolerant bacteria is found to show highest counts in lower estuarine region whereas, Pb tolerant bacteria are higher in upper middle region.

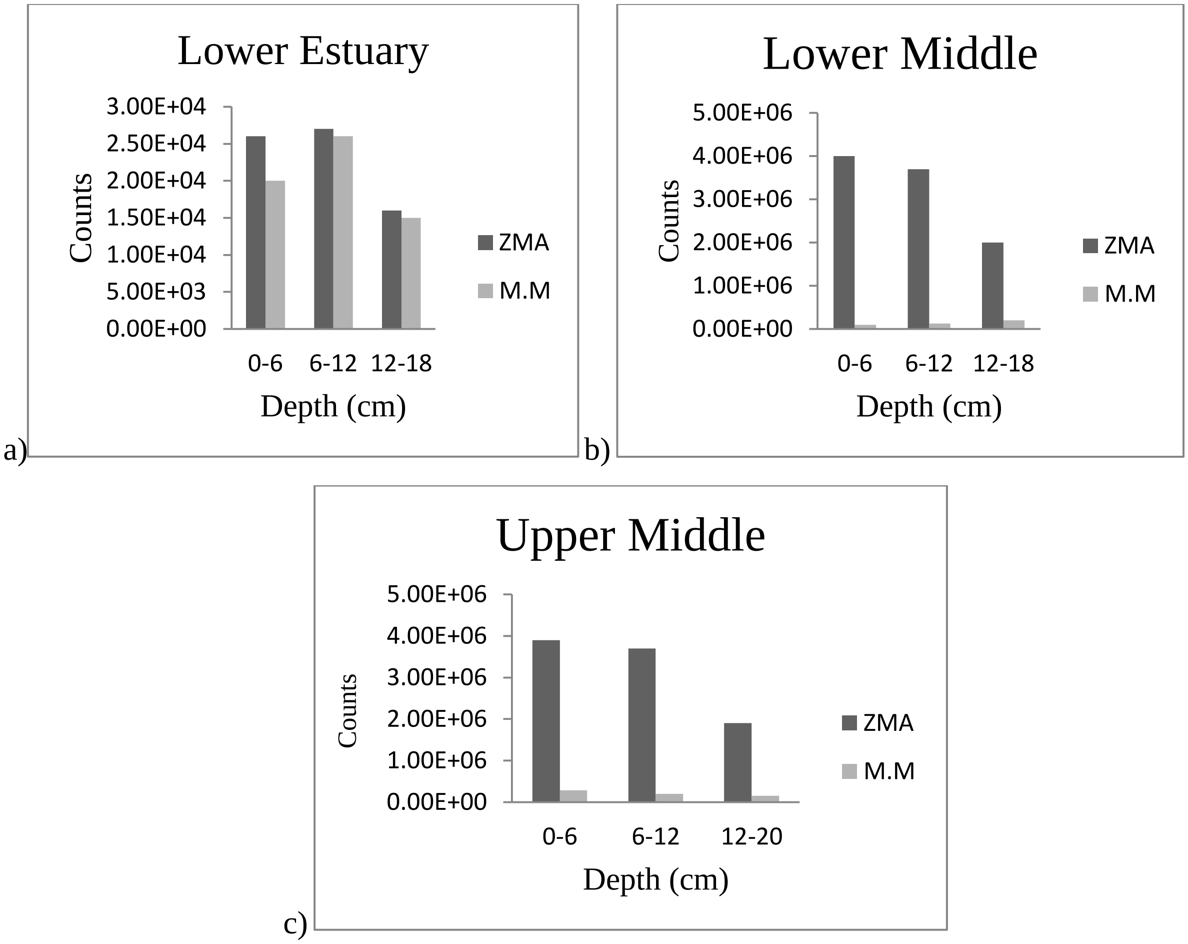
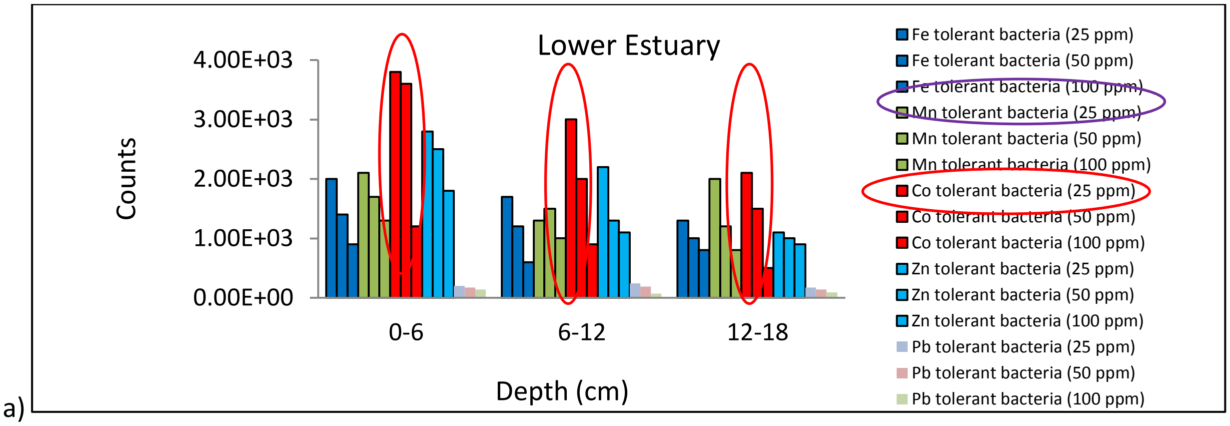


Fig. 5A.6. Total heterotropic counts (CFU g^{-1} sediment) in Zobell marine agar (ZMA) and Minimal media (M.M) in a) Lower estuary b) Lower middle estuary c) Upper middle estuary



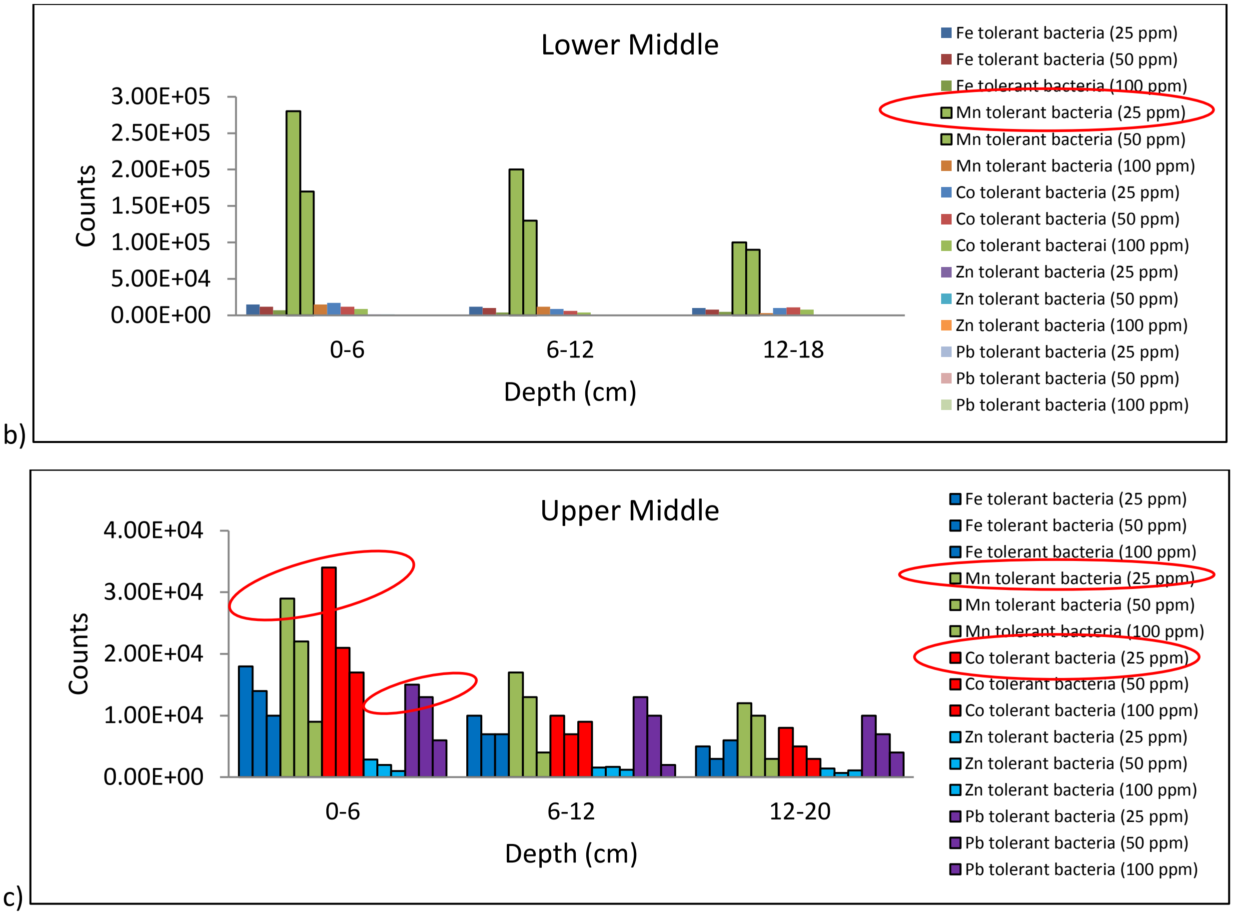


Fig. 5A.7. Metal tolerant bacteria (CFU g^{-1} sediment) in media containing metals such as Fe, Mn, Co, Zn and Pb in different concentrations (25, 50 and 100 ppm) from sediments of a) Lower estuary b) Lower middle estuary c) Upper middle estuary

5.6. Sediment parameters and microbes

Correlation analysis carried out to understand the association of total heterotrophic counts with sediment parameters in mudflat environment of lower estuary, lower middle and upper middle estuary revealed interesting results. In lower estuary it is observed that total heterotrophic counts show positive correlation with sediments components such as clay (r = 0.94) and silt (r = 0.68) and it also shows association with bulk concentration of metals such as Mn (r = 0.81), Co (r = 1.00), Zn (r = 0.93), Pb (r = 0.99) and Fe (r = 0.50). Further, efforts made to establish a relation between bioavailability of the metals within the different fractions and its effect on bacterial population (total heterotrophic counts), show that total bacterial population are significantly correlated with Mn (r = 0.68) in F1 phase and Co (r = 0.76), Zn (r = 0.70), Pb (r = 0.98) in F3 phase in lower estuary.

In lower middle estuarine region, total heterotrophic counts (THC) show significant correlation with clay (r = 0.97). Total metals are not correlated with THC. However, THC is significantly correlated with Mn and Co in F2 bioavailable phase with values of r = 0.99 and r = 0.83 respectively. Further, Zn is associated with THC with value of r = 0.67 in F3 phase. Pb show positive association with THC in F1, F3 and F4 phases.

In upper middle estuary, the THC show significant correlation with sediment components viz. clay (r = 0.72), organic carbon (r = 0.88) and silt (r = 0.95). THC population also show significant correlation with metals viz. Fe (r = 0.92), Mn (r = 0.65), Co (r = 1.00) and Pb (r = 0.96). THC shows significant correlation with Co in both F1 and F3 with values of r = 0.82, 0.88 respectively. Significant correlation of THC with Zn in F1 and F3 phases is also observed with r = 0.85 and r = 0.62 respectively.

Similarly, correlation analysis computed for estuarine sediments of mangrove environment of lower, lower middle and upper middle region, revealed sediment parameters and metals are not significantly correlated with THC in lower estuary. However, THC is significantly correlated with Fe in F2, F3 and F4 fractions with r values of 0.71, 0.82 and 0.79 respectively. THC is also significantly correlated with Mn (r = 0.53) in F3 fraction. Zn is seen to be positively correlated with THC with r values of 0.82 and 0.99 in its F3 and F4 phases whereas, Co with values of r = 0.58 for F2 and r = 0.79 for F3. Pb is also positively correlated with F2 (r = 0.58) and F3 (0.79).

In lower middle estuary, significant correlation is observed with silt (r = 0.99) and pH (0.76) and metals such as Co and Pb with r values of 0.89 and 0.85 respectively with THC. THC also shows significant correlation with Mn in F3 fraction.

Significant correlation of THC with silt (r = 0.96), organic carbon (0.97), Fe (r = 0.77), Co (r = 0.80) and Pb (r = 0.75) is noted in upper middle estuary. Fe in F3 and F1 is positively correlated with THC with r value of 0.99 and 1.00 respectively. THC shows correlation with Mn in F1 and

F4 fraction with r value of 1.00 and 0.74 respectively. Zn bound to F3 and F4 phases also show significant correlation with THC with r values such as 0.96 and 0.99 respectively whereas, Co shows significant association with THC with r values of 1.00 in F3 and F4 fractions.

Among the sediment parameters, finer sediments and organic matter are major factors which regulate mobilisation of metals and show impact on bacterial community structure (Cornall et al., 2013) in mudflat and mangrove environments. Not much role of coarser sediments is observed in the distribution of heterotrophic bacteria. The location specific environmental factors affect retention of metals in sediment, their speciation as well as toxicity of metals (Chariton et al., 2010) and thus influence on bacterial communities. Total metals and their species show significant correlation with bacterial population in almost all the estuarine regions in mudflat environment. Some of the metals like Co, Fe, Mn, Ni and Zn are essential as micronutrients for life processes (Marscher, 1995) however, Pb is persistent environmental pollutant. Interestingly, some natural microbes tolerate very high levels of trace metals although they are toxic. It is important to note that several genetic mechanisms are known in bacteria which maintain intracellular homeostasis of essential metals and regulate resistance against toxic metals. The mechanisms are metal efflux, intracellular sequestration by metal-binding metallothioneins, extracellular sequestration by exopolysaccharides, cell surface biosorption by negative groups, bioprecipitation and redox reactions which are present in microorganisms to counteract heavy metal stress (Kassab and Roane, 2006). Microorganisms must be using one or more of these mechanisms to resist metal stress. The correlation obtained shows that even with higher concentration of metals some of microorganisms survive in toxic conditions.

5.7. Metal species and metal tolerating bacterial population

Table 5A.12. Bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment sections of lower estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Mn) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	**62.26 (208.2)**	2.00E+04	**2.10E+03**	**1.70E+03**	**1.30E+03**
6-12	59.13 (176.05)	**2.60E+04**	1.30E+03	1.50E+03	1.00E+03
12-18	58.21 (140.07)	1.50E+04	2.00E+03	1.20E+03	8.01E+02

Table 5A.13. Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment sections in lower estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Pb) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	66.59 (23.28)	2.00E+04	2.01E+02	1.71E+02	**1.41E+02**
6-12	**70.02 (21.43)**	**2.60E+04**	**2.41E+02**	**1.91E+02**	7.10E+01
12-18	68.10 (14.48)	1.50E+04	1.71E+02	1.41E+02	9.10E+01

Table 5A.14. Bioavailable Co and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Co in different sediment sections in lower estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Co) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	**79.83 (8.7)**	2.00E+04	**3.80E+03**	**3.60E+03**	**1.20E+03**
6-12	72.28 (8.4)	**2.60E+04**	3.00E+03	2.00E+03	9.01E+02
12-18	79.29 (7.18)	1.50E+04	2.10E+03	1.50E+03	5.01E+02

The minimal media as control i.e. without metal spiked with various increasing metal concentrations viz. 25, 50, 100, 200, 400, 600, 800, 1000 ppm are used to study the growth of bacterial population with increasing metal concentration. It shows decrease of metal tolerant bacteria with increasing metal concentration in minimal media in all the estuarine regions. This study was carried out for all the metals viz. Fe, Mn, Zn, Co and Pb, but only those metals which are most bioavailable and show high risk to biota with respect to Risk Assessment Code (RAC) are discussed here. The study revealed the relation existing between metal tolerant bacteria and bioavailability of metals within the estuarine sediments at various depths in mudflats.

In sediments of lower estuary, Mn, Pb and Co show higher bioavailability i.e. higher concentration found in first four fractions and further results of RAC shows Mn, Co and Pb attributed higher risk to biota. In lower estuarine sediments the media without metal (Mn) show higher counts at 6 – 12 cm, although bioavailability is higher in surface sediments. The media with Mn of different concentrations show higher counts at the surface where bioavailability of metals is higher (Table 5A.12). Further in deeper sediment sections the sediments are anoxic and thus metals in such an environment exists as sulphides and this form of metal is not easily consumed by bacterial cells and thus counts of metal tolerant bacteria decreases towards deeper sediments. Same metal injected in different sediments sections with 25, 50 and 100 ppm concentrations show different counts of metal tolerant bacteria. At this location bioavailable concentration of Mn is sufficiently high to cause changes in bacterial community structure.

When Pb is added in minimal media it is observed that bacteria tolerating 100 ppm of Pb show higher concentration at surface (Table 5A.13) whereas, bacteria tolerating Pb at 25 and 50 ppm are more concentrated at depth between 6- 12 cm although bioavailable Pb is higher at surface. Higher Co tolerant bacteria is noted at the surface sediment section whereas, counts decrease towards bottom sediments (Table 5A.14). Bacterial species with increase metal contamination transform from sensitive to tolerant species. The catalytic activity of many enzymes depends on metal ions and thus is part of cofactors (Fontana et al., 2010).

Table 5A.15. Bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment sections of lower middle estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Mn) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	**91.34 (1836.6)**	1.00E+05	**2.80E+05**	**1.70E+05**	**1.50E+04**
6-12	81.78 (1809.18)	1.30E+05	2.00E+05	1.30E+05	1.20E+04
12-18	81.76 (1834.05)	**2.00E+05**	1.00E+05	9.00E+04	3.00E+03

Table 5A.16. Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment sections of lower middle estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Pb) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	70.14 (38.94)	1.00E+05	**1.00E+03**	**9.01E+02**	**4.01E+02**

6-12	**72.64 (43.12)**	1.30E+05	7.01E+02	7.01E+02	2.01E+02
12-18	66.82 (34.71)	**2.00E+05**	5.01E+02	3.01E+02	3.01E+02

In lower middle estuarine region, metals viz. Mn and Pb show high risk to very high risk with respect to RAC. When Mn is spiked in the minimal media with increasing concentration it is observed that metal tolerant bacteria decreases as metal concentration increases. The relation of bioavailable Mn and the counts of metal tolerant bacteria in 25, 50 and 100 ppm are higher in surface sediments (Table 5A.15). The bacterial counts found without metals show higher growth in bottom sediments. Pb tolerant bacteria are dominant at the surface whereas, bacterial counts in control i.e. without metal show higher bacterial population in bottom sediment section. The Pb shows higher bioavailability in middle sediment section (Table 5A.16).

Table 5A.17. Bioavailable Mn and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Mn in different sediment sections of upper middle estuary

CORE SECTION (CM) **(C)**	BIOAVAILABLE FRACTION (Mn) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	33.56 (1713)	**2.80E+05**	**2.90E+04**	**2.20E+04**	**9.00E+03**
6-12	36.69 (1396)	2.00E+05	1.70E+04	1.30E+04	4.00E+03
12-20	**47.17 (1699)**	1.50E+05	1.20E+04	1.00E+04	3.00E+03

Table 5A.18. Bioavailable Pb and microbes in media containing no metal and media with metal concentration of 25, 50 and 100 ppm of Pb in different sediment sections of upper middle estuary

CORE SECTION (cm)	BIOAVAILABLE FRACTION (Pb) (%) (F1+F2+F3+F4) values in bracket correspond to ppm	METAL TOLERANCE (BACTERIAL COUNTS) (CFU g^{-1} sediment)			
		MINIMAL MEDIA (NO METAL)	MINIMAL MEDIA (WITH METAL)		
			25 PPM	50 PPM	100 PPM
0-6	45.89 (38.2)	**2.80E+05**	**1.50E+04**	**1.30E+04**	**6.00E+03**
6-12	**61.97 (51.58)**	2.00E+05	1.30E+04	1.00E+04	2.00E+03
12-20	73.20 (47.40)	1.50E+05	1.00E+04	7.00E+03	4.00E+03

In upper middle region, Mn and Pb show higher risk with regards to RAC. Mn tolerant bacteria are found to be higher in media with metal and also in control towards the surface sediments (Table 5A.17). In case of Pb tolerant bacteria (Table 5A.18) the results is similar to that observed with respect to Mn i.e. counts are more prominent in surface sediments although, bioavailability of both Mn and Pb seems to be higher in the deeper sediments. Therefore, it can be stated that bioavailability of metals alone is not a parameter controlling the microbial growth cycles and there must be additional factors influencing their growth at this location.

The bioavailability of metals is changing depending on the conditions and processes operating at different depths within sediment column. Trace metal speciation is a function of redox potential. Various processes which influence the behaviour and adsorption of metals to different phases in an estuarine environment are diagenesis, redox condition and organic matter degradation, sediment texture and role of meiofauna within the sediment column, which have an important effect on the species of metals thereby effecting mobility. In general, metal tolerant bacterial population is higher at the surface and at some instances at bottom sediments. Even with higher concentration of metals in bioavailable form, some of the microorganisms survive in toxic conditions.

CHAPTER SIX

SUMMARY AND CONCLUSION

Estuaries are one of the important sub environments of the coastal zone. They are the favourable environments for deposition of sediments derived from various sources. Along the west coast of India estuarine processes are controlled largely by south west monsoon controlled seasons. Every component viz. suspended matter, sand, silt, clay, organic carbon, metals and their species have gained importance in estuarine studies to understand natural processes as well as changing environmental conditions. The Mandovi and Zuari rivers are considered as “life line” of Goa and they form an important tropical estuarine system along the west coast of India. Within the catchment area of both the rivers, the iron and manganese ores are mined using open cast method. Estuarine channels of both the rivers are used to transport large quantities of iron and manganese ores to Marmugao harbour. The mining activities in this region influence the geochemical and biological conditions of the estuarine processes.

Estuarine sediments are preferential sites for accumulation of particles of both terrestrial and marine origin and play a key role in the geochemical processes. Further, they are known to accumulate organic matter and metals within sediments. Factors such as grain size, organic matter, geomorphology, prevailing hydrodynamic conditions and different point and non-point sources play a vital role in concentration and distribution of metals in the estuarine system. Metals are known to exist in different forms and are associated with sediment components and organic matter. Remobilization of trace metals is regulated by Fe, Mn and Al. Over the years trace metal concentration increased due to enhanced input of material. Sediments may therefore act as either source or sink for metals. This is also strongly dependent on their chemical forms. Further, particular behaviour of metals in the estuarine environment is determined not only by their total concentration but also by their specific chemical forms.

Mandovi estuary receives input from multiple sources such as natural weathering products, domestic and industrial sewage outfalls, effluents from mines, dredge spoil disposal and agricultural runoff and deposited in the estuarine sub environments viz. mudflats and mangroves. It was therefore felt essential to examine the sediments within mudflats and mangroves of Mandovi estuary. To address this surface sediments collected from mudflats and mangroves along with water samples collected near surface and near bottom of Mandovi estuary in different seasons and core sediments were analysed. This thesis is an attempt to study the spatial and seasonal distribution of metals in mudflats and mangroves with following objectives.

- To study the abundance and distribution of metals in sediments from Mangroves and Mudflats.
- To understand the role of various physicochemical and geochemical parameters in the distribution and concentration of species in Mangroves and Mudflats.
- To investigate metal tolerance of bacterial isolates.

To address the objectives following parameters were analysed using standard methods.

a) Water samples - Salinity, Suspended particulate matter and particulate metals (Fe, Mn, Zn Ni and Co) representing three seasons.

b) Sediment samples - Sediment components (Sand, Silt, Clay and Organic carbon); Metal analysis (Fe, Mn, Al, Cr, Cu, Co, Ni, Zn and Pb); Total Heterotrophic counts (THC); Speciation of metals (Fe, Mn, Zn, Co and Pb) and metal tolerance studies on bacteria (Mn, Co and Pb) for both surface sediment samples representing three seasons and core samples.

The equipment used for the analysing the parameters include Atomic Absorption Spectrophotometer (AAS), Autoclave, Laminar Air Flow, pH meter and others.

The study focuses on understanding various sources and factors in governing the distribution of sediment components including metals in particulates, surface sediments and sub-surface sediment sections. In addition, to gain a precise understanding of the potential and actual impacts of elevated metal level in the sediment, metal speciation has been studied in sediment sections with depth and surface sediments with seasons. Various pollution indices and sediment quality guidelines were employed to know the pollution status. Further, based on metal speciation results, Risk Assessment Code (RAC) was used to assess the environmental risks. An attempt is also made to understand the relation between increasing metal toxicity on associated microbial population. Further, bioavailability of metals was related on survival of microbes and available heterotrophic metal tolerant forms.

Seasonal and spatial variations are observed in concentration of both total suspended matter (TSM) and particulate metals. High suspended matter is observed in pre-monsoon in both surface and bottom waters. The influence of monsoon on concentration of total suspended matter is clearly demonstrated in lower portion of estuary while, non-monsoon season in upper portion of the estuary especially in bottom waters. Post-monsoon and Pre-monsoon seasons exhibited high particulate metals in bottom waters as compared to surface waters. Salinity and Fe-Mn oxyhydroxides are found to be regulating the deposition of total suspended matter and metal adsorption as revealed by Pearson's correlation. The spatial and temporal variation of sediment components, metals and total heterotrophic counts within mudflats and mangroves of Mandovi estuary revealed presence of coarser sediments in lower estuary and finer sediments in middle estuary. Higher total suspended matter in bottom waters and higher finer sediments deposition in middle estuarine regions largely support each other. This is explained as relatively high wave activity at the lower estuary results in complex energy conditions thus facilitating deposition of coarser sediments in mudflats of lower estuary and which result in carrying finer ones to areas of lesser energy environments.

Mudflat sediments of middle estuary exhibited higher metal concentration compared to lower estuary. Deposition of higher finer sediments indicates presence of calmer hydrodynamic conditions and facilitates entrapment and accumulation of metals as they are generally associated with smaller grain size particles. Similarly mangrove sediments of lower middle and upper estuary favoured high metal concentration. Lower middle estuary harbours higher proportion of finer sediments and organic carbon and acts as a sink for metals. It is also important to note that the width of the river gets reduced to less than half as moved towards the upstream region which causes tidal turbulence resulting resuspension. Resuspension of bed load sediments due to tidal surge and geomorphological constriction involves drag and lift force resulting removal of less denser material from the surface sediments, which in turn helps in concentration of denser particles as well as the particles with adsorbed metals and therefore responsible for higher concentration of some of the metals along this region. Heterotrophic bacteria are found to be higher in mudflat sediments of middle estuary and towards middle and upper estuarine regions in mangrove areas. The difference in bacterial densities is related to factors such as nature and size of the sediment along with concentration of organic carbon. The occurrence of higher finer sediment concentration accompanied with higher organic matter seems to determine the heterotrophic counts in Mandovi estuary.

Mudflats of lower estuary show higher coarser sediments as compared to mangroves in all the seasons. Overall, most of the metals show higher concentration in mangrove sediments than mudflat sediments of lower estuary in all the three seasons and to some extent in lower middle estuary. However, in middle estuary, some of the metals show higher concentration in mudflats in different seasons. Seasonal variation in metal concentration between the two sub-environments is affected by addition of metals from different sources. Mangroves of lower estuary and lower middle estuary show higher heterotrophic counts than adjacent mudflats in monsoon and post-monsoon season. However, in upper middle estuary mudflats shows higher heterotrophic counts than mangroves during these seasons. Geoaccumulation indices indicated that mangroves are more enriched with metals compared to mudflats in all the three seasons in different locations of Mandovi estuary.

Sequential extraction technique was also employed to mudflat surface sediments of different seasons. Results reveal bioavailability of Mn is higher in lower middle region in all the three seasons followed by upper middle region and lower estuary. In lower middle estuary, the bioavailability of Mn is higher in pre-monsoon with organic fraction as a major phase. In monsoon carbonate is the main phase. Mn association with carbonate fraction shows its ability to replace calcium in carbonate minerals as a result of their similar ionic radii and charges. Mn is considered as essential micronutrient for growth of the organism. Mn in readily bioavailable phases namely exchangeable and carbonate must have been used during this season by the organisms as this season is known for higher productivity. Bioavailability of Co is found to be higher in lower estuary. Lower middle estuary with high finer sediments, organic matter content and higher bulk metal concentration is found to facilitate higher bioavailability of metals. Further, tides and tidal currents, redistribution of surface sediments must have led to changes in chemical properties of sediments that stimulate the mobilization of metals.

In addition, study carried out to understand distribution of metals (Fe, Mn, Co, Pb and Zn) in mudflat and mangrove regions in sediment depth sections revealed that Fe and Zn are higher in fraction 5 in all the three regions at different depths indicating less bioavailability of Fe and Zn in different sediment sections. This also holds well in the surface sediments of mudflat regions in different seasons. Mangrove sediments of lower estuary and upper middle estuary attributed higher bioavailability of Mn between different sediment sections whereas, higher bioavailability of Mn is noted in mudflat sediment of lower middle estuary between 0-6 cm and 12- 18 cm. Diagenesis could be possible reason for Mn remobilization from the reducing deeper sediment sections and accumulation in oxidized surface sections and thus increases bioavailability in mangrove region. Mangrove roots can diffuse oxygen into the rhizosphere, increasing redox potential in the sediment and this partly accounts for the oxidizing conditions observed at mangrove sites where root density is higher. In addition, anthropogenic input of Mn to the sediments cannot be ruled out. Bioavailability of Co is less in sediments of mangrove regions of lower, lower middle and upper middle at all the depths as that found in mudflat regions. Metals in bioavailable phases must be migrating from mangroves during ebb high tide to the estuarine waters. Adjacent mudflat environment being subjected to a periodic submergence and provide more aerobic condition must be facilitating co-precipitation of Co in this environment. In addition, in mangrove environment plant uptake may be responsible for depletion of metals in exchangeable fraction.

Risk Assessment Code (RAC), applied revealed that Mn mostly exists in the labile fraction and therefore is under high-risk – very high risk category and could easily enter the food chain. Speciation of Co and Pb shows medium to high risk to the aquatic environment. Fe and Zn did not show any risk to the environment. Based on results of Risk Assessment Code (RAC) attempt is made to study metal tolerance (Mn, Co and Pb) within mudflat sediments with respect to their detrimental effect on the bacterial community. Results attributed higher heterotrophic counts in enriched media (ZMA) followed by minimal media and the bacterial counts abruptly decrease with increase of metal toxicity within the media. It is observed that Co tolerant bacteria are significantly higher compared to Mn and Pb tolerant bacteria in lower estuary, whereas, in the lower middle region dominance of Mn tolerant bacteria is noted. In upper middle estuary, the sediments show higher Mn and Co bacterial colonies.

Correlation of heterotrophic counts with associated sediment parameters, bulk metals and metal species reveals that among the sediment parameters, finer sediments and organic matter are major factors which regulated mobilisation of metals and has greater impact on bacterial community structure in mudflat and mangrove environments. Total metals and their species show significant correlation with bacterial population in almost all the estuarine regions in mudflat environments. The correlation obtained shows that even with higher concentration of metals some of microorganisms survive in toxic conditions, though in general with higher bioavailable metal concentration, bacterial population decreases.

REFERENCES

Abeh, T., Gungshik, J. and Adamu, M. M. (2007). Speciation Studies of trace elements levels in sediments from Zaramagada stream in Jos, Plateau State, Nigeria. Journal of Chemical Society of Nigeria, 32(2), 218-225.

Abu-Hilal, A., Badran, M. and Vaugelas, J. (1988). Distrubution of trace elements in Callichirus Laurae burrows and nearby sediments in the Gulf of Aquada, Jordan (Red Sea). Marine Environmental Research, 25, 233-248.

Akcay, H., Oguz, A. and Karapire, C. (2003). Study of heavy metal pollution and speciation in Buyak Menderes and Gediz river sediments. Water Research, 37, 813-822.

Alagarsamy, R. (2006). Distribution and seasonal variation of trace metals in the surface sediments of the Mandovi Estuary, West coast of India. Estuarine Coastal and Shelf Science, 67, 333- 339.

Alavandi, S. (1990). Relationship between heterotrophic bacteria and suspended particulate matter in the Arabian Sea. Indian Journal of Marine Sciences, 30, 89-92.

Alloway, B. J. (1995). The origins of heavy metals in soils. In heavy metals in soils. 2nd edition, (ed. B. J. Alloway): Blackie Academic and Professional.

Alongi, D. M. (1998). Coastal Ecosystem Processes. CRC Press, 419.

Alongi, D. M. (2005). Mangrove-microbe-soil relations. In: Kristensen, E., Haese, R.R., Kostka, J.E. (Eds.), Macro- and Microorganisms in Marine Sediments. American Geophysical Union, Washington DC, 85–103.

Alongi, D. M., Tirendi, F. and Clough, B. F. (2000). Below ground decomposition of organic matter in forests of the mangroves Rhizophorastylosa and Avicenniamarina along the arid coast of Western Austriala. Aquatic Botany, 68 (2), 97- 122.

Anagnostou, C., Garbe-Schonberg, C. D., Kaberi, H. and Krasakopoulou (2000). Cycling of elements associated to particulate matter within a coastal system of the Mediterranean Sea: NW Aegean Sea METRO-MED Project Final Scientific Report.

Anawar, H. M., Yoshioka, T., Konohira, E., Akai, J. and Freitas, S. (2010). Sources of organic carbon and depositional environment in the Bengal delta plain sediments during the Holocene period. Limnology, 11, 133-142.
Anderson, B., Hunt, J., Phillips, B., Thompson, B., Lowe, S., Taberski, K. and Carr, R. S. (2007). Patterns and trends in sediment toxicity in the San Francisco Estuary. Environmental Research, 105, 145 – 155.

Angula, E. (1996). The Tomlinson Pollution Index applied to heavy metal, Mussel–Watch data: A useful index to assess coastal pollution. Science of the Total Environment, 187, 19–56.

Archer, D. and Devol, A. (1992). Benthic oxygen fluxes on the Washington Shelf and Slope – a comparison of in situ microelectrode and chamber flux measurements. Limnology and Oceanography, 37, 614–629.

Arunachalam, J., Emons, H., Krasnodebska, B. and Mohl, C. (1996). Sequential extraction studies on homogenized forest soil samples. The Science of the Total Environment, 181, 147–159.

Avery, S. V. (2001). Metal toxicity in yeast and the role of oxidative stress. Advances in Applied Microbiology, 49, 111–142.

Ayyamperumal, T., Jonathan, M. P., Srinivasalu, S., Armstrong, J. S. and Ram, M. (2006). Assessment of acid leachable trace metals in sediment cores from River Uppanar Cuddalore, South east. Environmental Pollution, 143 (1), 34-45.

Balachandran, K. K., Laluraj, C. M., Nair, M., Joseph, T., Sheeba, P. and Venugopal, P. (2005). Heavy metal accumulation in a flow restricted, tropical estuary. Estuarine, Coastal and Shelf Science, 65, 361–370.

Barreiro, B. (1991). Estudio de metales pesados en medio y organisms de unecosistema de Ria (Pontedeume, A. Coruna), Thesis Doctoral.

Baruah, N. K., Kotoky, P., Bhattacharyya, K. G. and Borah, G. C. (1996). Metal speciation in Jhanji River sediments. The Science of Total Environment, 193, 1–12.

Beavis, F. R., Ellis, D. J., Welch, S. A. and Kirste, D. (2006). Geochemistry of sediments in response to urbanization in merimbula/pambula estuaries. In proceeding of the CRC Leme Regolith Consolidation and dispersion of Ideas, 14-17.

Bendell-Young, L. I. and Harvey, H. H. (1992). The relative importance of manganese and iron oxides and organic matter in the sorption of trace metals by surficial lake sediments. Geochimica et Cosmochimica Acta, 56, 1175–1186.

Bianchi, A. and Garcin, J. (1993). In stratified waters the metabolic rate of deep-sea bacteria decreases with decompression. Deep-Sea Research - I, 40, 1703–1710.

Biksham, G., Subramaniam, V. and Van Griken, R. (1991). Heavy metal distribution in the Godavari river basin. Environmental Geology and Water Sciences, 17, 117–126.

Bilali, L., Rasmussen, P. E., Hall, G. E. M. and Fortin, D. (2002). Role of sediment composition in trace metal distribution in lake sediments. Applied Geochemistry, 17(9), 1171 – 1181.

Billerbeck, M., Werner, U., Polerecky, L., Walpersdorf, E., De Beer, D. and Hueltel, M. (2006). Surficial and deep pore water circulation governs spatial and temporal scales of nutrient recycling in intertidal sand flat sediment. Marine Ecology Progress Series, 326, 61-76.

Birch, G. (2003). A scheme for assessing human impact on coastal aquatic environment using sediment. In: C. D. Woodcoffe R. A. Furness (eds), Coastal GIS, Wollongong University, Paper in centre for Maritime policy, 14, Austrialia.

Bird, G., Brewer, P. A., Macklin, M. G., Serban, M., Balteanu, D. and Driga, B. (2005). Heavy metal contamination in the Aries river catchment, western Romania: implications for development of the Rosia Montana gold deposit. J. Geochemistry Exploration, 86, 26–48.

Boer, S. I., Arnosh, C., Beusekon Van, J. E. E. and Boetins, A. (2009). Temporal variation in microbial activities and carbon turnover in sub-tidal sandy sediments. Biogeosciences, 6, 1149-1165.

Bordas, F. and Bourg, A. (2001). Effect of solid/liquid ratio on the remobilization of Cu, Pb, Cd and Zn from polluted river sediment. Water Air Soil Pollution, 128, 391-400.

Boullion, S., Borges, A. V., Moya, C. E., Diele, K., DIttman, T., Duke, N. C., Kristensen, E., Lee, S. Y., Marchand, C., Middle burg, J. J., Monroy, V. H. R., Smith, T. J. and Wiley, R. R. (2008). Mangrove production and carbon sinks: A revision of global budget estimates. Global Biogeochemical Cycles, 22.

Bouillon, S., Moens, T., Overmeer, L., Koedam, N. and Dehairs, F. (2004). Resource utilisation patterns of epifauna from mangrove forests with contrasting inputs of local versus imported organic matter. Marine Ecological Progress Series, 278, 77 -88.

Bruins, M. R., Kapil, S. and Oehme, F. W. (2000). Microbial resistance to metals in the environment. Ecotoxicology and Environmental Safety, 45, 198–207.

Bruland, K. W. (1983). Trace elements in seawater. In chemical oceanography, 8 (ed. J.P. Riley and R. Chester), 157-220, Academic Press.

Buchman, M. F. (1999). NOAA screening quick reference tables. NOAA HAZMAT Report 99-1, (p. 12). Seattle, WA, Coastal Protection and Restoration Division, National Oceanic and Atmospheric Administration.

Burgos, M. and Rainbow, P. (2001). Availability of cadmium and zinc from sewage sludge to the flounder, *Platichthys flesus*, via a marine food chain. Marine Environmental Research, 51, 417-439.

Buykx, S. E. J., Bleijenberg, M., Van Der Hoop, M. A. G. T. and Loch, J. P. G. (2000). The effects of oxidation and acidification on the speciation of heavy metals in sulfide-rich water sediments using a sequential extraction procedure. Journal of Environmental Monitoring, 2, 23–27.

Calmano, W. and Forstner, U. (1983). Chemical extraction of heavy metals in polluted river sediments in central Europe. The Science of the Total Environment, 28, 77–90.

Campbell, P. G. C. (1995). Interactions between trace metals and aquatic organisms: A critique of the free-ion activity model. In A. Tessier, & D. Turner (Eds.), Metal speciation and bioavailability in aquatic systems England Wiley, 45–102.

Canfield, D. E. (1989). Reactive iron in marine sediments. Geochimica et Cosmochimica Acta, 53, 619– 632.

Canfield, D. E., Raiswell, R. and Bottrell, S. H. (1992). The reactivity of sedimentary iron minerals towards sulphide. American Journal of Science, 30, 604-608.

Carrel, D. G. (1995). Assessment of the Effects of Urbanisation on Peak Flow Rates', Proceedings of the Second International Symposium on Urban Stormwater Management, Integrated Management of Urban Environments, Institution of Engineers, Australia, 201-208.

Chakraborty, P., Raghunadh Babu, P. V. and Sarma, V. V. (2012). A study of Lead and Cadmium speciation in some estuarine and coastal sediments. Chemical Geology, 217- 225.

Chamley, H. (1989). Clay sedimentology, Springer-Verlag, Berlin, 623.

Chandrasekhar, K., Chary, N. S., Kamala, C. T., Raj, S. and Sreenivasa, R. (2003). Fractionation studies and bioaccumulation of sediment bound heavy metals in Kolleru lake by edible fish. Environmental International, 29, 1001-1008.

Chao, T. T. (1984). Use of participle dissolution techniques in geochemical exploration. Journal of Geochemistry Exploration, 20, 101-135.

Chapman, P. M., Wang, F., Janssen, C., Persoone, G. and Allen, H. E. (1988). Ecotoxicology of metals in aquatic sediments: binding and release, bioavailibility, risk assessment and bioremediation. Canadian Journal of Fisheries and Aquatic Sciences, 55 (10), 2221-2243.

Chariton, A. A., Roach, A. C., Simpsons, S. L. and Batley, G. E. (2010). Influence of the choice of physical and chemical variables on interpreting patterns of sediment contaminants and their relationships with estuarine macrobenthic communities. Marine Freshwater Research, 61, 1109-1122.

Chen, M. S., Wartel, S. and Tammerman, S. (2005). Seasonal variation of floc characteristics on tidal flats, the Scheldt Estuary. Hydrobiologia, 540, 181-195.

Chen, S. L., Zhang, G. A., Yang, S. L. and Shi, J. Z. (2006). Temporal variations of fine suspended sediment concentrations in the Changjang River estuary and adjacent coastal waters, China. Journal of Hydrology, 331, 132–145.

Chen, W. Y. (1992). Sediment transport and sediment dynamic environment of the mudflat with reference to the northern bank of Hangzhou Bay and southern bank of Changjiang Estuary (in Chinese with an English abstract). Acta Oceanologia Sinica, 13, 813–821.

Che, O. R. G. (1999). Concentration of 7 Heavy Metals in Sediments and Mangrove Root Samples from Mai Po, Hong Kong. Department of Applied Science, Hong Kong Technical College, Chai Wan, Hong Kong, People's Republic of China. Marine Pollution Bulletin, 39, 269-279.

Chester, R. (1990a). Marine Geochemistry, Unwin Hyman, London.

Chester, R. (1988). The solid state speciation of copper in surface water particulates and oceanic sediments. Marine Chemistry, 24, 261-292.

Chiffoleau, J. F., Cossa, D., Auger, D. and Truquet, I. (1994). Trace metal distribution, partition and fluxes in the Seine estuary (France) in low discharge. Marine Chemistry, 47, 145-158.

Chong, K. and Wang, W. X. (2000). Bioavailibility of sediment bound Cd, Cr and Zn to the green mussels Perna Viridis and the Manila Clam Ruditapes Philippinarum. Journal of Experimental Marine Biology and Ecology, 255, 75-92.

Clark, M. W., Mc Conchie, D., Lewis, D. W. and Saenger, P. (1998). Redox stratification and heavy metal partitioning in Avicennia - dominated mangrove sediments, a geochemical model. Chemical Geology, 149, 147 - 17l.

Coetzee, P. P. (1993). Determination of Speciation of Heavy Metals in Sediments of the Hortbees port dam by sequential chemical extraction. Water Research, 9(4), 291-300.

Cornall, A. M., Beyer, S., Rose, A., Joyce, C. S., McGuinness, K., Party, D. and Gibb, K. (2013). HCl extractable metal profiles correlate with bacterial population shift in metal- impacted Anoxic coastal sediments from Wet/ Dry Tropics. Geomicrobiology Journal, 30 (1), 48- 60.

Cossa, D. (1990). Seasonality in iron and manganese concentrations of the St. Lawrence River. Science of Total Environment, 98, 185–190.

Cundy, A. B., Croudace, I. W., Thomson, J. and Lewis, J. T. (1997). Reliability of salt marshes as ‘geochemical recorders’ of pollution input: a case study from contrasting estuaries in southern England. Environmental Science and Technology, 31, 1093–1101.

Dassenakis, M., Andrianos, H., Depiazi, G., Konstantas, A., Karabela, M., Sakellari, A. and Scoullos, M. (2003). The use of various methods for the study of metal pollution in marine sediments, the case of Euvoikos Gulf, Greece. Applied chemistry, 18, 781-794.

Davutluoglu, O. I., Seckin, G., Ersu, C. B., Yilmaz, T. and Sari, B. (2011). Heavy metal content and distribution in surface sediments of the Seyhan River, Turkey. Journal of Environmental Management, 92, 2250-2259.

Dellwig, O., Beck, M., Lemke, A., Lunau, M., Kolditz, K., Schnetger, B. and Brumsack, H. J. (2007a). Non-conservative behaviour of molybdenum in coastal waters: coupling geochemical, biological, and sedimentological processes. Geochimica Cosmochimica Acta, 71, 2745–2761.

Denton, G. R. W., Wood, H. R., Conception, L. P., Seigrist, H. G., Efflin, V. S., Nargis, D. K. and Pangelinan, G. T. (1997). Analysis of In- place contamination in Marine Sediments from Four Harbour Locations on Guam: A pilot study, water and Environmental Research Institute of Western Pacific, Technical Report No. 87, University of Guam, Mangilao, Guam.

Dessai, D. V. G. and Nayak, G. N. (2009). Distribution and speciation of selected metals in surface sediments, from the tropical Zuari estuary, central west coast of India. Environmental Monitoring and Assessment, 158, 117–137.

Dong, D., Nelson, Y. M., Lion, L. W., Shuler, M. L. and Ghiorse, W. C. (2000). Adsorption of Pb and Cd onto metal oxides and organic material in natural surface coatings as determined by selective extractions: new evidence for the importance of Mn and Fe oxides. Water Research, 34, 427– 436.

Drever, J. I. (1997). The Geochemistry of Natural Waters: Surface and Groundwater Environments 3rd Ed Prentice Hall Upper Saddle River NJ.

Dunbabin, J. S. and Bowmer, K. H. (1992). Potential use of constructed wet lands for treatment of industrial waste waters containing metals. Science of Total Environment, 111, 151-168.

Dyer, K. R. (1972). Sedimentation in Estauries, In: The estuarine environment, R. S. K. Barnes and J. Grcen (eds), Applied Science publiser, London, UK, 10-32.

Edzwald, J. K. and O'Melia, C. R. (1975). Clay distribution in recent estuarine sediments. Clays and Clay minerals, 23, 39–44.

Edzwald, J. K., Upchurch, J. B. and O'Melia, C. R. (1975). Coagulation in estuaries. Environment Science and Technology, 8, 58-63.

Ehrlich, H. L. and Newman, D. K. (2009). Geomicrobiology, 5th edn. Boca Raton, FL: CRC Press/Taylor & Francis.

Ergin, M., Kazan, B. and Ediger, V. (1996). Source and depositional controls on the heavy metal distribution in the marine sediments of the Gulf of Iskenderum, Eastern Mediterrarean. Marine Geology, 133, 223-229.

Falco, D. G., Magni, P., Teravuosri, L. M. H. and Matteucci, G. (2004). Sediment grain size and organic carbon distribution in the Cabras Lagoon, Sardinia, West Mediterranean. Chemistry and Ecology, 20, S367- S377.

Feely, R. A., Massoth, G. J., Baker, E. T., Gendron, J. F., Paulson, A. J. and Crecelius, E. A. (1986). Seasonal and vertical variations in the elemental composition of suspended and settling particulate matter in Puget Sound, Washington. Estuarine Coastal and Shelf Science, 22, 215-239.

Fernandes, H. M. (1997). Heavy metal distribution in sediments and ecological risk assessment: the role of diagenetic process in reducing metal toxicity in bottom sediments. Environmental Pollution, 97, 317 – 325.

Finney, B. P. and Huy, C. (1989). History of metal pollution in the Southern California Bight: an update. Environment and Science and Technology, 23, 194-303.

Fisher, H., Wanner, S. C. and Pusch, M. (2002). Bacterial abundance and production in river sediments as related to the biochemical composition of particulate organic matter (POM). Biogeochemistry, 61, 37-55.

Flory, P., Kerkar, S. and Krishnan, K. P. (2012). Bacterial response to dynamic metal concentrations in the surface sediments of a solar saltern, Goa, India. Environmental Monitoring and Assessment, 88, 217-233.

Folk, R. L. (1968). Petro Sedi Rocks, Hemphills, Austin, 177.

Fomina, M., Hillier, S., Charnock, J. M., Melville, K., Alexander, I. J. and Gadd, G. M. (2005a). Role of oxalic acid over-excretion in toxic metal mineral transformations by Beauveria caledonica. Applied Environmental Microbiology, 71, 371–381.

Fong, P., Zedler, J. and Donohoe, R. (1993). Nitrogen v/s Phosphorus limitation of algal biomass in shallow coastal lagoons. Limnology and Oceanography, 38, 906 - 923.

Fontana, L. F., Filho, J. G. M., Netto, A. D. P., Santos- Sabadinii, E., Figueiredo, A. G. and Grapez, M. A. C. (2010). Geomicrobiology of cores from Surui Mangrove- Guanabara Bay- Brazil. Marine Pollution Bulletin, 60, 1674 – 1681.

Fontana, L. Partridge, L. and Longo, V.D. (2010). Extending healthy life span- From Yeast to Humans. Science, 321–326.

Forstner, U. and Wittmann, G. T. W. (1979). Metal Pollution in the Aquatic Environment. Berlin, Springer-Verlag, 486.

Fox, L. E. And Wofsy, S. C. (1983). Kinetics of removal of iron colloids from estuaries. Geochimica et Cosmochimica Acta, 47, 211- 216.

Fox, W. M., Johnson, M. S., Jones, S. R., Leah, R. T. and Copplestone, D. (1999). The use of sediment cores from stable and developing salt marshes to reconstruct historical contamination profiles in the Mersey Estuary, UK. Marine Environmental Research, 47, 311–329.

Furukawa, K. and Wolanski, E. (1996). Sedimentation in Mangrove forests. Mangroves and Salt marshes, 1, 3 – 10.

Gadd, G. M. and Griffiths, A. J. (1978). Microorganisms and heavy metal toxicity. Mycological Ecological, 4, 303–317.

Gadd, G. M. (2007). Geomycology: biogeochemical transformations of rocks, minerals, metals and radionuclides by fungi, bioweathering and bioremediation. Mycological Research, 111, 3–49.

Gadd, G. M. (1992a). Metals and microorganisms: a problem of definition. FEMS Microbiological Letters, 100, 197–204.

Gallon, C., Tessier, A., Gobeil, C., Alfaro-De, La. and Torre, M. C. (2004). Modeling diagenesis of lead in sediments of a Canadian shield lake. Geochimica et Cosmochimica Acta, 68, 3531–3545.

Gardolinski, P. C., Hanrahan, G., Achterberg, E. P., Gledhill, M., Tappin, A. D, House, W. A. and Worsfold, P. J. (2002). Comparison of sample storage protocols for the determination of nutrients in natural waters. Water Research, 15, 3670–3678.

Gaudette H. E., Flight, W. R., Toner, L. and Folgerm, D. W. (1974). An inexpensive titration method for the determination of organic carbon in recent sediments. Journal of Sedimentary and Petrology, 44, 249 - 253.

Ghrefat, H. and Yusuf, N. (2006). Assessing Mn, Fe, Cu, Zn, and Cd pollution in bottom sediments of Wadi Al-Arab Dam, Jordan. Chemosphere, 65, 2114–2121.

Giller, K. E, Witter, E. and McGrath, S. P. (2009) Heavy metals and soil microbes. Soil Biology and Biochemistry, 41, 2031–2037.

Gleyzes, C., Tellier, S. and Astruc, M. (2002). Fractionation studies of trace elements in contaminated soils and sediments: a review of sequential extraction procedures. Trends in Analytical Chemistry, 21, 451–467.

Gobeil, C., Macdonald, R. W. and Sundby, B. (1997). Diagenetic sep- aration of cadmium and manganese in suboxic continental margin sediments. Geochimica et Cosmochimica Acta, 61, 4647-4654.

Goldhaber, M. B. and Kaplan, I. R. (1974). The sulphur cycle. In E. D. Goldberg (Ed.), The Sea, New York, Wiley, 569-656.

Gong, P., Siciliano, S. D., Srivastava, S., Greer, C. W. and Sunahara, G. I. (2003). Assessment of pollution induced microbial community tolerance of heavy metals in soils using ammonia-oxidising bacteria and biology assay. Human and Ecological Risk Assessment, 8 (5), 1067-1081.

Gonneea, M. E., Patyan, A. and Herrera-Silveira, J. A. (2004). Tracing organic matter sources and carbon burial in mangrove sediments over the past 160 years. Estuarine Coastal and Shelf Sciences, 61, 211-227.

Gonzalez, H. and Brugmann, L. (1991). Heavy metals in littoral deposits off Havana City, Cuba. Chemistry and Ecology, 5, 171-179.

Gonzalez, J., Boutier, B., Chiffoleau, D., Auger, D., Noel, J. and Truquet, I. (1991). Distribution of Cd and Hg in the Bay of Marenness- Oleron. Ocenologica Acta, 559- 568.

Gonzalez, M. J., Ramos, L. and Hernandez, L. M. (1994). Distribution of trace metals in sediments and their relationship with their accumulation in earthworms. International Journal of Environmental Analytical Chemistry, 57, 135-150.

Gounou, C., Bousserrhine, N., Varrault, G. and Mouchel, J. M. (2010). Inflence of the Iron-Reducing Bacteria on the release of heavy metals in Anaeobic River sediment. Water Air Soil Pollution, 212, 123- 139.

Govindarajan, R. K., Mathivanam, K., Srinivasan, R., Priyadharsini, J. I. and Rajaram, R. (2012). Investigation of anthropogenic influence on aquatic ecosystem quality along the Cuddalore coastal area, S. E. coast of Tamil nadu, India. Global Journal of Environmental Research, 6 (1), 44- 50.

Grant, J. A. (1986). The isocon diagram—a simple solution to Gresen's equation for metasomatic alteration. Economical Geology, 81, 1976 – 1982.

Grasshoff, K. (1983) Determination of nitrate. In: GRASSHOFF, K.; EHRHARDT, M.; KREMLING, K. (Eds.): Methods of Seawater Analysis. Weinheim: Verlag Chemie. 143.

Griffin, J. J., Windom, H. and Goldberg, E. D. (1968). The distribution of clay minerals in the World Ocean. Deep Sea Research, 15, 433-459.

Groengroeft, A., Jaehnig, U., Miehlich, G., Lueschow, R., Maass, V. and Stachel, B. (1998). Distribution of metals in sediments of the Elbe Estuary in 1994. Water Science and Technology, 37, 109–116.

Harbison, P. (1986). Harbison Mangrove muds: a sink and a source for trace metals. Marine Pollution Bulletin, 17, 246–250.

Harikumar, P. S. and Jisha, T. S. (2010). Distribution pattern of trace metal pollutants in the sediments of an urban wetland in the south west coast of India. International Journal of Engineering Science and Technology, 2 (5), 840 – 850.

Harter, R. D. (1992). Competative sorption of cobalt, copper and nickel ions by a calcium - saturated soil. Journal of Soil, Sediment and Association, 56 (2), 444 - 449.

Hatje, V., Apte, S. C., Hales, L. T. and Birch, G. F. (2003). Dissolved trace metal distribution in Port Jackson estuary (Sydnet Harbour), Australia, Marine Pollution Bulletin, 46, 428-434.

Heltai, G., Fehér, B. and Horváth, M. (2005). Element specific detection in speciation analysis using microwave induced plasma atomic emission spectrometry with high performance nebulization techniques, Colloquium Spectroscopicum Internationale XXXIV, University of Antwerp, Belgium, 187.

Hiller, S. (2003). In: G.V. Middleton, M. J. Church, M. Coniglio, L. A. Hardi and F. J. Longstaffe (eds.) Encyclopedia of Sediments and Sedimentary Rocks. Kluwer Academic, Publishers, Dordrecht. Clay mineralogy, 139-142.

Ho, H. H., Swennen, R., Cappuyns, V., Vassilieva, T. V. G. and Tran, V. T. (2013). Speciation and mobility of selected trace metals (As, Cu, Mn, Pb and Zn) in sediment with depth in Cam River – Mouth, Haiphong, Vietnam. Aguatic. Geochemistry, 19, 57 – 75.

Holden, J. F. and Adams, M. W. W. (2003). Microbe–metal interactions in marine hydrothermal vents. Current Opinion in Chemical Biology, 7, 160–165.

Horowitz, A. J. (1991). A primer on sediment Trace- element chemistry: Chelsea, Michigan, Lewis Publishers, 126.

Horsfall, M. and Spiff, A. (2005). Speciation and bioavailability of heavy metals in sediment of Diobu River, Port Harcourt, Nigeria. European Journal of Science Research, 6(3), 20-36.

Hossain, S. B. and Eyre, M. D. (2001). Suspended sediment transport dynamics in the sub-tropical micro-tidal Richmond River estuary, Australia. Estuarine Coastal Shelf Science, 52, 529–541.

Howari, A. and Banat, J. (2001). Assessment of Fe, Zn, Cd, Hg and Pb in the Jordan and Yarmouk River sediments in relation to their physicochemical properties and sequential extraction characterization. Water Air Soil Pollution, 1321, 43-59.

Huerta-Diaz, M. A. and Morse, J. W. (1992). Pyritisation of trace metals in anoxic marine sediments. Geochimica et Cosmochimica Acta, 56, 55-70.

Hui, Z., Zhong, Y., Ming, Z., Chang, Z., Juan, Y., Hua-Jua, H., Zhi-Feng, L. and Hong, J. (2012). Ecological risk assessment of heavy metals in sediments of Xiawan Port based on modified potential ecological risk index. Journal of Transmission of Non ferrous Metals Society, China, 22, 1470-1477.

Ibhadon, A. O., Wright, P. and Daniels, R. (2004). Trace metal speciation and contamination in an intertidal estuary. Journal of Environmental Monitoring and Assessment, 6, 679 –683.

Isuare, M., Manceau, A., Geoffrey, N., Laboudigue, A. and Tamura, N. (2005). Zinc mobility and speciation in soil covered by contaminated dredged sediment using micrometer-scale and bulk-averaging X-ray fluorescence, adsorption and diffraction techniques. Geochimica et Cosmochimica Acta, 69 (5), 1173-1198.

Iwegbue, C. M. A. (2007). Metal fractionation in soil profiles at automobile mechanic waste dumps. Waste Management Research, 25, 1-9.

Jagtap, T. G. (1987). Seasonal distribution of organic matter in mangrove environment of Goa. Indian Journal of Marine Science, 16, 103 – 106.

Jain, C. K. and Sharma, C. K. (2001). Distribution of trace metals in the Hindon River System, India. Journal of Hydrology, 253, 81 – 90.

Jain, C. K. (2004). Metal fractionation study on bed sediments of River Yamuna, India. Water Research, 38, 569–578.

James, R. O. (1977). The adsorption of aqueous heavy metals on inorganic minerals. Geochimica Cosmochimica Acta, 41, 1549– 1555.

Janaki-Raman, D., Jonathan, M. P., Srinivasalu, S., Armstrong-Altrin, J. S., Mohan, S. P. and Rammohan, V. (2007). Trace metal enrichments in core sediments in Muthupet mangroves, SE coast of India: Application of acid leachable technique. Environmental Pollution, 145, 245-257.

Johnson, W. P. and Logan, B. E. (1996). Enhanced Transport of Bacteria in Porous Media by Sediment- Phase and Aqueous-Phase Natural Organic Matter. Water Research, 30, 923-931.

Jonathan, M. P., Ram-Mohan, V. and Srinivasalu, S. (2004). Geochemical variations of major and trace elements in recent sediments, off the Gulf of Mannar, southeast coast of India. Environmental Geology, 45, 466-480.

Jones, B. and Turki, A. (1997). Distribution and Speciation of heavy metals in surficial sediments from the Tees Estuary, North-east England. Marine Pollution Bulletin, 34, 768–779.

Kanellopoulos, T. D., Angelidis, M. O., Karageorgis, A. P., Kaberi, H., Kapsimalis, V., Furukawa, Y., Wolanski, E. and Mueller, H. (2006). Currents and sediment transport in mangrove forests. Estuarine, Coastal and Shelf Science, 44, 301–310.

Kassab, D. M. and Roane, T. M. (2006). Differential responses of a mine tailings *Pseudomonas* isolate to cadmium and lead exposures. Biodegradation, 17, 379–387.

Kennish, M. J. (1998). Trace- metal sediment dynamics in estuaries: Pollution Assessment. In: Ware, G. W. (Ed). Review Environmental Contamination Toxicology, Springer-Berlin, 155, 69-110.

Kerdijk, H. N. and Salomons, W. (1981). Heavy metal cycling in the Scheldt Estaury. Delft Hydraulics Report M1640/M1736 (in dutch), 65.

Kessarker, P. M., Rao, V. P., Shynu, R., Mehra, P. and Veigas, B. E. (2010). The Nature and distribution of particulate matter in the Mandovi Estuary, Central West Coast of India. Estuaries and Coasts, 33, 30-44.

Kiratli, N. and Ergin, M. (1996). Partitioning of heavy metals in surface Black Sea sediments. Applied Geochemistry, 11, 775–788.

Koschinsky, A. (2001). Sequential leaching of Peru Basin surface sediment for the assessment of aged and fresh heavy metal association and mobility. Deep Sea Research Part II, 48 (17-18), 3683- 3699.

Krauskopf, K. B. and Bird, D. K. (1995). Introduction to Geochemistry, third ed. McGraw-Hill, New York, 589-591.

Krone, R. B. (1962). Flume studies of the transport of sediment in estuarine shoaling process, Hyd. Eng. Lab and sanitary Eng. Lab., University of California, Berkeley.

Krupadam, R. J. and Anjaneyulu, Y. (2002). 'Hydrogarphic features and distribution of nutrients in the Kakinada bay, East Coast of India'. Asian Journal of Microbiology, Biotechnology and Environment Sciences, 4(1), 21–36.

Kumar, S. P. and Edward, P. J. K. (2009). Assessment of metal concentration in the sediment cores of Manakudy Estuary, South west coast of India. Indian Journal Marine Science, 38 (2) 235-248.

Kumaran, K. P. N., Nair, K. M., Shindikar, M., Limaye, R. B. and Padmalal, D. (2005). Stratigraphical and Palynological appraisal of the Late Quaternary mangrove deposits of the west coast of India. Quaternary Research, 64 (3), 418-431.

Kuo, S., Heilman, P. E. and Baker, A. S. (1983). Distribution and forms of copper, zinc cadmium, iron and manganese in soils near a copper smelter. Soil Science, 135, 101–119.

Lacerda, L, D., Carvalho, C. E. V., Tanizaki, K. F., OvalJe, A. R. and Rezende, C. E. (1993). The biogeochemistry and trace metals distribution of mangrove rhizospheres. Biotropica, 25, 252 - 257.

Lacerda, L. D. (1994). Biogeochemistry of heavy metals in coastal lagoons. In: KJERFVE, B. (ed.), Coastal Lagoon Processes. Amster- dam, Netherlands. Elsevier, 221-241.

Lacerda, L. D. (1998). Biogeochemistry of Trace Metals and Diffuse Pollution in Mangrove Ecosystems. Okinawa International Society for Mangrove Ecosystems, 64.

Lakshmanaperumalasamy, P., Chandramohan, D. and Natarajen, R. (1986). Seasonal variation of microbial population from sediments of Vellar Estaury, South India. Gerban Ifremer, 3, 43- 54.

Laxen, D. P. H. (1983). The chemistry of Metal Pollution in water. In pollution causes, effects and control (ed), Roy, M. Horrison, The Royal Society of Chemistry, London, WIVOBN, 104.

Lin, J. G., Chen, S. Y. and Su, C. R. (2003). Assessment of sediment toxicity by metal speciation in different particle-size fractions of river sediment. Water Science and Technology, 47, 7–8, 233–241 © IWA Publishing.

Li, Q. S., Wu, Z. F., Chu, B., Zhang, N., Cai, S. S. and Fang, J. H. (2007). Heavy metals in coastal wetland sediments of the Pearl River Estuary, China. Environmental Pollution, 149, 158–164.

Liu, F. W., Yan, W., Hang, X. P. and Shi, P. (2003a). Distributional characteristics of heavy metal and its available phases in sediments from Zhujiang River mouth. Journal of Tropical Oceanography, 22 (5), 16–24 (in Chinese).

Li, W. C. and Wong, M. H. (2010). Effects of bacteria on metal bioavailability, speciation and mobility in different mine soils. Journal of soils and sediments, 10, 313-325.

Li, X. D., Shen. Z. G., Wai, O. W. H. and Li, Y. S. (2001). Chemical forms of Pb, Zn and Cu in the sediment profiles of the Pearl River Estuary. Marine Pollution Bulletin, 42, 215–223.

Li, X. D., Wai, O.W.H., Li, Y. S., Coles, B. J., Ramsey, M. H. and Thornton, I. (2000). Heavy metal distribution in sediment profiles of the Pearl river estuary, South China. Applied Geochemistry, 15, 567 – 581.

Li, Y. H. and Schoonmaker, J. E. (2005). Chemical composition and mineralogy of marine sediments. In: Mackenzie, F.T. (Ed.) Sediments, Diagenesis, and Sedimentary Rocks, vol. 7, First Edition: Treatise on Geochemistry, Holland, H.D., Turekian, K.K., Executive Directors, 1–35.

Loomb, C. A. M. (2001). Muddy Sedimentation in a Sheltered Estuarine Marine, Westpark Marina, Auckland, New Zealand. Thesis, The University of Waikato, 230.

Lopez- Sanchez, J. F., Rubio, R., Samitien, C. and Rauret, G. (1996). Trace metal portioning in marine sediments and sludges deposited off coast of Barcelona (Spain), Water Research, 30, 153 - 159.

Luoma, S. N. (1983). Bioavaibility of trace metals to aquatic organisms a review: The scene of total environment, 28, 1-22.

Mac Carthy, J. F. and Zachara, J. M. (1989). Sub surface transport of contaminants. Environment. Sci ence and Technology, 23, 496- 503.

Machado, W., Moscatelli, M., Rezende, L. G. and Lacerda, L. D. (2002). Mercury, zinc, copper accumulation in mangrove sediments surrounding a large landfill in southeast Brazil. Environmental Pollution, 120, 455– 461.

Macklin, M. G. and Dowsett, R. B. (1989). The chemical and physical speciation of trace metals in fine grained overbank flood sediments in the Tyne basin, north-east England. Catena, 16, 135–151.

Madiseh, S. D., Savary, A., Parham, H. and Sabzalizadeh, S. (2009). Determination of the level of contamination in Khuzestan coastal waters (Northern Persain Gulf) by using an ecological risk index. Environmental Monitoring and Assessment, 159, 521-530.

Mahalaxmi, M., Srinivasan, M., Murugan, M., Balkrishna, S. and Devanathan, S. (2011). Isolation and identification of total heterotropic bacteria and human pathogens in water and sediment from Cuddalore fishing harbour after Tsunami. Asian Journal of Biological Sciences, 1-9.

Maiz, I., Arambarri, I., Garcia, R. and Millan, E. (2000). Evaluation of heavy metal availability in polluted soils by two sequential extraction procedures using factor analysis. Environmental Pollution, 110, 3–9.

Mance, G. (1987). Pollution threat of heavy metals in Aquatic environments, Elsevier Applied Science Publishers Ltd, New York, United States of America.

Man, K. W., Jinshu, Z., Leung Amy, P. K., Paul Lam, K. S., Hon-Wah Lam, M. and Yu- Fong, Y. (2004). Distribution and behavior of trace metals in the sediment and porewater of a tropical coastal wetland. Science of the Total Environment, 327, 295–314.

Marchand, C., Allenbach, M. and Lallier- Verges, E. (2011). Relationships between heavy metal distribution and organic matter cycling in mangrove sediments (Conception Bay, New Caledonia). Geoderma, 164 (3-4), 444-456.

Marchand, C., Fernandez, J. M., Moreton, B., Landi, L., Verges, E. L. and Bultzer, F. (2012). The partioning of transistional metals (Fe, Mn, Ni and Cr) in mangrove sediments. Chemical Geology, 300, 70-80.

Marchand, C., Lallier-Verges, E., Baltzer, F., Alberic, P., Cossa, D. and Baillif, P. (2006). Heavy metals distribution in mangrove sediments along the mobile coastline of French Guiana. Marine Chemistry, 98, 1–17.

Marin, B., Valladon, M., Polve, M. and Monaco, A. (1997). Reproducibility testing of a sequential extraction scheme for the determination of trace metal speciation in a marine reference sediment by inductively coupled plasma-mass spectrometry. Analytica Chimica Acta, 342, 91–112.

Marscher, H. (1995). Mineral nutrition of higher plants. 2nd Edn, Academic Press, Inc.San Diego. 889.

Martincic, D., Kwokal, Z. and Branica, M. (1990). Distribution of Zn, Pb, Cd and Cu between different size fractions of sediments In the Limski Kanal (north Adriatic Sea). Science of the Total Environment, 95, 201–215.

Martino, M., Turner, A., Niemmo, M. and Millward, G. (2002). Resuspension, reactivity and recycling of trace metals in the Mersey Estuary, Marine Chemistry, 77, 171-186.

Massolo S., Bignasca A., Sarkar S. K., Chatterjee M., Bhattacharya, B. D. and Alam, A. (2012). Geochemical fractionation of trace metals in sediments of Hugli River (Gangas) and Sundarban Wetland (West Bengal, India). Environmental Monitoring Assessment, 184, 7561–7577.

Mendiguchai, C., Moreno, C., Manueal- Vez, M. P. and Garcia- Vargas, M. (2006). Preliminary investigation on the enrichment of heavy metals in marine sediments originated, from intensive aquaculture effluents. Aquaculture, 254, 317- 325.

Mikulic, N., Orescanin, V., Elez, L., Pavicic, L., Pezelj, D., Lovrencic, I. and Lulic, S. (2008). Distribution of trace elements in the coastal sea sediments of Maslinica Bay, Croatia. Environmental Geology, 53, 1413–1419.

Modak, D. P., Singh, K. P, Chandra, H. and Ray, P. K. (1992). Mobile and bound forms of trace metals in sediments of the lower Ganges. Water Research, 26 (11), 1541–1548.

Morillo, J., Usero, J. and Garcia, I. (2004). Heavy metals distribution in marine sediments from the southwest coast of Spain, Chemosphere, 55, 431-442.

Morillo, J., Usero, J. and Gracia, I. (2002). Partitioning of metals in sediments from the Odiel River (Spain). Environment International, 28, 263–271.

Morillo, J., Usero, J. and Rojas, R. (2008). Fractionation of metals and as in sediments from biosphere reserve (odiel saltmarshes) affected by acidic mine drainage. Environmental Monitoring and Assessment, 139, 329 - 337.

Morris, A. W., Bale, A. J. and Howland, R. J. M. (1982). Chemical variability in the Tamar Estuary, south-west England. Estuarine Coastal Shelf Science, 14, 649–661.

Muller, B. and Sigg, L. (1990). Interaction of trace metals with natural particle surfaces comparison between adsorption experiments and field measurements. Aquatic Science, 52, 75-92.

Muller, G. (1979). Schwermetalle in den sediment des Rheins, Veranderungem Seit 1971. Umschau, 79, 778-783.

Muzuka, A. N. N. and Shaghude, Y. W. (2000). Grain size distribution along the Msasani Beach North of Dares Salaam Harbor. Journal of African Earth Sciences, 30 (2), 417- 126.

Nagvenkar, G. S. and Ramaiah, N. (2010). Arsenite tolerance and biotransformation potential in estuarine bacteria. Ecotoxicology, 19, 604-613.

Nasrabadi, T., Nabi Bidhendi, G., Karbassi A. and Mehrdadi N. (2010). Evaluating the efficiency of sediment metal pollution indices in interpreting the pollution of Haraz River sediments, southern Caspian Sea basin. Environmental Monitoring and Assessment, 171, 395–410.

Nayak, G. N. and Bukhari, S. S. (1992). Spatial and temporal distribution of total suspended matter and other associated parameters in Zuari estuary, Goa. Journal of Indian Association of Sedimentologists, 11, 55-69.

Nedwell, D. B., Blackburn, T. H. and Wiebe, W. J. (1994). Dynamic nature of the turnover of organic carbon, nitrogen and sulphur in the sediments of a Jamaican mangrove forest. Marine Ecology Progress Series, 110, 223-231.

Neincheski, L. F. and Baumgarten, M. G. Z. (2000). Distribution of particulate trace metals in the southern part of Patos Lagoon estuary. Aquatic Ecosystem Health and Management, 3(4), 515-520.

Nelson, Y. M., Lion, L. W., Ghoirse, W. C. and Shuler, M. L. (1999). Production of biogenic Mn oxides by Leptothrix Discophora SS- 1 in a chemically defined growth medium and evaluation of their Pb adsorption characteristics. Applied Environment and Microbiology, 65 (1), 175-180.

Nolting, R., Helder, W., de Baar, H. and Gerringa, L. (1999). Contrasting behavior of trace metals in the Scheldt Estuary in 1978 compared to recent years. Journal of Sea Research, 42, 275 – 290.

Nolting, R., Sundby, B. and Duinker, J. (1990). Behaviour of minor and major elements in suspended matter in Rhine and Meuse Rivers and Estuary. Science of Total Environment, 97, 169 – 183.

Ogbanna, D. N. (2010). Seasonal dynamics of microbial population and physicochemical characteristics of water body receiving industrial pollutants in port Harcoutt, Nigeria. Agricultural and Biology journal of North America, 1 (6), 1333- 1339.

Olutuna, O. G., Aribisala, G. O. and Akintunde, E. A. (2012). A study of chemical speciation of metals in aquatic bottom sediment of Aiba reservoir, Iwo, Nigeria. African Journal of Environmental Science and Technology, Vol. 6(8), 312-321.

Omiema, D. S. and Ideriah, T. J. K. (2012). Distribution of microorganisms in water and sediment along Abonnema shoreline, Eastern Niger Delta, Nigeria. Journal of Chemical, Biological and physical Sciences, 2(4), 2114 – 2122.

Ongeri, D. M. K., Lalah J. O. and Wandiga, S. O. (2010). Speciation of Cd, Cu, Zn, Fe and Pb in sediments sampled from Lake Victoria basin, Kenya. Environmentalist, 30, 254–259.

Orson, R. A., Simpson, R. L. and Good, R. E. (1992). A mechanism for the accumulation and retention of heavy metals in tidal fresh water marshes of the upper Delaware River estuary. Estuarine Coastal and Shelf Science, 34, 171-186.

Owens, R. E., Balls, P. W. and Price, N. B. (1997). Physicochemical processes and their effects on the composition of suspended particulate material in estuaries: implications for monitoring and modelling. Marine Pollution Bullettin, 34, 51–60.

Palanques, A., Plana, F. and Maldonado, A. (1995). Recent influences of man on Ebro margin sedimentation system (Norwestern Mediterranean Sea). Marine Geology, 95, 247-263.

Paludan, C. and Morris, J. F. (1999). Distribution and speciation of Phosphorous along a salinity gradient in intertidal marsh sediments. Biogeochemistry, 45, 197-221.

Panquin, P. R., Gorsuch, J. W., Apte, S., Batley, G. E., Bowles, K. C., Campbell, P. G. C., Delos, C. G, Di Toro, D. M., Dwyer, R. L., Galvez, F., Gensemer, R. W., Goss, G. G., Parameshwari, E., Lakshamanan, A. and Thilagavatti, T. (2009). Chromate resistance and reduction by bacterial isolates. Australian journal of Basic and Applied Sciences, 3(2), 1363- 1368.

Pardo, R., Barrado, E., Perez, L. and Vega, M. (1990). Determination and association of heavy metals in sediments of the Pisucraga, river. Water Research, 24(3), 373-379.

Patrick, W. H., Gambrell, R. P. and Khalid, R. A. (1977). Physiochemical factors regulating solubility and bioavailability of toxic heavy metals in contaminates dredge sediment. Journal of Environmental Science and Health, A12, 475 – 492.

Pedersen, T. F. and Price, N. B. (1982). The geochemistry of manganese carbonate in Panama Basin sediments. Geochimica Cosmochimica Acta, 46, 59–68.

Pereira, F., Kerkar, S. and Krishan, K. P. (2012). Bacterial response to dynamic metal concentration in the surface sediments of a solar saltern (Goa, India). Environmental Monitoring and Assessment, 6 (2), 217- 230.

Perin, G., Craboledda, L., Lucchese, M., Cirillo, R., Dotta, L., Zanetta, M. L. and Oro, A. A. (1985). Heavy metal speciation in the sediments of northern Adriatic sea. A new approach for environmental toxicity determination. In: Lakkas TD (Ed.). Heavy Metals in the Environment, CEP Consultants, Edinburgh. Environmental Pollution, 110, 3–9.

Pertsemli, E. and Voutsa, D. (2007). Distribution of heavy metals in Lakes Doirani and Kerkini, Northern Greece. Journal of Hazardous Materials, 148(3), 529-537.

Petersen, W., Wallmann, K., Li, P. L. and Schroeder, F. (1993). A review of factors effecting the release and bioavailability of metals during sediment disturbance events. Environmental International, 973- 980.

Petersen, W., Wallmann, K., Li, P. L., Schroeder, F. and Knauth, H. D. (1995). Exchange of trace elements at the sediment–water interface during early diagenesis processes. Marine and Freshwater Research, 46, 19–26.

Pizarro, I., Gomez, M., Camara, C., and Palacios, M. A. (2003). Arsenic speciation in environmental and biological samples. Analytica Chimica Acta, 495, 85– 98.

Pokrovsky, O. S., Shirokova, L. S., Zabelina, S. A., Vorobieva, O., Yu Moreya, Klimoy, S. I., Chupakov, A. V., Shorina, N. V., Kokryatskaya, N. M., Audry, S., Viers, J., Zoutien, C. and Freydier, R. (2012). Size fractionation of trace elements in a seasonally Stratified Boreal lake: Control of organic matter and Iron colloids. Aquatic Geochemistry, 18, 115–139.

Poppe, L. J. and Commeau, J. A. (1992). Mineralogy of silt fractions in surficial sediments from united states mid-Atlantic shelf, slope and rise. Marine Geology, 103, 185-197.

Postma, H. (1967). Sediment transport in the estuarine environment. In: Estuaries, ed. G. H. Lauff. American Association for Advancement of Science Washington, D.C. Publication 83, 158–179.

Praveena, S. M., Aris, A. Z. and Radojevic, M. (2010). Heavy metal dynamics and soutce in intertidal mangrove sediment of Saban Borneo Island. Environment Asia, 37, 79- 83.

Praveena, S. M., Radojevic, M. and Abdullah, M. H. (2007). The assessment of Mangrove sediment quality in Menkabong Lagoon: An Index Analysis Approach. International Journal of Environmental and Science Education, 2(3), 60-68.

Qasim, S. Z. and Sen Gupta, R. (1981). Environmental characteristics of the Mandovi – Zuari estuarine system in Goa. Estuarine Coastal and Shelf Sciences, 13, 557-578.

Qiao, S., Yang, Z., Pan, Y. and Guo, Z. (2007). Metals in suspended sediments from the Changjiang (Yangtze River) and Huanghe (Yellow River) to the sea, and their comparision. Estuarine Coastal and Shelf Science, 74, 539-548.

Qi, S., Leipe, T., Rueckert, P., Di, Zhou. and Harff, J. (2010). Geochemical sources, deposition and enrichment of heavy metals in short sediment cores from the Pearl River Estuary, Southern China. Journal of Marine Systems, 82, S28- S42.

Raghunath, R. and Sreedhara Murthy, T. R. (1996). Carbonate and organic matter studies of the shelf of Kasargod, west coast of India. Indian Journal Marine Science, 25, 355 – 357.

Rajendran, N. and Karthiksen, K. (2007). Microbial flora associated with submerged mangrove leaf litter in India. Reviews Biosphere Tropical, 55 (2), 393-400.

Ramkumar, S., Kannapuram, E. and Palanisamy, M. (2012). Prevelance and distribution of total heterotropic bacteria from Kottaipattinam coast, Palk Strait, South esat coast of India. Archives of Applied Science Research, 3 (5), 593 – 598.

Ramos, C. A., da Silva, A. P. and de Oliveira, S. R. (2006). Concentration, stock and transport rate of heavy metals in a tropical red mangrove, Natal, Brazil. Marine Chemistry, 99, 2–11.

Ramos, L., Hernandez, L. M. and Gonzalez, M. J. (1994). Sequential fractionation of copper, lead, cadmium and zinc in the soil from or near Donana National Park. Journal of Environmental Quality, 23, 50–57.

Ratasuk, P., Parkpian, P., Jugsujinda, A. and DeLaune, R. D. (2003). Factors governing adsorption and distribution of copper in Samut Prakarn coastal sediment, Thailand. Journal Environmental Science and Health, 38, 1793–1810.

Ratheesh Kumar, C. S., Joseph, M. M., Gireesh Kumar, T. R., Renjith, K. R., Manjir, M. N. and Chandramohanakumar, N. (2010). Spatial variability and contamination of heavy metals in the intertidal systems of tropical environment. International Journal of Environmental Research, 14 (4), 691-700.

Rees, J. G., Williams, T. M., Nguli, M. M., Kairu, K. K. and Yobe, A. C. (1996). Contaminant transport and storage in the estuarine creek systems of Mombasa, Kenya. British Geological Survey Technical Report, WC/96/42, 67.

Regnier, P. and Wollast, R. (1993). Distribution of trace metals in suspended matter of the Scheld Estuary. Marine Chemistry, 43, 3-19.

Reimann, L. G., Steward, F. and Azam, F. (2000). Dynamics of bacterial community composition and activity during a mesocosm diatom bloom. Applied Environmental Microbiology, 66, 578- 587.

Riba, I., Blasco, J., Jimenez-Tenorio, N., Gonzalez, M. L. and Angel Delvalls, T. (2005). Heavy metal bioavailability and effects: II. Histopathology-bioaccumulation relationships caused by mining activities in the Gulf of Cadiz (SW, Spain). 58 (5), 671-682.

Riba, I., DelValls, T. A., Forja, J. M. and Gomez- Parra, A. (2004). The influence of pH and salinity values in the toxicity of heavy metals in sediments to the estuarine clam Ruditapes Philippinarum. Environmental Toxicology and Chemistry, 23 (5), 1100 -1107.

Richards, K. J. (1982). Modeling the benthic boundary layer. Journal of Physical Oceanography, 12, 428- 439.

Rini, S. (2002). Some biogenic compounds and their derivatives in selected Mangrove Ecosystems. Ph.D thesis, Cochin University of Science and Technology, 150.

Rosales- Hoz L, Cundy, A. B. and Bahena – Manjarrez, J. L. (2003). Heavy metals in sediment cores from a tropical estuary affected by anthropogenic discharges: Coatzacoalcos Estuary, Mexico. Estuarine Coastal and Shelf Science, 58, 117–126.

Rubio, B., Nombela, M. A. and Vilas, F. (2000). Geochemistry of major and trace elements in sediments of the Ria de Vigo (NW Spain): an assessment of metal pollution. Marine Pollution Bulletin, 40, 968–980.

Rubio, R., Lopez-Sanchez, J. F. and Rauret, G. (1991). La especiaciỏn sỏlida de trazas de metales en sedimentos. Aplicaciỏn a sedimentos muy contaminados. Analytical Chemistry, 87, 599-605.

Ryan, P. R., Germaine, K., Franks, A., Ryan, D. J. and Dowling, D. V. (2005). Bacterial endophytes: Recent developments and applications, 278 (1), 1-9.

Sahu, K. C. and Bhosale, U. (1991) Heavy metal pollution around the island city of Bombay, India. Part I: quantification of heavy metal pollution of aquatic sediments and recognition of environmental discriminants. Chemical Geology, 91, 263- 283.

Salomons, W. and Forstner, U. (1984). Metals in the hydrocycle. Berlin: Springer-Verlag.

Sandhuram, Y., Sarma, V. V., Murthy, T. V. R. and Rao, P. B. (2005). Seasonal variability of physico- chemical characteristics of the Haldia channel of Hooghly Estuary. India, J. Earth System Science, 114 (1), 37 – 49.

Santhanam, B. (1976). Distribution of bacteria in estuarine waters of the Cochin. Estuaries, 2, 34-48.

Santos Echeandia, J., Vale, C., Cretano, M., Pereira, P. and Prego, R. (2010). Effect of tidal flooding on metal distribution in pore waters of marsh sediments and its transport to water column (Tagus Estuary, Portugal), Marine Environmental Research, 70, 358-367.

Santschi, P. H., Hohener, P., Benoit, G. and Bucholtz Ten Brink, M. (1990). Chemical processes at the sediment- water interface. Marine Chemistry, 30, 269-315.

Sathyanarayanan, D., Prasada Reddy, B. R., Swamy, A. S. R. and Krishna Rao G. (1985). Chemical ocenanography of harbor & coastal environment of Visakhapattinam (Bay of Bengal) part ii; Nature and composition of sediments. Indian Journal of Marine sciences, 14, 147-150.

Schorer, M. (1997). Pollutant and organic matter content in sediment particle size fractions. Freshwater Contamination, proceedings of Rabit Symposium S4, IAHS publication, 243.

Schubauer, M. K., Dierkes, J. R., Monson, P. D. and Ankley, G. T. (1993). pH-dependent toxicity of Cd, Cu, Ni, Pb and Zn to Ceriodaphnia dubia, Pimephales promelas, Hyalella azteca and Lumbriculus variegatus. Environmental Toxicology and Chemistry. 12, 1261-1266.

Scully, M. E. and Friedrichs, C. T. (2007). Sediment pumping by tidal asymmetry in a partially mixed estuary. Journal of Geophysical Research, 112.

Selvaraj, K., Mohan, V. R. and Szefer, P. (2004). Evaluation of metal contamination in coastal sediments of the Bay of Bengal, India: Geochemical and statistical approaches. Marine Pollution Bulletin, 49, 174–185.

Severmann, S., Lyons, T. W., Anbar, A., McManus, J. and Gordon, G. (2008). Modern iron isotope perspective on the benthic iron shuttle and the redox evolution of ancient oceans. Geology, 36, 487–490.

Shan, X. Q. and Chen, B. (1993). Evaluation of sequential extraction for speciation of trace metals in model soil containing natural minerals and humic acid. Analytical Chemistry, 65, 802–807.

Shaw, T. J., Goskes, J. M. and Jahnke, R. J. (1990). Early diagenesis in differing depositional environment. The response of transition metals in a pore water, Geochimica et Cosmochimica Acta, 54, 1233- 1246.

Shetye, S. R., Kumar, M. D. and Shankar, D. (2007). The Mandovi and Zuari estuaries. National Institute of Oceanography, Goa, India. 145.

Shetye, S. R., Michael, G. S. and Pradnya, V. C. (2007b). Mixing and intusion of salt water. in: Shetye, S.R., Kumar, M.D., Shankar, D. (ed) The Mandovi and Zuari Estuaries. National Institute of Oceanography, Dona Paula, Goa, 49-58.

Shynu, R., Rao, P. V., Kessarkar, P. M. and Rao, T. G. (2012). Temporal and Spatial variability of trace metals in suspended matter of Mandovi estuary, central west coast of India. Environmental Earth Sciences, 65, 725- 739.

Silver, S. and Phung, L. T. (2009). Heavy metals, bacterial resistance. In Encyclopedia of Microbiology, pp. 220–227. Edited by M. Schaechter. Oxford: Elsevier.

Singh, K. P., Mohan, D., Singh, V. K. and Malik, A. (2005). Studies on distribution and fractionation of heavy metals in Gomti river sediments–A tributary of the Ganges, India. Journal of Hydrology, 312, 14−27.

Siraswar, R. and Nayak, G. N. (2011). Mudflats in lower middle estuary as favourable location for concentration of metals, west coast of India. Indian Journal of Geo-marine Sciences, 40 (3), 372 – 385.

Soto-Jimenaz, M. F. and Paez-Osuna, F. (2001). Distribution and Normalisation of heavy metal concentrations in mangroves and lagoonal sediments from the Mazatlan Harbor (SE Gulf of California). Estuarine Coastal and Shelf Science, 53(3), 259- 274.

Spencer, K. L. and Macleod, C. L. (2002). Distribution and Partitioning of Heavy Metals in Estuarine Sediment Cores and Implications for the Use of Sediment Quality Standards. Hydrology and Earth System Sciences, 6, 989-998.

Srivastava, J., Farooqui, A. and Hussain, S. M. (2012). Sedimentalogy and salinity status of Pichavaram mangrove wetland, south east coast of India. International Journal of Geology, Earth and Environmental Sciences, ISSN: 2277-2081 (Online), 2 (1), 7-15.

StatSoft (1999). Statistica computer program, version 5.5. StatSoft, Tulsa, OK.

Stecko, J. R. P. and Bendell- Young, L. I. (2000). Contrasting the geochemistry of suspended particulate matter and deposited sediments within an estuary. Applied Geochemistry, 15 (6), 753- 775.

Subramanian, V. and Mohanachandran, G. (1990). Heavy metals distribution and enrichment in the sediments of Southern East Coast of India. Marine Pollution Bulletin, **2,** 324–330.

Summers, R. W. (1980). The diet and feeding behaviour of the flounder Platichthys flesus (L.) in the Ythan Estuary, Aberdeenshire, Scotland. Estuarine, Coastal and Shelf Science, 2, 217–232.

Sundaray, K. K. (2007). Water quality assessment of Mahanadi River, Orissa, India using multivariate statistical approach. Doctoral thesis, Utkal University, Bhubaneshwar.

Sundaray, S. K., Nayak, B. B., Lin, S. and Bhatta, D. (2011). Geochemical speciation and risk assessment of heavy metals in the river estuarine sediments- a case study, Mahanadi Basin India. Journal of Hazardous material, 186, 1837-1846.

Sundby, B., Anderson, L. G., Hall, P. O. J., Iverfeldt, A., Rutgers van der Loeff, M. M. and Westerlund, S. F. G. (1986). The effect of oxygen release on uptake of cobalt, manganese, iron and phosphate at the sediment-water interface. Geochem et Cosmochimica Acta, 50, 1281–1288.

Sung, W. and Morgan, J. J. (1980). Kinetics and product of ferrous iron oxygenation in aqueous systems. Environmental Science Technology, 14, 561–568.

Surija, B. and Branica, M. (1995). Distribution of Cd, Pb, Cu and Zn in carbonate sediments from the Krka river estuary obtained by sequencial extraction. The science of Total Environment, 170, 101- 118.

Swallow, K. C., Westall, J. C., McKnight D. M., Morel N. M. L. and Morel, F. M. M. (1980). Potentiometric determination of copper complexation by phytoplankton exudates. Limnology and Oceanography, 23, 538–542.

Tarn, N. F. Y. and Wong, Y. S. (2000). Spatial variation of heavy metals in surface sediments of Hong Kong Mangrove swamps. Environmental Pollution, 110, 195 – 205.

Tavakoly Sany, B., Sulaiman, A. H., Monazami, G. H. and Salleh, A. (2011). Assessment of Sediment Quality According To Heavy Metal Status in the West Port of Malaysia. World Academy of Science, Engineering and Technology, 50.

Taylor, D. S. (2000). Biology and Ecology of Rivulus Marmoratus: New insights and a review: Florida Scientist, 63 (4), 242 - 255.

Teasdale, P. R., Apte, S. C., Ford, P. W., Batley, G. E. and Koehnken, L. (2003). Geochemical cycling and speciation of Copper in waters and sediments of Macquaire Harbour, Western Tasmania, Estuarine, Coastal and Shelf Science, 57, 475-487.

Templeton, D. M., Ariese, F., Cornelis, R., Danielsson, L., Muntau, H., Van Leeuwen, H. P. and Lobinski, R. (2000). Guidelines for terms related to chemical speciation and fractionation of elements. Definations, structural aspects, and methodological approaches. Pure and Applied Chemistry, 72 (8), 1453-1470.

Tessier, A. and Campbell, P. G. C. (1987). Partitioning of trace metals quality classification. Environ. Sci. Policy 2:75–86. In sediments, relationships with bioavailability. Hydrobiologia, 149.

Tessier, A., Campbell, P. G. C. and Bisson, M. (1979). Sequential extraction procedure for the speciation of particulate trace metals. Analytical Chemistry, 51 (7), 844 – 851.

Thamdrup, B., Fossing, H. and Jorgensen, B. B. (1994). Manganese, iron and sulphur cycling in coastal marine sediment, Aarhus Bay, Denmark. Geochimica Cosmochimica Acta, 58, 5115 – 5129.

Thill, A., Moustier, S., Garnier, J. M., Estournel, C., Naudin, J. and Bottero, J. (2001). Evolution of particle size and concentration in Rhone river mixing zone: influence of salt flocculation. Continental Shelf Research, 21, 2127 – 2140.

Tijani, M. N. and Onodera, S. (2009). Hydrogeochemical assessment of metal contamination in an Urban Drainage System: A case Study of Osogbo Town ship, SW Nigeria. Journal of Water Resource and Protection, 3, 164- 173.

Tokalioglu, S., Kartal, S. and Elci, L. (2002). Determination of heavy metals and their speciation in lake sediments by flame atomic absorption spectrometry after a four-stage sequential extraction procedure. Analytica Chimica Acta, 413, 33–40.

Tokalioglu, S., Kartal, S. and Elci, L. (2000). Speciation and determination of heavy metals in lake waters by Atomic Absorption Spectrometry after sorption on Amberlite XAD- 16. Analytical Sciences, 16, 1169-1173.

Tomlinson, D. C., Wilson, J, G., Harris, C. R. and Jeffery, D. W. (1980). Problems in the assessment of heavy metals levels in estuaries and the formation of a pollution index, Helgol. Wiss. Meeresunters, 33 (1-4), 566-575.

Turekian, K. K. and Wedepohl, K. H. (1961). Distribution of the elements in some major units of the Earth's crust. Geological society of American Bulletin, 72 (2), 175-192.

Turner, A. and Millward, G. (2002). Suspended particles: Their role in estuarine biogeochemical cycles. Estuarine Coastal and Shelf Sciences, 55, 857 – 883.

Turner, A. (1999). Diagnosis of chemical reactivity and pollution sources from particulate trace metal disturbance in estuaries, Estuaries Coastal and Shelf Science, 48, 177- 191.

Turner, A. (2000). Trace metal contamination in sediments from U.K. estuaries: An empirical evaluation of the role of hydrous iron and manganese oxides. Estuarine, Coastal and Shelf Science, 50, 355–371.

Ukpong, I. E. (1997). Mangrove swamp at a saline/ fresh water interface near creek Town, Southeastern Nigeria. Catena, 29 (1), 61- 71.

Valdés, J., Vargas, G., Sifeddine, A., Ortlieb, L. and Guinez, M. (2005). Distribution and enrichment evaluation of heavy metals in Mejillones Bay (23°S), Northern Chile: Geochemical and statistical approach. Marine Pollution Bulletin, 50, 1558–1568.

Van Leussen, W. (1994). Estuarine macroflocs and their role in fine grained sediment transport Disseration, University of Utrecht Utrecht, 488.

Van Nugteren, P., Herman, P. M. J., Moodley, L. and Middelburg, J. J. (2009). Spatial distribution of detrital resources determines the outcome of the competition between bacteria and facultative detrivorous worm. Limnology and Oceanography, 54, 1413-1419.

Verlaan, P. A., Donze, M. and Kuik, P. (1997). Marine vs. Fluvial suspended matter in the Scheldt estuary. Estuarine Coastal Shelf Science, 46, 873–883.

Vijver, M., Jages, T., Posthuman, L. and Peijnenburg, W. (2003). Metal uptake from soils and soil- sediment mixture by larvae of Jenebrio molitor (Coleoptera). Ecotoxicology and Environmental Safety, 54, 277-289.

Violante, A., Huang, P. M. and Gadd, G. M. (editors) (2008). Biophysicochemical Processes of Heavy Metals and Metalloids in Soil Environments. Chichester: Wiley.

Volvoikar, S. P. and Nayak, G. N. (2013). Depositional environments and geochemical response of mangrove sediments from creeks of northern Maharashtra coast, India. Marine pollution Bulletin, 69 (1-2), 223-227.

Wakeham, S. (2002). Diagenesis of organic matter at water- sediment interface. In: A. Gianguzza, E. Pelizzeti and S. Sammartano (eds). Chemistry of Marine water and sediments. Springer, Berlin, 146-164.

Wang, L., Ruilian, Yu., Gongren, Hu. and Xianglin, Tu. (2010). Speciation and assessment of heavy metals in surface sediments of Jinjiang River tidal reach, Southeast of China, Environmental Monitoring and Assessment, 165, 491- 499.

Warren, L. A. and Zimmerman, A. P. (1994). Suspended particulate oxides and organic matter interactions in trace metal sorption reactions in a small urban river. Biogeochemistry, 23, 21–34.

Wilde, K. L., Stauber, J. L., Markich, S. J., Franklin, N. M. and Brown, P. L. (2006). The effect of pH on the uptake and toxicity of Cu and Zn in a tropical freshwater alga (Chlorella sps.). Archives of Environmental Contamination and Toxicology, 51, 174- 185.

Wolanski, E., Mazda, Y. and Ridd, P. (1992). Mangrove hydrodynamics. In: Robertson, A.I., Alongi, D.M. (Eds.), Coastal and Estuarine Studies. American Geophysical Union, 329.

Wolters, E. C., Huang, C. C. and Clark, C. (1989). Positron emission tomography in manganese intoxication. Annual Neurology, 26, 647-651.

Yuan, C., Shi, J., He, B., Liu, J., Liang, L. and Jiang, G. (2004). Speciation of heavy metals in marine sediments from the East China Sea by ICP-MS with sequential extraction. Environment International, 30, 769–783.

Yu, K. Y., Tasi, L. J., Chen, S. H. and Ho, S. T. (2006). Chemical binding of heavy metals in anoxic river sediments. Water Research, 35 (7), 4086 – 4094.

Zhang, C., Wang, L., Li, G., Dong, S., Yang, J. and Wang, X. (2002). Grain size effect on multi-element concentrations in sediments from the intertidal flats of Bohai Bay, China. Applied Geochemistry, 17, 59 – 68.

Zhang, J., Huang, W. W. and Martin, J. M. (1988). Trace metals distribution in Huanghe (Yellow River) estuarine sediments. Estuarine, Coastal and Shelf Science, 26, 499–526.

Zhaung, C., Wang, L., Dong, S., Yang, J. and Wang, X. (2002). Grain size effect on multi-elment concentration in sediments from the intertidal flats of Bohai Bay, China. Applied Geochemistry, 17, 59 – 68.

Zhou, H., Peng, X. and Pan, J. (1998). Distribution, source and enrichment of some chemical elements in sediments of Pearl River estuary, China. Continental shelf research, 24, 1857 – 1875.
Zhou, J. L., Liu, Y. P. and Abrahams, P. W. (2003). Trace metal behavior in the Conwy estuary, North Wales. Chemosphere, 51, 429-440.

Zoumis, T., Schmidt, A., Grigorova, L. and Calmano, W. (2001). Contaminants in sediments: Remobilisation and demobilization. Science of Total Environment, 266, 195 – 202.

Zwolsman, J. J. G. and van Eck, G. T. M. (1999). Geochemistry of major elements and trace metals in suspended matter of the Scheldt estuary South west Netherlands. Marine Chemistry, 66, 91-111.

Zwolsman, J. J. G., Berger, G. W. and Van Eck, G. T. M. (1993). Sediment accumulation rates, historical input, post-depositional mobility and retention of major elements and trace elements in salt marsh sediments of the Scheldt estuary, SW Netherlands. Marine Chemistry, 44, 73–94.