METAL FRACTIONATION IN SEDIMENTS: A COMPARATIVE ASSESSMENT OF FOUR SEQUENTIAL EXTRACTION SCHEMES

Zakir, H. M.^{1,2} and Naotatsu SHIKAZONO³

¹ Ph. D. Student, Laboratory of Geochemistry, Dept. of Applied Chemistry, Keio University
(3-14-1, Hiyoshi, Kohoku-ku, Yokohama 223-8522, Japan)

E-mail: zakirhm.ac.bau@gmail.com

² Assistant Professor, Dept. of Agricultural Chemistry, Bangladesh Agricultural University
(Mymensingh 2202, Bangladesh)

³ Professor, Laboratory of Geochemistry, Dept. of Applied Chemistry, Keio University
(3-14-1, Hiyoshi, Kohoku-ku, Yokohama 223-8522, Japan)

E-mail: sikazono@applc.keio.ac.jp

Four sequential chemical extraction schemes for metal analysis (a 4-step modified protocol originally proposed by the Standards, Measurements and Testing Programme (SM&T - formerly BCR) of the European Union, Tessier and Hall 5-step procedures and 6-step Grimalt's scheme) were compared. Each procedure was applied to four sediment samples collected from two rivers, Nakagawa (NR) and Tamagawa (TR), Tokyo, Japan. Additionally, a certified marine sediment reference material (JMS-1) was also extracted for the comparison. To evaluate the partitioning of metals among different geochemical forms, the concentration of chromium (Cr), copper (Cu), nickel (Ni), lead (Pb) and zinc (Zn) were measured by Zeeman-corrected atomic absorption spectrometry. Other major elements and mineralogy of sediments were also determined on bulk prior to extracting by X-ray fluorescence (XRF) and X-ray diffraction (XRD) analysis, respectively, which allowed qualitative correlation between the fractionation results obtained and the presence of defined geochemical phases. The total concentrations of metals were determined after strong acid attack and the analytical precision was verified by using JMS-1. Significant proportions of elements were found in non-residual fractions among the schemes (average 83, 91, 76, 93 and 59% for Cr, Cu, Pb, Zn and Ni, respectively) in the more anthropogenically impacted NR sediments. The highest mobility was observed for Pb and Zn among the four schemes while the lowest was for Cr and Ni. Results of the comparison among the total metal levels for the four schemes based upon the sum of sequential extraction steps for each element showed significant differences for Cr, Ni and Zn. On the other hand, the sum of non-residual phases of the four schemes showed no significant differences for Cr, Cu and Zn but the SM&T and Hall schemes showed significant differences from others for Pb and Ni, respectively. Generally, good recoveries were achieved with the Hall scheme (range 92-109%) and the Fe-Mn oxides occluded phase of this scheme was the most effective owing to the high tendency to extract metals. In addition, Grimalt scheme produced the lowest recovery of Cr and the concentrations of residual fraction of other elements were also lower.

Key Words : Sequential extraction, Fractionation, Metal distribution, Sediment

1. INTRODUCTION

Metals like Cr, Cu, Ni, Pb, Zn and others are most important among the frequently observed contaminants in soil and sediments. Both industrial activities and urbanization have greatly increased the

metal burden in the environment. Deposition of metal-rich wastes through different anthropogenic activities (such as mine tailings, metal smelting, leather tanning, electroplating, emissions from gas exhausts, energy and fuel production, intensive agriculture and sludge dumping and others)

contaminate environmental systems with non-biodegradable toxic metals¹⁾. The list of sites contaminated with metals grows every year, presenting a serious problem for human health and a formidable danger to the environment²⁾. The west bank of NR at Kazeno Hiroba (Higashi-Ohjima of Edogawa Ward, Tokyo, Japan) is one such location where a significant amount of industrial metal contained waste was deposited, particularly Cr. Data obtained by speciation and knowledge concerning to the distribution of metals in soils and sediments can be used as useful tools to extrapolate the degree to which metals and their compounds are mobile, extractable and plant-available^{3, 4, 5)}. Sequential extraction schemes are recognized to be a necessary tool to obtain such information as total concentration does not provide suitable information about the bioavailability of potentially toxic elements. Four to eight extractants are usually employed in an order in which the earlier ones are the least aggressive and the most specific, and subsequent extractants are progressively more destructive and less specific. Regardless of the schemes used there have been a number of problems identified such as reagents that are insufficiently specific to dissolve target phases and there is potential for substantial redistribution of metals among the phases during extracting processes. Thus, there is no reagent with a single phase specific to the removal of metals⁶⁾. It is generally assumed that the availability of the metals to the living organisms follows a decreasing order from the first step towards the last step in a sequential extraction scheme. The extent of overlap occurring between the various fractions is a subject of discussion. However, it is probable that up to the oxide occluded forms in different schemes of sequential extractions represent the forms of the metals that are more available than the metals in the last fractions. Therefore, the changes in the proportion of metals bound in these fractions can be used as an indication of possible important environmental changes^{7, 8)}.

There are a wide variety of sequential extraction procedures available in literature based on a different sequence of extractants and/or different operating conditions^{9, 10, 11, 12, 13)}. Among those, the 4-step SM&T (formerly BCR scheme, modified by Rauret et al.¹⁴⁾) and the 5-step Tessier¹⁵⁾ schemes are most widely accepted. A similar method to the Tessier scheme is described by Grimalt¹⁶⁾ and includes a water soluble phase (deionized H₂O). In the Hall scheme¹⁷⁾, the oxide/ reducible phase is divided into metals associated with amorphous Fe oxyhydroxide and crystalline oxides of Fe. Both the SM&T and Hall schemes combine the exchangeable and

carbonate phases into one fraction.

The aim of this study was to compare the results from four sequential extraction procedures used for partitioning metals in two different types of sediments. In comparison, emphasis was given to the non-residual phases of metal as these are widely used to determine the anthropogenic pollution levels in solid materials. The methods were chosen because of their similarities and capacity to generate specific information on the type of materials considered. The study was focused on the five elements of toxicological interest (Cr, Cu, Pb, Ni and Zn), that may be associated with anthropogenic activities. The methods have been applied to four sediment samples collected from the two rivers (NR and TR) near Tokyo, Japan and JMS-1, a certified marine sediment¹⁸⁾ for which no information regarding phase dependent composition is available although the Cr content of JMS-1 has been analyzed recently¹⁹⁾ by using X-ray near edge structure.

2. EXPERIMENTAL MATERIALS AND METHODS

(1) Reagents and instrumentation

All chemicals and reagents were of analytical reagent grade quality (Sigma-Aldrich, USA and Wako, Japan). Millipore water was used throughout all the experiments. Before use, all glass and plastic ware were soaked in 14% HNO₃ for 24 hrs. The washing was completed with Millipore water rinse.

Atomic absorption measurements were performed with a Hitachi Z-6100 Polarized Zeeman spectrophotometer, equipped with single element hollow-cathode lamps. The instrument was operated at maximum sensitivity with an air-acetylene flame. Lamp current and slit width were used according to the manufacturer's recommendation. The instrumental parameters used for each element are shown in Table 1. A centrifuge (Kokusan H-27F, Tokyo, Japan) for complete separation of the extract from the residue, a Yamato SA-320 (Tokyo, Japan) shaker and a Toa-Dkk HM-20P (Tokyo, Japan) pH meter were used throughout all the experiments.

A preliminary quantitative determination of major elements of the samples was carried out by X-ray Fluorescence spectroscopy, employing a Rigaku RIX 1000 (Tokyo, Japan) XRF. For the preparation of beads, 0.4000 ± 0.0005 g dry oxidized (at 900°C for 14 hrs) samples, lithium tetraborate (4.0 g) and lithium iodide (50 mg) were mixed

Table 1 Instrumental and experimental parameters for the metals

Parameters	Metals				
	Cr	Cu	Ni	Pb	Zn
Electric current (mA)	7.5	7.5	10.0	7.5	5.0
Wavelength (nm)	359.3	324.8	232.0	283.3	213.9
Slit width (nm)	1.3	1.3	0.2	1.3	1.3
Analytical working range ($\mu\text{g ml}^{-1}$)	0.5-4.0	0.5-8.0	0.5-4.0	1.0-8.0	0.1-1.0
Determination limit ($\mu\text{g ml}^{-1}$)	100	30	20	200	4

together and used a Bead Sampler NT-2100 (Tokyo, Japan). The X-ray tube features a 20-60 kV/ 80 mA power supply, chromium transmission target and a beryllium side window. Analyzing crystals are in four positions consist with lithium fluoride (LiF [200]), penta-erythritol (PET [002]) and germanium (Ge [111]). Gas flow proportional counter (PC) and source slit diameter are same (0.45 mm) while the scintillation counter (SC) diameter is 0.15 mm. Scintillation counter consist of a scintillating crystal of NaI doped with thallium attached to a photomultiplier (R580 HTV). Plate calibration was performed using geological standards (JB-3, JF-1, JG-2, JGb-2, JH-1 and JSy-1) following the manufacturer's recommendations. The results are calculated considering the ignition loss and expressed in mass percent. A Bruker AXS: D-8 Advance (Berlin, Germany) X-ray diffractometer was employed for XRD analysis.

(2) Collection and preparation of samples

Bed sediment samples were collected from Nakagawa River (located in the eastern part of Tokyo, Japan) and Tamagawa River (located in the western part of Tokyo, Japan). Between them, NR is shorter in length, depth and width than TR and receives significant amount of industrial discharges, municipal wastewater and others. And NR possesses almost muddy sediments without coarse sand while TR has typical sediments with coarse sand. The samples from NR were collected at Kazeno Hiroba (Higashi-Ohjima of Edogawa Ward, Tokyo) and from TR at the Taishi Bridge (Taishigawara, Kawasaki). Kazeno Hiroba residents indicated industrial wastes containing metals including Cr were deposited about 30 years ago. Two samples were collected from each river, and sampling sites were separated by a distance of about 300 m. The sample mass collected in each case was about 500 g. Sub-samples of the material were oven dried at 50°C for 24 hrs and sieved (aperture 125 μm). The lower

particle size fraction was homogenized by grinding in an agate mortar and stored in glass bottles until the sequential extraction analyses were carried out.

(3) Procedures

The sequential extraction procedures applied are shown in Table 2. All the operations were carried out in 50 mL polypropylene centrifuge tubes (Nalgene, New York) and Teflon (PTFE) containers provided with screw stoppers. The initial sediment mass was 1.0000 ± 0.0005 g. The extracts were separated from the solid residue by centrifugation at 4000 rpm for 30 min. The residue was washed with 10 mL millipore water in between the successive steps and the supernatant washings added to the same container. The final solution volume was 50 mL. An additional filtration step through Millipore 0.45 μm filter was introduced, in order to prevent the errors due to suspended materials and to avoid the AAS nebulizer blocking with micro particles potentially remaining in the suspension. Analytical determinations were carried out on four independent replicates for all samples. Blanks were run simultaneously at all stages of the procedure. Only blank values below the instrumental limits of detection were considered acceptable.

For total metal determinations, 1.0000 ± 0.0005 g of dried sediment was digested as described by Tessier et al.¹⁵⁾ in a PTFE container. The recovery values (%) were investigated using the results of total metal analyses. Sequential and total extractions were also performed on JMS-1 provided by the Geological Survey of Japan. All strong acid mixtures were prepared just before the analysis of total metal contents.

(4) Statistical analyses

Statistical analyses were performed to compare the results as described by Douglas²⁰⁾. Comparisons were made by using one way analysis of variance

Table 2 Comparative summary of the four sequential extraction procedures

Steps	SM&T scheme	Tessier scheme	Grimalt scheme	Hall scheme
Soluble	-	-	Millipore water (10 mL); then 90 min. stirring	-
Exchangeable*	20 mL of 0.11 M HOAc; 16 h stirring at 22±5°C	8 mL of 1 M MgCl ₂ (pH=7); then stirring 1 h	8 mL of 1 M NH ₄ OAc; then 60 min. stirring	20 mL of 1 M NaOAc (pH=5) and 6 h stirring
Associated to Carbonates*		8 mL of 1 M NaOAc (pH=5); then stirring 6 h	8 mL of 1 M NaOAc (pH=5); then 6 h stirring	
Amorphous Fe oxyhydroxide**	20 mL of 0.5 M NH ₂ OH.HCl, pH=2 adjusted with 2 M HNO ₃ ; 16 h stirring at 22±5°C	20 mL of 0.04M NH ₂ OH.HCl in 25% HOAc; digest for 6 h at 96±3 °C	15 mL of 0.04 M NH ₂ OH.HCl and total evaporation; again add 10 mL of 0.04 M NH ₂ OH.HCl (dryness); Redissolution with 10 mL of 1 M HNO ₃	20 mL of 0.25 M NH ₂ OH.HCl in 0.25 M HCl heated for 2 h at 60°C and vortex 3 times
Crystalline Fe oxide**				30 mL of 1 M NH ₂ OH.HCl in 25% HOAc; 90°C for 3 h, vortex 9 times
Oxidisable/ Associated to organic matter and sulfides	5 mL 8.8 M H ₂ O ₂ , digest 1 h at 25°C; then digestion for 1 h at 85±2°C; 50% evaporation; add 5 mL 8.8 M H ₂ O ₂ and digest for 1 h at 85±2°C; 90% evaporation ; 25 mL of 1 M NH ₄ OAc (pH=2); 16 h stirring at 22±5°C	3 mL of 0.02 M HNO ₃ + 5 mL of 30% H ₂ O ₂ (pH=2) digest for 2 h at 85±2°C; add 3 mL of 30% H ₂ O ₂ (pH=2), heated again for 3 h at 85±2°C; cool and then add 5 mL of 3.2 M NH ₄ OAc in 20% HNO ₃ and stir for 30 min.	5 mL of 0.02 M HNO ₃ + 5 mL of 30% H ₂ O ₂ digest for 30 min. at 85°C; again add 3 mL of 30% H ₂ O ₂ and digest for 30 mins. at 85°C; add 5 mL of 3M NH ₄ OAc and 30 min. stirring	Add 750 mg KClO ₃ + 5 mL of 12 M HCl, vortex; again add 10 mL of 12 M HCl, vortex; after 30 min. add 15 mL water, vortex; finally add 10 mL of 4 M HNO ₃ and vortex. Heat the content at 90°C for 20 min.
Residual and silicates	6 mL of aqua regia (HNO ₃ : HCl = 1 : 3); evaporate to dryness at 150°C; Repeat - with 4 mL of aqua regia; dissolution with 2 M HNO ₃	HF:HClO ₄ = 5:1 (12 mL) (dryness); again HF:HClO ₄ = 10:1 (11 mL) (dryness); then add 1 mL HClO ₄ (until white fumes); then dissolution with 12 N HCl	10 mL of HNO ₃ : HClO ₄ = 4:1 digest at 90°C for 2 h	2 mL of 16 M HNO ₃ , heat at 200°C to dryness, cool, add 2 mL 12 M HCl, heat; add 10 mL of HF: HClO ₄ : HNO ₃ = 5: 3: 2 heat at 120°C to dryness; add 1 mL HCl + 3 mL HNO ₃ + 3 mL H ₂ O and warm.

* combined in SM&T and Hall schemes as Exchangeable + Carbonatic and AEC (adsorbed, exchangeable and carbonate), respectively

** combined in three schemes as Reducible (SM&T)/ Bound to Fe-Mn oxides (Tessier)/ Associated to oxides (Grimalt)

(ANOVA) with log transformed data, which was followed by post hoc (Tukey's) tests, as appropriate. Measurements are given as mean $\pm$ standard error, unless noted.

3. RESULTS AND DISCUSSION

(1) Major elemental composition of the samples

Major oxide components of sediment samples are listed in Table 3. It is apparent from Table 3 that NR sediment samples contained higher amounts of iron (approx. 4 times), magnesium (almost double), calcium (about twice) and phosphorus (about 12 times) than TR sediment. Major component concentrations suggest, besides the obvious presence of quartz, other aluminosilicate minerals, as well as the presence of iron oxides, and to a lesser extent, manganese, titanium and phosphorus containing mineral phases. Analytical recoveries for the major elements from JMS-1 ranged 94-108 % except phosphorus (83%).

Additional independent information on the component geochemical phases of all the samples studied was obtained by XRD analysis. Due to the lack of sensitivity of the technique in the case of trace/ minor components, only major crystalline phases could be identified. Quartz, feldspars (such as albite/ anorthite) and calcite were common in both river sediment samples. It is worth mentioning that the chlorite was detected only in NR sediments. Minerals of the mica group (muscovite, biotite) together with quartz and feldspar were detected in NR sediments and JMS-1 standard material. Additionally, the presence of iron hydroxide group

minerals, specifically goethite, were detected in NR and JMS-1 reference material and iron phosphate group mineral in NR sediment. The mineral composition of all the samples agrees qualitatively with the major elemental composition determined by XRF.

(2) Comparison on the basis of total metal contents and elemental recoveries

A comparison between total concentrations obtained by strong acid attack and the sum of the individual concentrations in the fractions of sequential extraction scheme was done for all the schemes and samples used and the results are presented in Table 4, where, each value is the mean of four independent determinations, and the standard deviation ($\pm$ SD) is also added. This comparison offers an alternative path to check for possible systematic errors arising from element losses, contamination or other undetermined causes during the numerous manipulations carried out in the course of sequential extractions. Validation of the method was performed by similar analysis of JMS-1. The determined and certified reference concentration values have been included in Table 4 and found to be in acceptable agreement. However, a rigorous validation of the complete sequential methods through assessment of accuracy was not possible because standard reference materials certified regarding the phase dependent composition were not available to us although there are reports of some such certified materials found in literature^{21, 22, 23}.

A comparison was also done between the total metals contents obtained by single extraction and the

Table 3 Major constituents and their concentrations (mass %) obtained by XRF in different sediment samples and geological standard (JMS-1)

Samples		Major constituents									
		SiO ₂	TiO ₂	Al ₂ O ₃	Fe ₂ O ₃	MnO	MgO	CaO	Na ₂ O	K ₂ O	P ₂ O ₅
NR - 1		58.62	0.81	13.33	15.47	0.13	3.30	2.87	2.55	1.90	1.03
NR - 2		57.84	0.71	13.20	16.81	0.12	3.17	2.43	2.44	1.80	1.39
TR - 1		76.94	0.45	11.11	4.25	0.11	1.75	1.26	1.99	2.09	0.06
TR - 2		73.71	0.59	12.54	5.28	0.10	1.63	1.41	2.24	2.36	0.15
JMS-1	Measured*	60.73	0.83	17.65	8.19	0.13	3.04	2.35	4.31	2.60	0.17
	Certified**	60.71	0.79	17.87	7.52	0.12	3.24	2.41	4.60	2.53	0.20

* Ignited matter basis; ** Dry matter basis

Table 4 Comparison between total metal concentrations (in $\mu\text{g g}^{-1} \pm \text{SD}^{\text{a}}$) and the sum of fractions of different schemes

Sample	Schemes	Heavy Metals				
		Cr	Cu	Ni	Pb	Zn
NR-1	Total	916 $\pm$ 7.9	362 $\pm$ 2.0	126 $\pm$ 0.9	229 $\pm$ 1.8	1197 $\pm$ 1.23
	SM&T	884 $\pm$ 11.2	339 $\pm$ 6.2	122 $\pm$ 2.6	227 $\pm$ 1.2	1147 $\pm$ 4.1
	Tessier	914 $\pm$ 2.6	379 $\pm$ 6.7	104 $\pm$ 1.3	239 $\pm$ 2.6	1169 $\pm$ 14.3
	Grimalt	672 $\pm$ 7.1	307 $\pm$ 3.4	104 $\pm$ 2.5	188 $\pm$ 1.7	1057 $\pm$ 8.2
	Hall	911 $\pm$ 3.6	390 $\pm$ 2.0	118 $\pm$ 1.5	235 $\pm$ 1.1	1165 $\pm$ 6.5
NR-2	Total	822 $\pm$ 4.7	344 $\pm$ 2.1	113 $\pm$ 1.7	204 $\pm$ 2.2	1157 $\pm$ 5.3
	SM&T	807 $\pm$ 9.4	325 $\pm$ 4.9	109 $\pm$ 2.0	209 $\pm$ 2.1	1103 $\pm$ 6.4
	Tessier	832 $\pm$ 5.2	359 $\pm$ 4.6	96.3 $\pm$ 2.7	204 $\pm$ 3.3	1163 $\pm$ 9.7
	Grimalt	630 $\pm$ 9.9	306 $\pm$ 5.0	95.5 $\pm$ 0.9	195 $\pm$ 3.1	1125 $\pm$ 7.0
	Hall	828 $\pm$ 6.1	379 $\pm$ 3.4	103 $\pm$ 0.8	230 $\pm$ 2.3	1160 $\pm$ 5.4
TR-1	Total	266 $\pm$ 2.8	27.3 $\pm$ 0.2	85.5 $\pm$ 2.2	34.7 $\pm$ 0.5	136 $\pm$ 2.2
	SM&T	233 $\pm$ 6.7	28.8 $\pm$ 1.7	85.5 $\pm$ 0.4	34.2 $\pm$ 1.3	133 $\pm$ 3.9
	Tessier	241 $\pm$ 6.4	26.5 $\pm$ 1.0	81.6 $\pm$ 2.8	34.2 $\pm$ 0.6	133 $\pm$ 4.7
	Grimalt	139 $\pm$ 5.8	27.3 $\pm$ 0.8	72.0 $\pm$ 1.5	28.1 $\pm$ 1.4	115 $\pm$ 4.7
	Hall	245 $\pm$ 4.1	28.9 $\pm$ 0.9	80.3 $\pm$ 1.7	35.9 $\pm$ 0.4	141 $\pm$ 3.1
TR-2	Total	296 $\pm$ 3.2	33.1 $\pm$ 1.2	61.0 $\pm$ 1.4	28.6 $\pm$ 1.0	159 $\pm$ 3.0
	SM&T	256 $\pm$ 5.9	30.5 $\pm$ 2.1	60.9 $\pm$ 0.5	27.6 $\pm$ 1.4	159 $\pm$ 2.1
	Tessier	274 $\pm$ 7.5	33.3 $\pm$ 2.2	58.2 $\pm$ 1.9	29.5 $\pm$ 3.2	157 $\pm$ 6.1
	Grimalt	165 $\pm$ 7.5	28.6 $\pm$ 0.6	53.5 $\pm$ 2.5	21.7 $\pm$ 1.5	129 $\pm$ 3.6
	Hall	273 $\pm$ 5.2	36.3 $\pm$ 1.3	54.7 $\pm$ 1.6	31.1 $\pm$ 1.2	159 $\pm$ 2.0
JMS-1	Total	138 $\pm$ 2.9	84.9 $\pm$ 2.0	57.9 $\pm$ 1.3	49.2 $\pm$ 1.5	274 $\pm$ 2.2
	SM&T	115 $\pm$ 1.6	89.9 $\pm$ 2.4	61.5 $\pm$ 1.2	49.4 $\pm$ 0.9	270 $\pm$ 2.6
	Tessier	140 $\pm$ 1.5	81.2 $\pm$ 0.4	57.7 $\pm$ 0.8	49.3 $\pm$ 2.1	272 $\pm$ 3.3
	Grimalt	89.1 $\pm$ 1.9	75.6 $\pm$ 1.3	59.3 $\pm$ 1.4	42.4 $\pm$ 1.8	260 $\pm$ 4.1
	Hall	142 $\pm$ 1.1	91.8 $\pm$ 1.9	58.1 $\pm$ 0.8	48.8 $\pm$ 0.8	263 $\pm$ 3.0
	Certified values	133 $\pm$ 2	88 $\pm$ 2	53 $\pm$ 2	49 $\pm$ 2	264 $\pm$ 3

^a Number of replicates: 4, SD: Standard deviation

levels for the four schemes based upon the sum of sequential extraction steps for each element by using Student's t -test²⁰). When it was done, there was no significant difference observed for Cu and Pb at 95% confidence level for all river sediment samples and JMS-1. But there were significant differences for Cr, Ni and Zn for NR-1, NR-2, TR-1, TR-2 and JMS-1 at $\alpha = 0.05$. It is evident from Table 4 that the Cr and Zn

contents especially the Cr content obtained from the Grimalt scheme were very much lower than those of the others schemes used. On the other hand, the Ni content for the SM&T scheme was higher than that of the others schemes at 99% confidence level. But their standard deviation values, which were lower than those of the others, indicate that the sequential extraction schemes produce good

reproducible results.

Validation of the analytical results was also tested by recovery calculations. An important consideration in the reliability of sequential extraction data is the percent recovery relative to a total digestion. The apparent recovery was calculated following the method as described by Sutherland et al.²⁴⁾:

$$\text{Recovery}_n = \frac{\sum_n \text{Sequential extraction procedure} \times 100}{\text{Total digestion with strong acids}}$$

where, n is the concentration of a given element in fraction n.

Single (total) digestion procedure employing strong acids mixture was applied to each sample and replicated four times. The average analytical results acquired are illustrated in Fig. 1. The recovery values of the tested elements for all the four schemes were calculated according to the equation mentioned above and generally, agreed with each other although some recoveries deviated from acceptable values (between 90 and 110%) at the 90% confidence level. It is apparent from Fig. 1 that the recoveries obtained with the Grimalt method were lower for all the metals studied in all the samples, with the exception of Ni in JMS-1. Recovery of Cr using the Grimalt scheme was poor with values of 75% for NR; 54% for TR and 65% for JMS-1. The residual fraction of this scheme for all the elements showed a negative departure consistently. This indicated that for the materials studied the last extraction stage of the Grimalt scheme was unable to remove elements strongly fixed in the crystalline lattice. It must be remarked from Table 2 that no HF was employed and the extraction time was also shorter in that step, preventing an exhaustive attack to the residue. The recovery for Cr was also comparatively lower with the SM&T scheme in TR (87%) and JMS-1 (83%) samples, but for NR sample the recovery was 97%, may be due to its muddy nature. Thus, it can be concluded that in this case the other two schemes provided a more complete and reliable fractionation of the metal. On the other hand, the Tessier scheme produced 91-105% recovery with an exception of Ni (84 %) in NR sediment. This may be due to the single extraction, which was done after the method described by Tessier et al.¹⁵⁾. Generally, good recoveries were achieved with the Hall procedure and the range was 92 - 109% (Fig. 1). However, during the use of sequential extraction schemes for fractionation of metals, sample contamination or loss could occur and this phenomenon may cause the experimental errors, which means, obtaining low or high recovery.

Consequently, because the experimental precision depends not only upon the care taken in performing the multiple extraction steps and the instrumental calibration and measurement, but also on the homogeneity of the treated material, these results are not unexpected²²⁾.

Fig.1 Comparison between the total metal concentrations and sum of fractions among the schemes used on the basis of recovery (%) in sediments and standard samples.

(3) Influence of sequential extraction schemes on the fractionation pattern of metals at each extraction step

The comparison among the one 4-step (the SM&T), the two 5-step (Tessier and Hall) and the one 6-step (Grimalt) sequential extraction procedures was carried with JMS-1 and the four different sediment samples collected from two rivers in Japan. The metal concentrations in percentage based on the summation of all the sequential extraction steps excluding the residual phase fractions, determined at each extraction step for the four different sequential extraction schemes are illustrated in Figs. 2-6. The residual phase was not considered in the comparison because the SM&T and Grimalt schemes did not employ HF in the dissolution of metal at this phase. Thus, there is some comparability in total phase partitioning of metal, especially for Cr and Ni. It was also reported earlier that without HF, the extractants are unable to remove metals trapped in the silicate matrices. Thus, preventing an exhaustive attack to the residue²⁵⁾. However, the results obtained from the Figs. 2-6, indicate that each metal has a characteristic distribution pattern. The order of the non-residual fractions (i.e. the first 3-steps for the SM&T scheme, the first 4-steps for the Tessier and Hall schemes and the first 5-steps for the Grimalt scheme) of the metals in all the samples obtained by the schemes are: Zn > Pb > Ni > Cu > Cr for SM&T, Pb > Zn > Cu > Cr > Ni for Tessier, Pb > Zn > Cu > Cr > Ni for Grimalt and Pb > Cu > Zn > Ni > Cr for Hall scheme.

In an average, the lowest mobility was observed

for Ni and Cr for the four extraction schemes. The following sections describe element phase distributions according to the sequential extraction procedures applied in this study. For better comparison and to get much more information on the relative advantages of each schemes, the data obtained from the non-residual phases of the four schemes were grouped into the following three phases: Easily extractable (upto carbonate), Fe-Mn oxides occluded and Organic matter (sulfides) bounded.

a) Chromium

The majority of Cr was released from non-residual phases of all the four schemes used for NR (overall range 85-93%, exception in SM&T scheme, 65%) and JMS-1 (overall range 68-69%, exception in Tessier scheme, 39%) samples but in case of TR samples, the residual phase was higher (76-87%) with the exception in the Grimalt (52%) and Hall (49%) schemes. From Fig. 2, it is evident that a relatively high proportion of Cr in NR samples (90%) was bound to organic matter phase of SM&T scheme and 64-76% was in Fe-Mn oxides occluded phases of other three schemes. In case of TR and JMS-1 samples, a similar trend was also observed for non-residual Cr. It was apparent from the results for all the samples that the extractants and techniques used in the Fe-Mn oxides occluded and organic matter bounded phases of Hall scheme were more aggressive compared to the extractants used in the others schemes and for this reason the residual phase of this scheme was the lowest among the schemes. On the other hand, the most effective extractants were 0.25 M $\text{NH}_2\text{OH} \cdot \text{HCl}$ in 0.25 M HCl + heating at 60°C for 2 hrs and 1 M $\text{NH}_2\text{OH} \cdot \text{HCl}$ in 25% HOAc + heating at 90°C for 3 hrs used for the leaching of Cr bound to Fe-Mn oxides occluded phase of the Hall scheme (amorphous Fe oxyhydroxide and crystalline Fe oxide, respectively) with an extractable ratio (extracted: total released by single extraction) of 70%, 24% and 43% for NR, TR and JMS-1, respectively. This may be due to the formation of relatively stable organo-metal complexes with more concentrated strong reductant ($\text{NH}_2\text{OH} \cdot \text{HCl}$) in presence of HCl and subsequently weak acid (HOAc). Furthermore, heating process may also accelerate the extraction process. This agrees, with some reports^{26, 27} published earlier. However, it is worth mentioning that when the one way ANOVA was done, no significant difference was observed between any two schemes on the basis of the total non residual Cr contents for all the samples.

Fig.2 Distribution of Cr among the different non-residual phases of different schemes in sediments and standard samples.

Fig.3 Distribution of Cu among the different non-residual phases of different schemes in sediments and standard samples.

b) Copper

The analytical results from each extraction step of all the schemes and samples used for Cu are illustrated in Fig. 3. The total extractable Cu content with extractants used in the SM&T, Tessier and Grimalt schemes were almost similar for NR and JMS-1 (89-91 and 72-79%, respectively) samples whereas for the Hall scheme, the amount was 97 and 87%, respectively. On the other hand, in case of TR sediments, total non-residual Cu content with the extractants used among the schemes showed the sequence as: Hall > Grimalt > Tessier > SM&T. From the Fig. 3, it is evident that the extractants used in the Hall scheme in bound to Fe-Mn oxides occluded phase was more aggressive followed by Grimalt scheme (0.04 M $\text{NH}_2\text{OH} \cdot \text{HCl}$ and dissolution in 1 M HNO_3) and the extractant used in Tessier scheme (0.04 M $\text{NH}_2\text{OH} \cdot \text{HCl}$ in 25% HOAc) was the least effective, because there was no non-coordinating counter-ion like HNO_3 (Table 2). May be this was the reason to obtain the highest amount (66-96%) of non-residual Cu for the extractants used in organic matter bounded phase (0.02 M HNO_3 + 30% H_2O_2 (pH=2) + heating and

after cooling, shaking 30 min. with 3.2 M NH_4OAc in 20% HNO_3) in Tessier scheme. However, there was no significant difference between any two schemes on the basis of the total non-residual Cu contents for all the samples by using one way ANOVA.

c) Nickel

There were significant differences observed between the average non-residual Ni levels of the Hall scheme and the others by using one way ANOVA followed by post hoc test at 99% level of confidence for all the samples used but among the others three schemes, there were no significant differences found for total non-residual Ni contents. The results for each extraction step are depicted in Fig. 4. It should be noted here that the average residual Ni contents for Hall scheme was lower than the others for all the samples examined. The Fe-Mn oxides occluded phase of Hall scheme (amorphous Fe oxyhydroxide and crystalline Fe oxide) was the most effective extractants for leaching of Ni with an extractable ratio (extracted: total released by single extraction) of 40%, 53% and 38% for NR, TR and JMS-1, respectively and the reason may be the use of more concentrated $\text{NH}_2\text{OH.HCl}$ with HCl and subsequently HOAc as well as the differences in the extraction conditions (heating) than other schemes. It has been reported by many authors^{26, 27, 28} that higher concentration of extractant affected extractability. On the other hand, there are reports that temperature can substantially affect the extractability²⁷ and resulted in faster rates of leaching with higher amount of metals²⁶ at Fe-Mn oxides occluded phase.

Fig.4 Distribution of Ni among the different non-residual phases of different schemes in sediments and standard samples.

d) Lead

The analytical results for each extraction step for Pb are illustrated in Fig. 5. The total extractable Pb differed between schemes with the overall sequence

being: Hall > Grimalt > Tessier > SM&T. But it should be mentioned here that the sum of total concentration obtained by the different phases of Grimalt scheme was always lower than others for all the samples used (Table 4). It is apparent from Fig. 5 that the Fe-Mn oxides occluded phase of Tessier scheme extracted the lowest amount (18%, 21% and 66% for NR, TR and JMS-1, respectively) of Pb among the similar phases of other schemes for all the samples used. However, the subsequent step (organic matter bounded phase) of the Tessier scheme extracted the highest amount (34-80%) of Pb compared to the other schemes. This reflects the lower concentration of $\text{NH}_2\text{OH.HCl}$ (0.04 M) and weak acid (HOAc), used in Fe-Mn oxides occluded phase of Tessier scheme. On the other hand, in the same phase of the SM&T and Grimalt schemes, HNO_3 (non-coordinating counter-ion) and in the Hall scheme, HCl were used with concentrated $\text{NH}_2\text{OH.HCl}$ and replicate extraction was done with more concentrated $\text{NH}_2\text{OH.HCl}$ and HOAc, which may be contributed to extract more Pb (Table 2). This agrees with the reports^{26, 27, 28} published earlier. Finally, the organic matter bounded phase (750 mg $\text{KClO}_3 + 12 \text{ M HCl} + \text{H}_2\text{O} + 4 \text{ M HNO}_3 + \text{heating at } 90^\circ\text{C for 20 min.}$) of Hall scheme showed the poorest effect for leaching this metal may be due to the effectiveness of the previous phases. On the other hand, it is well known that Pb forms stable complexes with acetate^{1, 29}, which was used in other three schemes with H_2O_2 . However, the total non-residual Pb contents for the SM&T scheme differ significantly from the others schemes at 95% level of confidence for all the samples used by using one way ANOVA followed by post hoc test but among the other three schemes, there were no significant differences for total non-residual Pb content.

Fig.5 Distribution of Pb among the different non-residual phases of different schemes in sediments and standard samples.

e) Zinc

Fig.6 Distribution of Zn among the different non-residual phases of different schemes in sediments and standard samples.

The total non-residual Zn contents were approximately 52-92%, 58-92%, 72-94% and 84-96% for the schemes SM&T, Tessier, Grimalt and Hall, respectively. The results for each extraction step of the four extraction procedures are depicted in Fig. 6. As a consequence, there were no significant differences between the total non-residual Zn contents for all the samples and all sequential schemes at 95% confidence level. However, it is evident from Fig. 6 that the easily extractable phase of the SM&T scheme (extractant: 0.11 M acetic acid + 16 hrs stirring) had slightly more leaching capacity than the others three schemes (extractants for all: 1M sodium acetate of pH 5 + 6hrs stirring) for all the samples. It should also be mentioned here, in Hall scheme the amount of extractant was more than twice than Tessier and Grimalt schemes [Table 2], which may contribute to release more Zn in this phase. So this was obviously, due to the nature and amount of extractants as well as extraction condition used in the scheme. Furthermore, Fig. 6 also reflects that the Fe-Mn oxides occluded phase of the three schemes (except SM&T) had more effectiveness for the fractionation of Zn. The sequence of the schemes on the basis of the most effective extractant in this phase for all the samples was: Hall > Tessier $\geq$ Grimalt. Shiowatana et al.²⁶⁾ reported that ambient temperature is unable to completely extract the reducible phase. They also found further leaching occurring when temperature raised from 65°C to 80°C especially for Zn and Fe. Differences in the extraction conditions among the schemes, and in particular, the ability of the chemical reagents used in each case to target particular geochemical phases, should affect all metals more or less in the same way. Furthermore, during sequential extraction

procedures each metal would behave according to its physicochemical properties. According to Mester et al.¹⁾, among the metals studied, Zn has the most stability over a wide Eh-pH range in solution and it could be easily extracted without any redistribution effects.

4. CONCLUSIONS

The results of heavy metal fractionation in sediment samples obtained by four commonly used sequential extraction schemes were compared in terms of the distribution and bioavailability of metals at each extraction step. The highest mobility was observed for Pb and Zn among the four schemes while the lowest was for Cr and Ni. A significant fraction of the four metals (except Ni) appear as bound to Fe-Mn oxides especially for NR sediments, in agreement with the occurrence of minerals of the iron oxide group observed by the XRD analysis. This is well known that the sequential extraction procedures and the reagents used have substantially different effectiveness for extracting and/ or partitioning of the metals from the sediment samples, that is, the extractable heavy metal contents can be altered depending on the operational conditions used. In addition, there are a number of factors affecting a sequential extraction procedure such as, readsorption, concentration and type of the reagent, aim of application and others. However, with little exception, similarities in the distribution patterns resulting from all the sequential extraction schemes were observed.

Finally, on the basis of the obtained results of this study, the Hall method may be recommended as its extractability, reproducibility as well as stability of the results was comparatively higher than others. For all the studied elements, the Fe-Mn oxides occluded phase of this scheme was the most effective owing to the high tendency to form stable complexes for both the target phases and the analyte elements.

ACKNOWLEDGMENT: This work is partially supported by the 21st century COE program, Life Conjugated Chemistry (LCC), Keio University, Japan. The first author also gratefully acknowledges the financial support as Fellowship provided by the Yoshida Scholarship Foundation, 3-22-1 Kamezawa, Sumida-ku, Tokyo, Japan.

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(Received November 2, 2007)