

Study of speciation of metals in an industrial sludge and evaluation of metal chelators for their removal

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Abstract

Leachability and mobility of metals were evaluated in sludge obtained from electroplating industry, using toxicity characteristic leaching procedure (TCLP) and diethylene triamine penta acetic acid test (DTPA). Sequential leaching procedure was used to determine the chemical fractionation of metals. Further, removal of metals from sludge using various chelators (EDTA, citric acid, siderophore) was evaluated. The leaching test indicated that nickel in the sludge sample exceeded 5 ppm concentration. This categorized the sludge, as a toxic waste. The mobility of the metals in the sludge was in the following order: Ni, Fe, Zn, Cr, Pb. Metals were found to be associated with various fractions of sludge. The metal concentration in the sludge was very high as compared to normal abundance and thus not within the range to be used as a fertilizer. The low removal of metals from the sludge by various chelators may be due to overloading of metals and use of aged sludge rather than artificially contaminated one for the study. The results of this study bring forth the prospect of the use of siderophores for bioremediation, because it is biodegradable and ecofriendly. This can be achieved with further optimization of the method, exploration of more potent siderophores and by inclusion of metal bound sludge fraction specific treatments.

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1. Introduction

Rapid industrialization and urbanization has resulted in increased release of toxic heavy metals entering the biosphere. Solid and/or liquid wastes containing toxic heavy metals are generated in various industrial processes, e.g., in chemical manufacturing, thermal power generation, coal and ore mining, smelting and metal refining, metal plating and others. The sludge generated during the various industrial processes contains undesirable levels of toxic metals. Heavy metals containing sludge disposal is problematic because metals are non-biodegradable and tend to bioaccumulate. Metal sludge when applied to soil affects human and animal health, soil quality, water quality [1]. Excessive levels of heavy metals introduced to soil by sludge can lead to elevated uptake of heavy metals by plants, which causes

damage to plants and affect human health upon consumption of crops grown on the soil. To reduce the availability of heavy metals in sludge is therefore one of the major concerns in land application of sludge.

The Tamil Nadu Pollution Control Board (TN PCB) estimated that a factory in Ranipet, India, manufacturing chromium salts for a local leather tanning industry, accumulated about 1,500,000 tonnes of chromium sludge. This sludge when applied to land caused decrease in the fertility of the land. Foul smell and ulcerations on skins were the other troubles associated with the sludge, potentially affecting 3,500,000 people [62].

The critical, most polluted sites need immediate attention to remediate them. To be successful on a specific site, the remediation technique must be selected according to the form of retention of the heavy metal in the sludge.

Different forms of heavy metals have different mobility and phytoavailability [2]. Generally, the plant uptake of heavy metals is correlated to extractable forms of the metals rather than to the total metal contents in the soils [3]. Therefore, in order to assess the potential environmental impacts of sludge contaminated with metals, the determination of total, trace and heavy metal content

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alone is not sufficient, as it is the chemical form of the metal in the matrix that determine its behaviour in the environment and its mobilization capacity [4–8]. Leachability, availability and metal speciation studies provide more relevant information.

The Tessier et al. scheme [9] has been widely employed for metal fractionation in soils and sediments and sludges. The distribution of heavy metals among the exchangeable, carbonate, oxide, organic and residual fractions using sequential extraction analysis could assess the potential phytoavailability of heavy metals in industrial sludge. The form of metal retention in sludge is influenced by various external factors including pH, temperature, redox potential, organic matter decomposition, leaching, ion exchange processes and microbial activity [10].

The basic disposal methods for large quantities of sludge include land application, landfilling, incineration, ocean dumping and lagooning [11]. But new technologies have to be developed to manage toxic metal sludges in more economical ways. One such method of treating sludges is detoxication, so that they can be disposed off inexpensively as non-hazardous waste, or recycled. Some of the methods which are used for metal removal from sludges are hydrometallurgy for metal removal, bioleaching, hybrid process for heavy metal removal, electrokinetic removal or electroreclamation and use of plants for heavy metal removal (phytoextraction). These methods have their own advantages and disadvantages. Use of biomolecules like siderophores [12,13] can be an innovative ecofriendly method for the removal of metals from industrial sludge and has not been reported yet.

Siderophores are class of microbial chelating agents, which are low molecular weight ligands synthesized and excreted by bacteria for capturing and supplying iron to support metabolic activity. Siderophores are specific Fe(III) ligands and are able to bind to other metals, such as magnesium, manganese, chromium(III), gallium(III) and radionuclides, such as plutonium(IV) [14]. Siderophores binding to metals, including toxic ones like lead and cadmium, has been reported [15]. Reports are there of siderophore complexation with metals such as divalent heavy metals like zinc and actinides-thorium, plutonium [16].

The objectives of this research were to understand the mobility and leachability of metals in an industrial sludge. The speciation of the metals in the sludge was also studied. Subsequently, various metal chelators (organic, inorganic and biomolecule-siderophore) were evaluated for the removal of metals from sludge to convert toxic sludge in nontoxic for use as fertilizer and serve as one of the appropriate and beneficial methods for the sludge disposal.

2. Experimental

2.1. Sampling of industrial sludge

Industrial sludge was collected from an electroplating industry in Maharashtra, India. The semisolid sludge was collected from disposal ponds after thorough mixing, stored at 4 °C during transportation and then air-dried on arrival to the laboratory and, analyzed immediately.

2.2. Chemical characterization of the sludge

The dried sludge samples were sieved to remove the particles greater than 2 mm. The sludge samples were homogenized and kept in plastic bags for further analyses.

The sludge was analyzed chemically for pH and heavy metal content. The pH of the sludge was measured by soaking it in deionized water (H₂O:soil ratio of 2.5:1).

2.3. Metal analysis

2.3.1. Total metals

One gram of sludge (dry weight) was digested with aqua regia (a mixture of concentrated HCl and concentrated HNO₃) [17]. The suspension was cooled and filtered through a Whatman filter paper No. 40 and the filtrate was analyzed for metals by inductively coupled plasma spectrometry (ICP-AES-JY-24, Jobin Yvon, France).

2.3.2. Toxicity characteristic leaching procedure (TCLP)

Leachable metal concentrations were determined using the TCLP test as per USEPA Method 1311 [18]. The extraction fluid (5.7 mL of glacial acetic acid and 64.3 mL of 1.0N of sodium hydroxide in 500 mL of reagent grade water and diluted to a volume of 1 L with reagent grade water, pH 4.93) was added to sludge sample in 20:1 liquid to solid ratio and mixed in a rotary shaker for 18 h. Samples were then filtered through Whatman filter paper No. 40 and analysed for metals using ICP-AES.

2.3.3. Diethylene triamine pentaacetic acid test (DTPA)

The DTPA soil extraction was used to predict micronutrient available to plants in neutral to calcareous soils [19] and toxicity of these heavy metals [20]. Readily available, i.e. mobile fraction of metal was analysed using DTPA test [19]. The DTPA extraction solution (0.005 M DTPA, 0.01 M calcium chloride and 0.10 triethanol amine (TEA) adjusted to pH 7.3 ± 0.05 with 1.0N HCl) was added to the sludge in 2:1 liquid to solid ratio and mixed in a rotary shaker for 2 h. Samples were then filtered through Whatman filter paper No. 40 and analysed for metals using ICP-AES.

2.3.4. Sequential extraction

Sequential extraction procedures provide useful information for risk assessment, since the amount of metals mobilized under different environmental conditions can be estimated (weakly acidic conditions: exchangeable fraction, reducing conditions: Fe/Mn oxide fraction and oxidizing conditions: organic fraction). The mobility and bioavailability of the metals decrease approximately in the order of the extraction sequence.

A sequential extraction scheme developed by Fiedler et al. [21] was used to study partitioning of metals in the sludge into fractions namely water extractable, exchangeable, Fe–Mn oxides, organic matter bound and silicate minerals (residual). ICP-AES was used to quantify levels of heavy metals in different fractions. The experiment was performed in triplicate.

2.4. Siderophore production and quantification

Pseudomonas azotoformans (MTCC 7616) isolated by NEERI from an oil sludge collected from Mathura Refinery, UP, India, was used for the production of siderophore using sterile sodium succinate medium (SSM). The medium used does not contain any added source of iron and all the glasswares were washed with 6 M HCl to remove any traces of iron present. The standard SS medium contained (g L^{-1} in distilled water): K_2HPO_4 , 6.0; KH_2PO_4 , 3.0; $(\text{NH}_4)_2\text{SO}_4$, 1.0; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.2; sodium succinate, 4.0. [22]. The pH of the medium was adjusted to 7.0 by addition of NaOH prior to sterilization. For siderophore production, 10 mL of the culture was added to 500 mL of SS medium in 1000 mL Erlenmeyer flask and incubated at 30 °C in a rotary shaker at 175 rpm. After 48 h, the cells were separated from inoculated SS medium by centrifugation at 8000 rpm for 20 min and the supernatant was filtered through 0.45 μm membrane filter to ensure the complete removal of cells. Hydroxamate and catechol functionalities, respectively, of siderophore produced were determined by the Csaky test [23] and the Arnow reaction [24]. For these assays, hydroxylamine and 2,3-dihydroxybenzoic acid, respectively, were used as standards, and optical densities were read at 543 and 700 nm [25], respectively.

2.5. Batch studies for metal removal

A batch study was set up to investigate the efficiency of environmentally compatible, biological washing agent—siderophore for desorption of sludge bound metals in excess of mobile metal removed by distilled water. Efficiency of siderophore for metal removal was compared with an organic chelator – citric acid and a chemical chelator – ethylenediamine tetraacetic acid (EDTA). Uninoculated SSM was kept as the control. Siderophore produced in the production media was used directly for mobilization and leaching of metals from the sludge.

The sludge (5 g) was separately mixed with 25 mL of the washing solutions consisting of distilled water, siderophore, citric acid, EDTA (1:5, w/v). The pH of washing solution was adjusted to 7.0 ± 0.5 . The sample was shaken at 150 rpm. After 24 h, it was centrifuged at 5000 rpm for 10 min. The desorption of metals was studied by estimating the concentration of metal in the supernatant by ICP-AES. A series of five such washings were

given at an interval of 24 h and the metal concentration in the supernatant was determined after each washing. Changes in pH of the filtrate were monitored simultaneously. Same procedure as mentioned above was repeated with each washing solution. All samples were run in triplicate.

After sequential washings, the sludge was air-dried and metal concentration in the sludge was estimated for mass balance determination. Any change of pH before and after treatment of sludge with siderophore and other washing solutions was also noted.

2.6. Determination of leachable and mobile metals in treated sludge

Residual leachable metal concentration was determined by toxicity characteristic leaching procedure as per USEPA Method 3051 [18] and residual readily mobile metals in the treated metal sludge was analysed using diethylene triamine penta acetic acid (DTPA) test [19].

2.7. Mass balance determination

To achieve a final mass balance of metals in the sludge, residual metals in the treated sludge was determined. The treated sludge was mixed thoroughly, air-dried and 1.0 g of each was acid digested (aqua regia), filtered and the volume made up to 100 mL with double distilled water. Metal concentration was estimated by ICP-AES.

3. Results and discussion

3.1. Characterization of sludge

The sludge collected from the industry in Maharashtra was alkaline in nature with a pH of 9.02. The concentrations of Zn, Ni, Cr and Fe in the sludge were 48780.0, 176000.0, 36300.0 and 86420.0 mg kg^{-1} , respectively. The results of the chemical characterization of the sludge are presented in Table 1.

3.2. Leachable and mobile metal concentration of the sludge

The leaching test was applied in order to assess the behaviour of metals in terms of mobility so as to categorize the sludge as hazardous or nonhazardous and accordingly, can be disposed in landfills or used as fertilizer. The waste is considered toxic when the leaching test shows that extracted hazardous metals exceeded USEPA prescribed TCLP limits, which are (in ppm) arsenic (5), barium (100), cadmium (1), chromium (5), lead (5) and zinc (5).

Leachable metal concentrations determined with the TCLP test of the sludge is presented in Table 2. The results indicated the metal concentrations in the leachate were 22.0 and 12.54 mg kg^{-1} for Ni and Zn, respectively. Therefore, the metal sludge was classified as a hazardous waste. Mobile metal concentrations are presented in Table 3. In the sludge, available fractions of metals were very low as compared to that of the

Table 1
Characterization of raw sludge ($n = 5$)

Parameter	Average \pm S.D.
pH	9.02
Zn (mg kg^{-1})	48780.0 \pm 6.88
Pb (mg kg^{-1})	967.6 \pm 0.23
Cd (mg kg^{-1})	153.4 \pm 0.031
Co (mg kg^{-1})	1174.0 \pm 1.48
Cu (mg kg^{-1})	554.2 \pm 0.17
Ni (mg kg^{-1})	176000.0 \pm 121.03
Mn (mg kg^{-1})	643.4 \pm 0.16
Fe (mg kg^{-1})	86420.0 \pm 38.40
Cr (mg kg^{-1})	36300 \pm 13.79

Table 2
Metal removal by TCLP extraction from sludge

Metals	TCLP (permissible limit) ^a	TCLP (initial) ^b (mg kg ⁻¹)	TCLP (after treatment with washing agents) ^b (mg kg ⁻¹)				
			Untreated ^c	SSM ^c	DW ^c	SID ^c	CA ^c
Zn	5.0	12.54	8.42	21	21.0	35.2	306.0
Pb	5.0	5.26	0.3	0.6	0.28	0.58	1.56
Cd	1.0	0.22	1.36	1.34	1.4	1.34	1.4
Co	^d	0.65	0.08	0.1	0.06	0.14	0.14
Cu	^d	0.59	0.78	0.78	1.02	1.02	3.5
Ni	5.0	22.0	135.4	65.8	94.2	72.6	159.8
Mn	^d	1.26	2.28	3.92	2.12	5.36	3.5
Fe	^d	0.68	UD	UD	UD	UD	UD
Cr	5.0	5.46	0.34	0.16	1.48	0.86	0.1

UD, Undetectable; SSM, sodium succinate medium; DW, distilled water; SID, siderophore; CA, citric acid; EDTA, ethylenediamine tetraacetic acid.

^a USEPA, vide no. SW-846.

^b Determined by USEPA Method 3051 (USEPA, 1992).

^c Treatments.

^d Not enlisted.

metal loading. The mobility of the metals in the sludge is in the following order: Ni, Fe, Zn, Cr and Pb.

As shown in Tables 2 and 3, the TCLP and DTPA tests removed similar amounts of all metals except Ni. The DTPA soil test has been designed to prevent excess dissolution of CaCO₃ and thus extracts only the soluble and exchangeable fractions of contaminant metals [19]. The TCLP extraction is an acetic acid digestion at pH of 4.93 to dissolve carbonates and some more soluble oxides and thus removes the metals associated with these fractions in addition to the soluble and exchangeable. The absence of Fe concentration in the TCLP extract of the sludge (Table 2) indicates that no significant amount of iron oxide was extracted by the TCLP. Thus, the similarity between the mobile and extractable metal (except Ni) concentration indicates that they represent the soluble and exchangeable metal fractions and that there is no significant amount of carbonate-bound metals in the sludge under study.

3.3. Sequential extraction

The results of the distribution of metals in various fractions of the sludge are incorporated in Fig. 1.

Water soluble: For the sludge, the water soluble fraction was small or almost nonexistent for Zn, Pb, Ni, Co and Mn. Water

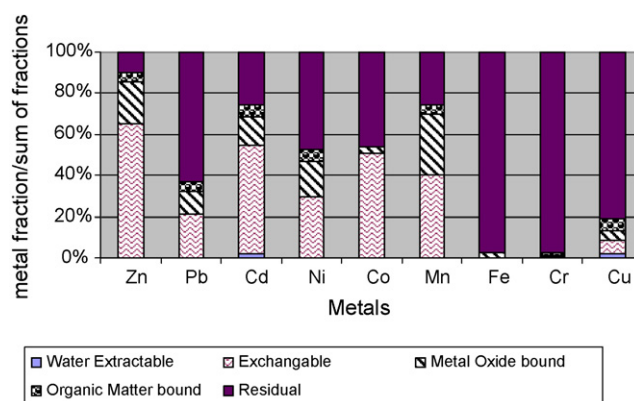


Fig. 1. Metal fractionation of sludge.

extractable Cu and Cd in the sludge was found to be 2.0% (Fig. 1).

Exchangeable: Most of the metals found in the sludge was in the exchangeable fraction. The percentage of Zn, Cd, Co, Mn, Ni, Pb and Cu in the sludge bound to the extractable fraction was 65, 52, 50, 40, 30, 21 and 6%, respectively (Fig. 1).

Metal oxide fraction: In the sludge, 21% of the Zn was found bound to Fe/Mn oxides, whereas only 17, 14, 11, 5, 4% of the

Table 3
Metal removal by diethylene triamine penta acetic acid (DTPA) extraction from sludge

Metals	DTPA (initial) ^a (mg kg ⁻¹)	DTPA (after treatment with washing agents) ^a (mg kg ⁻¹)				
	Untreated ^b	SSM ^b	DW ^b	SID ^b	CA ^b	EDTA ^b
Zn	4.73	22.40	6.84	75.00	23.60	235.60
Pb	1.22	1.09	0.17	1.06	1.077	1.13
Cd	0.30	0.49	0.13	0.46	0.50	0.33
Co	0.17	0.11	0.03	0.12	0.09	0.08
Cu	0.70	0.84	0.54	2.07	0.62	1.49
Ni	319.32	549.50	93.50	493.50	585.46	319.50
Mn	0.06	0.06	0.06	0.09	0.14	0.28
Fe	5.00	3.43	1.59	2.70	3.03	2.17
Cr	2.00	1.53	0.18	0.31	2.63	0.41

^a Determined by Lindsay and Norvell (1978) extraction method.

^b Treatments.

Ni, Cd, Pb, Cu and Co, respectively, was found on this fraction (Fig. 1).

Organic fraction: In the sludge, this metal fraction was between 0.1 and 6%, respectively. The organic fraction held 6% of Cu, Ni and Cd, 5% of the Pd, 4% of Zn and Mn, respectively. This fraction retained insignificant levels of all the other metals (Fig. 1). Heavy metals associated with sulfides can be available into reducible, carbonate and exchangeable fractions after an increase in the redox potential [26].

Residual fraction: Metals in this fraction are generally considered to be bound within the lattice of silicate minerals and can become available only after digestion with strong acids at elevated temperature. Heavy metals in the residual fraction were found to be as high as 97% in the sludge. The metals in the sludge are in the following order: Fe, Cr, Cu, Pb, Ni, Co, Cd, Mn and Zn.

Exchangeable metals are most readily available to biota. Metals in the acid extractable phase are very sensitive to pH changes and they are readily leached when the pH is decreased. These two metal fractions are thus considered mobile. The organic fraction released under oxidizing conditions is not considered mobile and bioavailable. These metals are incorporated into stable high molecular weight humic substances, which release small amount of metals over long time periods [27]. Degradation of organic matter under oxidizing conditions can lead to a release of the metals bound to these organic components [28]. Organic substances exhibit a high degree of selectivity for divalent ions compared to monovalent ions [29]. The residual or nonextractable metals are retained within the crystal lattices of minerals and inside crystallized oxides [30]. Metals confined in the residual fractions are usually not expected to be released over short period of time under the conditions usually encountered in nature [9,31,32]. Redox potential (Eh), organic matter, pH and other factors affects the chemical forms of metals in soil [31–33].

It is generally observed that the sum of the five metal fractions obtained from the sequential extraction procedure is higher than the total metal contents following acid digestion [34] and may be due to incomplete digestion with the mixed acid compared to the slow and increasingly stronger attack by the reagents of the sequential extraction. For this reason, it is more appropriate to refer to fractions in relative figures (metal concentration in certain fraction/sum of fractions). In our experiment fractions in relative figures was considered to avoid errors but our result is not in accordance with the above. The total heavy metal content was higher than the sum of the metal fraction from extraction procedure. This can be attributed to the limitations of the sequential extraction procedures which include metal redistribution during extraction, extractability being largely determined by minor pH differences, as well as possible chemical overloading of the system [35].

The results have shown that metals like Zn, Cd, Ni, Co, Mn were found to be highly mobile in the industrial sludge characterized. This observed high metal mobility is contradictory to the findings of Qiao and Ho [36], who reported that metals in sludge are generally found in very stable, insoluble forms which are mostly associated with the organic fraction, that fail to be

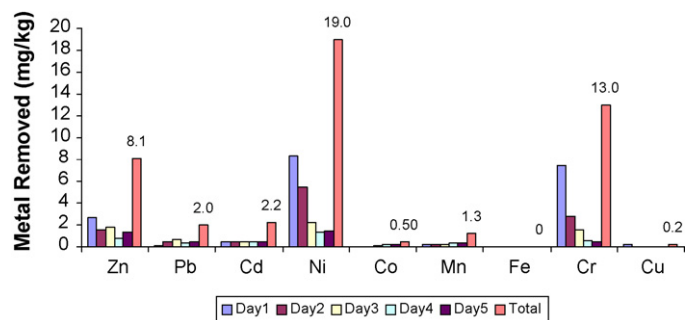


Fig. 2. Metal removal following five 24 h of sequential sludge washes by double distilled water.

extracted with DTPA, an extractant known to indicate metals available to plants [30].

In summary, it may be concluded that the industrial sludge was highly contaminated with Ni, Cr and Zn significantly in excess of the permissible levels for the application of sludge to agricultural land as regulated by the USEPA. Apart from these high total metal contents, substantial amounts of Mn and Fe were present in the most mobile forms, being the exchangeable and the acid extractable.

3.4. Siderophore production

With incubation, the colour change in the SS medium was observed which changed colourless to green and then brown on further incubation. This was indicative of siderophore production. The concentration of catechol type siderophore produced was found to be $3.88 \mu\text{g mL}^{-1}$. Whereas the concentration of hydroxamate type siderophore was found to be $1.46 \mu\text{g mL}^{-1}$. Thus, *P. azotoformans* in SS medium was found to produce a mixed type of siderophore both catechol and hydroxamate type, and quantitatively 5.3 mg L^{-1} .

3.5. Batch study for metal decontamination from sludge

Dissolution of the metal–mineral bond followed by the dispersion of the pollutant metal in the washing liquid as an emulsion, complex or suspension controls extraction of heavy metals from contaminated soil [37].

In this experiment, extractants included SSM, distilled water, siderophore (5.3 mg L^{-1}), citric acid (0.01 M) and EDTA (0.01 M). Metal removal rates by the various washing agents showed separate trends over the five extractions. The removal rates were represented as percent removal per day. The relative metal extraction efficiencies of distilled water, siderophore, EDTA, citric acid and SS medium were different for the contaminated sludge evaluated and is presented in Figs. 2–6.

Distilled water removed the available or weakly bound fraction of metals (water extractable), that act as mobile metal fraction. Distilled water showed two separate trends of metal removal. Metals like Pb, Co, Mn showed an increased removal over the five washings whereas Zn, Ni, Cr showed a decline in metal removal over the period of washing. Overall dis-

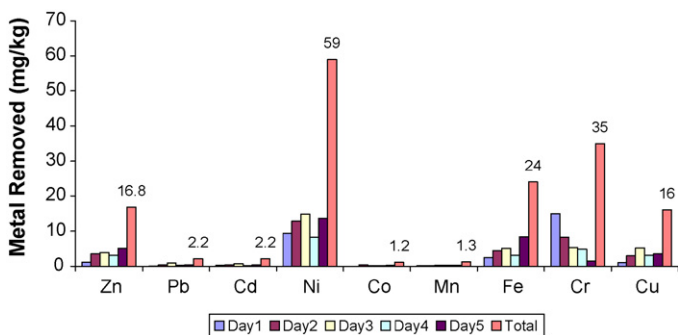


Fig. 3. Metal removal following five 24 h of sequential sludge washes by siderophore.

tilled water removed 8.1, 18.8 and 12.5 mg kg⁻¹ of Zn, Ni, Cr, respectively, and removal of other metals were negligible (Fig. 2).

Removal of metals by the washing agents indicated that the washing agents remove not only the leachable or available fraction of metal but also bound metal unlike by distilled water that removed the mobile metal fraction only.

Removal of metals by siderophore from the industrial sludge resulted in almost constant increasing rate throughout the five washings except for Cr that showed decreasing trend in removal over the extractions (Fig. 3). Nickel removal rates were less constant, but significant removal was observed throughout the five extractions. Siderophore showed highest removal of 59 mg kg⁻¹ of Ni followed by significant removal of 34.9, 23.6 mg kg⁻¹ of Cr and Fe, respectively. Siderophores are strong chelators of Fe [38]. Removal of metals other than Fe by siderophores has also been reported [14,15,39]. A higher removal of Ni and Cr as compared to Fe can be attributed to high concentration of Ni and Cr in the sludge. The stability constant of siderophore–Fe complex has been established [40]. Results showing greater complexation of siderophore with other metals initiate the further study of stability constants of these metal–siderophore complexes for better understanding. The constant or increasing extraction rates observed support the hypothesis that the metals are being solubilized with consecutive washes as suggested by the TCLP results. Thus, metal remediation using biological agents would require prolonged soil washing. These results confirm that the steady rate of metal removal observed with repeated extractions was due to such factors as increased chelant availability and mineral dissolution.

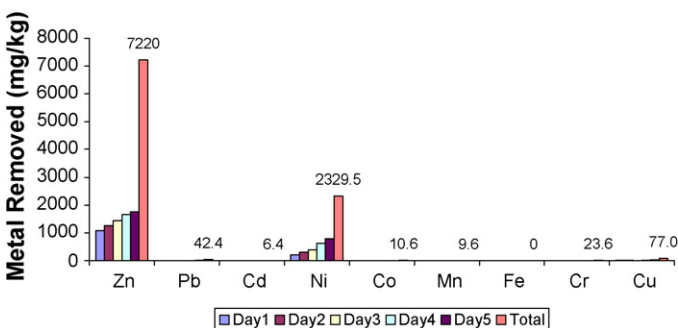


Fig. 4. Metal removal following five 24 h of sequential sludge washes by EDTA.

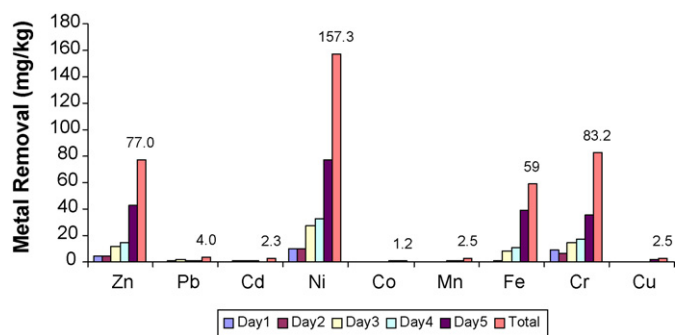


Fig. 5. Metal removal following five 24 h of sequential sludge washes by citric acid.

EDTA was found to be efficient for metal removal from sludge. Metal removal by EDTA followed an increasing pattern in the sludge (Fig. 4). Removal rates of metals from the sludge increased during all the extractions for Zn, Co and Ni. In contrast, inconsistent removal rate was observed for Pb, Mn, Cr from the sludge. Though EDTA showed high removal of Zn and Ni, i.e. 7220 and 1325 mg kg⁻¹, respectively, but there are many problems associated with use of EDTA in remediation. Concern has also arisen with both the potential toxicity of EDTA and its recalcitrance in the environment [41,42]. EDTA is biodegraded in soil under aerobic conditions at an estimated half-life of 120–300 days. In soil all metal species degraded at the same rate with exemption of the Ni–EDTA complex that degrades more slowly [63]. Both Tiedje [43] and Bolton [44] have demonstrated EDTA degradation by a variety of soils and sediments, but the residence time is extremely long. Tiedje [43] documented 13–46% mineralization of 2–4 mg EDTA kg⁻¹ soil in 13 surface soils after 15 weeks, but minimal degradation was observed in subsurface soil samples. Similarly, Bolton [44] observed 1–18% degradation of 0.01 mM EDTA in subsoils after 16 weeks. Means et al. [45] found EDTA associated with 12–15-year-old radioactive wastes, thus documenting the extreme persistence of EDTA in the environment. The addition of conditioning fluids, such as EDTA, ammonia, sodium acetate and water, has been shown to improve metal recoveries by increasing the fraction of metals in solution [46]. However, addition of conditioning fluids has the potential to mobilize the metals and spread the contamination to underlying soils and groundwater.

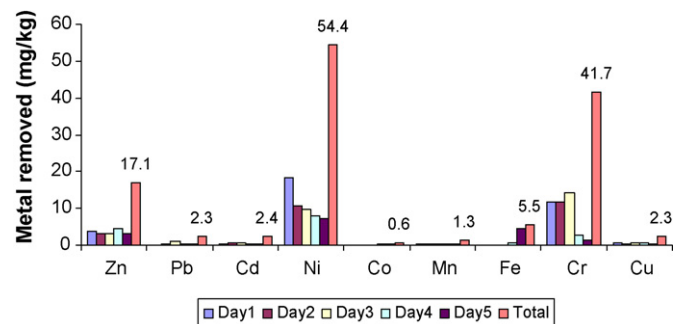


Fig. 6. Metal removal following five 24 h of sequential sludge washes by sodium succinate medium.

Citric acid showed a consistent increasing extraction rate of metals like Zn, Ni, Fe and Cr. Removal of other metals were significantly very less. Removal was in the order of Ni > Zn > Cr > Fe (Fig. 5).

The variation of results can be explained by the higher strength of complexation between metals and the synthetic chemical [47,48] as compared with between metals and the biological agents. This is in accordance with the results are reported in this study. But Hernlem et al. [12] and Neubauer et al. [13] indicated that siderophore has great potential as a bioremediation tool. Similarly, the results of this study bring forth the prospect of the use of siderophore in bioremediation because it is biodegradable and ecofriendly.

Aged contaminated soil rather than artificially contaminated soils, helps to realistically approximate the variation in metal lability found at contaminated sites [49,50]. Accurate assessment of the effectiveness of a washing agent for remediation of metal-contaminated soils requires the use of aged contaminated soils rather than artificially contaminated soils. In contaminated soils, bioavailability and mobility of metals are not only affected by soil physical properties and contamination history, but also by exposure to different environmental conditions [51–55]. Use of aged sludge rather than artificially contaminated one for the experiment establishes a real time application but suffers disadvantage of low removal of metals. This was proved for soils and may even stand significant to metal removal from sludge too. Frazer [56] also reported that although 80–100% of cadmium and lead can be removed from artificially contaminated soil but in field samples, the results were more in the range of 20–80%.

Thus, a low metal removal rate by siderophore as against the synthetic chelators does not end its scope for use in bioremediation of metal contaminated sludge, further optimization of the method and use of more potent siderophores produced by other organisms can facilitate the same.

Sludge remediation process using biomolecules can be made a closed system, more efficient and more applicable, by eliminating the major loopholes in system. Metal bound sludge fraction specific treatment can be of immense usage. Similar study of removal of metals from Fe/Mn by adding reducing agents has been carried out on sea nodules. The enforced reducing environment resulted in improved leaching by breaking up the oxide matrices [57–60]. Similarly, use of bioacid for removal of metals bound to silicate minerals was studied [60]. Such modifications should be incorporated in the sludge washing system. Further, after the washing the solubilized metal should be recovered safely in order to avoid environmental recontamination. Such a study on electrochemical lead recovery from leachate was carried out [61]. Such a method of metal concentration and recycle should be a part of the remediation system.

In control experiment with SSM, negligible amount of metal could be removed from sludge as compared to the total metals present. This removal was insignificant in comparison to the metals removed by washings with distilled water. Metal removal from sludge by SSM is presented in Fig. 6. Removal of metals from sludge by SSM can be attributed to the presence of sodium succinate in the medium, which is utilized by microorganisms during growth and is negligible in the media with produced

Table 4
Percent removal of metals by washing agents

Metals	Percent removal of metals by washing agents				
	DW	SID	EDTA	CA	SSM
Zn	0.02	0.03	14.80	0.16	0.03
Pb	0.21	0.22	4.38	0.41	0.24
Cd	1.41	1.43	4.17	1.49	1.56
Co	0.04	0.10	0.90	0.10	0.05
Cu	0.04	2.88	13.89	0.45	0.42
Ni	0.01	0.03	1.32	0.09	0.03
Mn	0.20	0.20	1.5	0.39	0.20
Fe	0	0.02	0	0.07	0.64
Cr	0.40	0.096	0.067	0.23	0.11

Table 5
Variation of pH in the filtrate

Number of days	Treatments				
	DW	SID	EDTA	CA	SSM
1	8.86	8.95	9.07	9.1	9.25
2	8.88	9.62	9.16	9.04	9.59
3	8.84	9.96	9.13	9.12	9.55
4	8.67	9.68	9.21	9.33	9.51
5	8.63	9.14	9.14	9.33	9.23

siderophore. The percent removal of the five washing agents for each metal is presented in Table 4.

The pH of the supernatant after treatment is indicated in Table 5. With the number of washings the pH variation was seen in the supernatant, which is perhaps due to metal dissolution and probable interaction of sludge, metal and chelators.

The final TCLP analysis of sludge was also significant (Table 2). Metal concentrations in the final TCLP extracts were observed to be either same or increased for almost all samples. The increase observed, suggested that the sequential washes solubilize not only the mobile fraction, but also a portion of the fractions previously characterized as immobile by the TCLP extraction after treatment with washing agents.

The pH of the metal contaminated sludge treated with washing solutions is presented in Fig. 7. The pH of the sludge after treatment with only DW has gone down whereas for other treatments it either increases or remains the same. This difference in the final pH can be attributed to the removal of metals by the

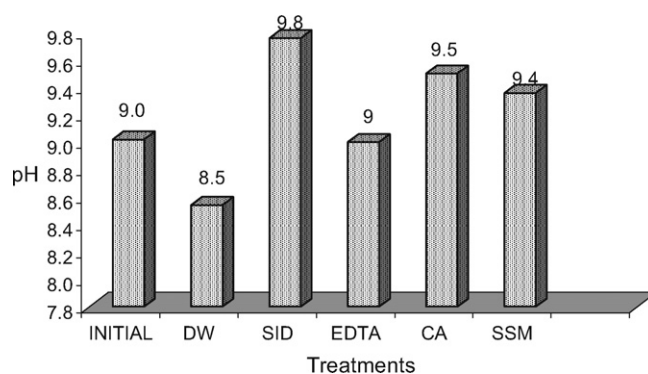


Fig. 7. Variation in pH of sludge treated with various washing agents.

Table 6
Metal mass balance

Metals	Zn	Pb	Cd	Ni	Co	Mn	Fe	Cr	Cu
Removed by DW	8	2	2.2	19	0.5	1.3	0	13	0.2
Remaining in sludge	75900	957	208	126000	828	651	70100	33600	530
Recovery (%)	111	99	108	72	71	101	81	93	96
Removed by siderophore	16.8	2.2	2.2	59	1.2	1.3	24	35	16
Remaining in sludge	74200	944	206	136000	742	604	65600	30900	503
Recovery (%)	108	98	107	77	63	94	76	85	94
Removed by EDTA	7220	42.4	6.4	2329.5	10.6	9.6	0	23.6	77
Remaining in sludge	37000	723	123	175000	651	539	77100	33000	423
Recovery (%)	65	79	68	101	56	85	89	91	90
Removed by CA	77	4	2.3	157.3	1.2	2.5	59	83.2	2.5
Remaining in sludge	39000	702	119	159000	595	537	77200	31600	492
Recovery (%)	57	73	63	90	51	84	89	87	89

All the values are in mg kg⁻¹ except recovery (%).

respective washing solutions from the sludge and interaction between sludge, metal and washing agents.

3.6. Heavy metal mass balances

Metal contaminated sludge washed by different extractants after the experiment were tested for the residual metal concentrations. Mass balances were found to be significant during the experiments. Percent metal recovery is presented in Table 6. The small difference in mass balances of the metal may be due to the analytical and/or operational errors, which are unavoidable during the experiments. Metals remaining in the washing agent treated sludge are also presented in Table 6.

4. Conclusions

- The industrial sludge is highly contaminated with metals like Ni, Zn and Cr. The TCLP test and DTPA test classifies it as hazardous waste and needs a treatment.
- The mobility of the metals in the sludge was in the following order: Ni, Fe, Zn, Cr, Pb. Most of the metals were found to be associated with silicates, as exchangeable or bound to Fe–Mn oxides except for Cr and Cu, which were mainly present on the silicate fraction. The various chelators used for metal removal from sludge showed differential removal, EDTA being the highest followed by citric acid and siderophore. The results of this study bring forth the prospect of the use of siderophore in bioremediation because it is biodegradable and ecofriendly. More potent siderophores produced by other microorganisms are to be explored and utilized. Similarly, metal bound sludge fraction specific treatment can be of immense usage and should be combined with washing of sludge with biomolecules.

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