Toxic metals (Ni$^{2+}$, Pb$^{2+}$, Hg$^{2+}$) binding affinity of dissolved organic matter (DOM) derived from different ages municipal landfill leachate

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Abstract
Solid waste production is rapidly increasing in Bangladesh and landfill leachate is the consequence of the decomposition of this waste. These leachates contain heavy metals and significant amount of dissolved organic matter (DOM). DOM is known to have considerable role in heavy metals speciation. Hence, it is important to characterize DOM/leachate and evaluate toxic metals binding affinity of DOM. The objectives of this study were to characterize the DOM in landfill leachate through physico-chemical and optical analyses and to investigate the toxic metals (Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$) binding affinity of three different ages (fresh sample L-1, young sample L-2 and mature sample L-3) DOM samples. Results suggested that leachate is a potential pollutant which contained very high organic pollutant load. Conditional stability constant (LogK) and percentages of fluorophores that correspond to metal binding (%f) values indicated that young DOM sample (L-2) had the highest binding affinity to all the three metals ions. In general, DOM samples showed the following order affinity to the metal ions; Ni$^{2+}$ binding affinity: L-2 > L-3 > L-1, Pb$^{2+}$ binding affinity: L-2 > L-3 > L-1 and Hg$^{2+}$ binding affinity: L-2 > L-1 > L-3.

Keywords Landfill leachate · Solid waste · DOM · Toxic metals · Binding affinity · Bangladesh

Introduction
Solid waste management has become a tremendous work for densely populated developing country due to the huge production rate of wastes and poor management infrastructure. Bangladesh is one of the rapidly developing countries with very dense population and poor waste management facilities in its major cities. More than 16,380 tons of wastes are produced in Bangladesh per day (Anonymous 2004) whereas about 7000 tons of solid wastes are produced only in Dhaka, capital city of Bangladesh (Hasan et al. 2009). As a developing country, the socio-economic situations of the country are mounting through fast industrialization and urbanization, so the amount of solid waste generation will continue to increase in future (Ahmed and Ali 2006). In Bangladesh, the composition of solid waste varies greatly as the wastes are not separated properly and all are dumped together. All types of waste such as redundant vegetables, food, plastics bags, broken batteries, thermometers, chemical waste are discarded together without appropriate segregation. In addition various industries dispose heavy metals containing wastes (Chamon et al. 2009). These wastes ultimately find their ways in different rivers, ponds, canals, roadside open spaces and only about half of them are dumped in open landfill sites as a cheapest mean of management (Ahmed and Ali 2006; Sujauddin et al. 2008). Huge amount of leachate is produced by the decomposition of these wastes that contain substantial amount of organic matter and heavy metals which possess serious threat to surrounding surface water, groundwater and soil (Renou et al. 2008), if disposed without proper treatment.

Unfortunately, high concentrations of heavy metals are found in the landfill leachate of Bangladesh. Several factors could contribute for the heavy metal input in landfill leachate. About 2.8 million metric tons of electronic wastes containing mercury (Hg) and lead (Pd) are generated each year which are disposed to landfills, agricultural land and water bodies (EDSO 2010). The presence of heavy metals in fertilizers (Yusuf et al. 2003; Naser et al. 2009) and...
anthropogenic activities such as agriculture and industrial activities increases the Pb and nickel (Ni) contents of soils and waters which have effects on the metal contents in vegetables (Alegría et al. 1991). These metals containing discarded vegetables in solid waste may also contribute to high metal level in landfill leachate. In composition, dissolved organic matter (DOM) is a major fraction of leachate which contributes more than 85% of total organic matter in terms of organic carbon in leachates (Zhang et al. 2009). This DOM has potential ability to bind heavy metal that affects their speciation (Wu et al. 2011). In previous study, it was found that, a considerable fraction of heavy metal was associated with DOM of landfill leachate (Baun and Christensen 2004). Strong affinity of humic substances (in DOM) toward heavy metals was found due to the presence of large number of functional groups in their structures (Wu et al. 2012; Terbouche et al. 2010) which help to form DOM–metal complexes. In aquatic environment and soil, DOM–metal complexes may completely alter the bioavailability and transport of many toxic metals (Zhao and Nelson 2005).

Dissolved organic matter composition depends and varies on the basis of leachate composition, broadly on solid wastes composition. In Bangladesh, DOM removal facility is not properly installed in the landfill sites and metals binding capacity of leachate-derived DOM was not investigated yet which could be different from other international studies’ results depending on the composition of solid waste of the country. Hence, this study was designed to determine the physico-chemical and optical properties of leachate (DOM) and to evaluate the toxic metals (Ni2+, Pb2+ and Hg2+) binding affinity of leachate-derived DOM. This research could be a promising work in the context of DOM–metal interactions in landfill leachate of Bangladesh.

Materials and methods

Sample collection and preparation

In this study, fifteen (15) leachate samples were collected. Five fresh leachate samples were prepared at laboratory in a controlled environment using solid waste collected from Amin Bazar Sanitary Landfill Site after 25 days of decomposition. Five young leachate samples and other five mature leachate samples were collected from leachate ponds of the Amin Bazar Sanitary Landfill Site, Dhaka, Bangladesh. In this article, fresh leachate samples, young leachate samples and mature leachate samples have been indicated by L-1, L-2 and L-3, respectively, based on their maturation/age. This landfill site was started as an open dumping site in 2007 which is now semi-aerobic sanitary landfill site. The samples were collected in pre-cleaned (by HNO3 and Milli-Q water) sampling bottles for physico-chemical analyses and filtered through pre-combusted (450 ºC for 3 h in a digital muffle furnace) 0.45 µm Whatman glass microfiber filter paper for optical and fluorescence analyses. For elemental analysis, each sample (250 mL) was acidified with 2 mL of concentrated HNO3 (APHA 1995). The sampling bottles were transferred to the laboratory by ice box that controlled the temperature below 4 ºC as early as possible.

Physico-chemical and optical analyses

Physico-chemical properties such as hydrogen ion concentration (pH), dissolve oxygen (DO), total dissolve solid (TDS) of leachate samples were determined by digital multi-meter (HACH, 51910). Electrical conductivity (EC) was measured using an electric conductivity meter (HANNA, HI 8033) and turbidity was measured by turbidity meter (HANNA, HI 93703). Biochemical oxygen demand (BOD) and chemical oxygen demand (COD) were measured using 5-day incubation method (APHA 1995) and closed reflux colorimetric method (Colorimeter HACH, DR/890), respectively. Then, the leachate samples were centrifuged with a rotating speed of 4000 r/min for 60 min using centrifuge machine (80-2, Shanghai Surgical Instrument Factory, China) and filtrated through glass-fiber membrane (0.45 µm) to remove suspended materials that may react with DOM. The organic matter in the filtrate was DOM. Dissolved organic carbon (DOC), specific UV absorbance at 254 nm (SUVA254), specific fluorescence intensity at 350/450 nm (SFI350/450) were measured using total organic carbon analyzer (TOC-L CPN, Shimadzu, Japan), UV spectrophotometer (SPECTORD 210 Plus, analytijkena) and fluorescence spectrophotometer (F-4600, Hitachi, Japan), respectively. Functional groups of DOM samples were identified using FT-IR spectrophotometer (IRPrestige-21, Shimadzu, Japan).

Elemental analysis of DOM samples

Initial concentrations of Ni, Pb and Hg present in the leachate samples were determined using atomic absorption spectroscopy (AA-7000, Shimadzu, Japan). Flame AAS was used to measure Ni and Pb concentration. Cold vapor AAS (CVAAS) was used for the measurement of Hg. The calibration curves were prepared for each metal individually applying linear correlation by least square method.

Fluorescence titration

Prior to fluorescence complexation titration, the DOM samples were diluted 100 times with Milli-Q water to ensure that the maximum fluorescence signals were below the upper detection limit of the spectrometer. For the purpose of complexation titration Ni(NO3)2, Pb(NO3)2 and Hg(NO3)2 salts were added to the samples to prepare 1, 5, 10, 15 and 20 µM
Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$ metal-DOM solutions. For this analysis, three representative samples were taken from L-1, L-2 and L-3 and after mixing with metal salts total numbers of samples were 48 (3 raw samples and 45 metals added samples). All the titrated solutions were shaken for 24 h at 25 °C to achieve complexation equilibrium. To maintain a constant pH condition before and after titration, the pH values of the metal titrants were adjusted to 6.3 for Ni(NO$_3$)$_2$, 5.0 for Pb(NO$_3$)$_2$ and 6.0 for Hg(NO$_3$)$_2$. Titration experiments were performed in duplicate. After titration, the fluorescence Excitation – Emission Matrices (EEM) spectra of the samples were measured using a Fluorescence Spectrophotometer (F-4600, Hitachi, Japan). Excitation wavelengths were scanned from 225 to 500 nm in 5 nm steps and the emitted fluorescence detected between 240 and 600 nm in 2 nm steps. Excitation, emission slit widths were 5 nm and scan speed was 1200 nm/min.

Modified Stern–Volmer equation was used to calculate complexation parameters (conditional stability constants and binding capacities) (Esteves Da Silva et al. 1998; Lu and Jaffe 2001).

\[
\frac{F_0}{\Delta F} = \frac{1}{fK[M]} + \frac{1}{f}
\]

where $F_0$ is the initial fluorescence intensities of the samples without metal addition, $F$ is the fluorescence intensities of the samples after metal addition, $\Delta F = F_0 - F$, $f$ is the fraction of the initial fluorescence that corresponds to the binding fluorophores, $K$ is the conditional stability constant, $[M]$ is the metal concentration. The logarithm of the conditional stability constant (Log $K$), percentage of fluorophores that correspond to metal binding (%$f$) were measured by using this equation. From the linear plot of $\frac{F_0}{\Delta F}$ versus $1/[M]$, $K$ and $f$ values were estimated using the slope $[(1/fK)]$ and the intercept $(1/f)$ (Esteves Da Silva et al. 1998).

### Results and discussion

#### General characteristics of leachate and DOM

Physicochemical and optical characteristics of leachate and DOM are listed in Table 1. Mean pH value for laboratory prepared fresh leachate samples (L-1) was 5.68 whereas for young (L-2) and mature samples (L-3), the values were 7.94 and 7.87, respectively. This indicated that in the early stage the leachate samples were acidic which gradually tended to become basic with increasing time. From Table 1 it is very clear that fresh leachate samples (L-1) had very high mean values for each physico-chemical parameter compared to others. The pH and TDS values in L-1 and EC, turbidity, BOD, COD and DO values of all the samples were beyond the standards for inland surface water discharge of Bangladesh (ECR 1997), which indicated that the leachate is a potential

| Sample ID | pH  | EC (mS/m) | TDS (mg/L) | Turbidity (FTU) | BOD (mg/L) | COD (mg/L) | DO (mg/L) | DOC (mg/L) | SF$_{450/350}$ (RU) | SUVA$_{254}$ (L/mg C m) |
|-----------|-----|----------|------------|-----------------|------------|------------|-----------|------------|---------------------|------------------------|
| L-1       | 5.68| 3,360    | 16,000     | 794             | 460        | 690        | 1.9       | 1.6        | 0.23                | 0.93                   |
| L-2       | 7.94| 720      | 460        | 159             | 247        | 380        | 3.9       | 3.1        | 0.29                | 0.58                   |
| L-3       | 7.87| 690      | 380        | 127             | 247        | 380        | 3.9       | 3.1        | 0.09                | 1.46                   |

**Table 1.** Physicochemical and optical properties (mean value of five samples) of leachate and DOM samples.

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**Inland surface water discharge standard of Bangladesh (ECR 1997)**

| Standard* | EC 2100 | pH 7.0 | TDS 1000 | Turbidity 50 | BOD 20 | COD 100 | DO 8.0 | DOC 25 | SF 0.25 | SUVA > 0.45 |
|-----------|---------|--------|----------|--------------|-------|---------|-------|-------|---------|-------------|
| 6–9       | 120     | 10     | 200      | 10           | 10    | 50      | 50    | 50    | -       | -           |

*Inland surface water discharge standard of Bangladesh (ECR 1997).
pollutant. As the landfill site is flooded during every monsoon, there is a chance of surface water pollution from this site.

Mean values of DOC in DOM samples indicated very high organic carbon load in all the samples. Specific fluorescence intensities (SFI$_{350/450}$) of the samples were measured which were used as the initial fluorescence intensity ($F_0$) of the samples. The highest SFI$_{350/450}$ mean value of L-2 indicated that, L-2 contained the maximum number of fluorophores than L-1 and L-3 at Ex$_{350}$/Em$_{450}$ that was responsible for fluorescence intensity. SUVA$_{254}$ (specific UV absorbance at 254 nm) mean values of the samples L-1, L-2 and L-3 were 0.43, 0.58 and 1.46 L/mg C m, respectively. These relatively lower values of the SUVA$_{254}$ in all the samples indicated that the aromatic moieties in all the samples were hydrophilic (Ahmed et al. 2012). To identify the substances that are responsible for fluorescence properties of DOM, 3D EEMs were created and data were analyzed. For all EEMs, three fluorescence peaks were identified; protein like substances (e.g., tryptophan, tyrosine, etc.) detected at Ex/Em 250–300/290–370 nm, humic-like substances detected at Ex/Em 375–450/380–580 nm and fulvic acids detected at Ex/Em 300–400/380–480 nm. Similar types of peaks were identified in many other previous studies (Baker and Curry 2004; Henderson et al. 2009; Chen et al. 2003).

Functional groups of DOM

IR spectra of three different ages DOM in Fig. 1 represent the information about the structural changes of DOM in term of chemical bonds and groups over time. The FT-IR spectrum of L-1 (green line in Fig. 1) showed major signals at 3377, 2929 and 1593 cm$^{-1}$, which were related to hydrogen bonded alcohols, phenols of O–H, C–H stretches of alkanes and aromatic rings of C=C bonding, respectively (Shouliang et al. 2008; Li et al. 2014a). There were other signals at 1411, 927, 854 and 777 cm$^{-1}$ which attributed to the C–H bond of alkanes. Peak at 1319 cm$^{-1}$ was related to nitro compounds (NO$_2$). Peaks at 1280, 1265, 1122 and 1093 cm$^{-1}$ attributed to C–O stretching of alcohol, ethers, carboxylic acids and esters (Skoog et al. 2007). L-2 showed major peaks at 3466 and 3292 cm$^{-1}$ which corresponded to N–H stretching of amines and amides, and O–H stretching of hydrogen bonded alcohol and phenol, respectively (black line in Fig. 1). A signal at 2538 cm$^{-1}$ related to O–H stretching of hydrogen bonded carboxylic acids. Peak at 1649.14 cm$^{-1}$ indicated the presence of C=C bond of alkanes. C–H bond of alkanes and aromatic rings was found at 1446, 1462, 792.74, 848 and 879 cm$^{-1}$ (Skoog et al. 2007; Seo et al. 2007; Marley et al. 1996). A signal at 1186 cm$^{-1}$ indicated the C–N is stretching of amines and amides.

In L-3 (red line in Fig. 1) major signals were at 3466, 2929 and 2551 cm$^{-1}$ which are related to N–H stretching of amines and amides, C–H stretches of alkanes and O–H

![Fig. 1 The FT-IR spectra of DOM samples showing changing chemical bonds over time](image-url)
stretching of hydrogen bonded carboxylic acids, respectively (Skoog et al. 2007; Shouliang et al. 2008). C–H stretching of alkanes was also found at around 1446 cm$^{-1}$ (Marley et al. 1996). C–C bond of alkanes was found at 1647 and 1685 cm$^{-1}$. Peak at 1186 cm$^{-1}$ attributed to C–N stretching of amines and amides (Li et al. 2014a). C–O stretching of alcohol, ethers, carboxylic acids and esters was found at 1033 and 1076 cm$^{-1}$. C–H bond of alkanes was also found at 848 and 879 cm$^{-1}$. Overall spectra analysis showed an increasing number of amines and amides chemical bonds in DOM over time.

**Toxic metals (Ni, Pb and Hg) concentrations in DOM samples before adding metal salts**

As Ni, Pb and Hg were used as quenching titration metals, the initial concentrations of those metals in DOM samples were determined. Ni was found in all samples and the mean concentrations were 0.81, 0.11 and 0.09 mg/L in L-1, L-2 and L-3, respectively. On the other hand, mean concentrations of Pb were 0.86, 0.26 and 0.25 mg/L in L-1, L-2 and L-3. Hg concentration in L-1 was 0.04 mg/L, whereas L-2 and L-3 contained 0.23 and 0.24 mg/L, respectively. The sources of these metals in leachate samples might be the damage batteries, broken thermometers, electronic devices etc. in the solid waste in landfill site (EDSO 2010; Chamon et al. 2009). Pesticides also could be a source of metals in leachate that may come through the decomposition of discarded vegetables, fruits, etc. (Naser et al. 2009).

**Comparison of fluorescence spectra before and after complexation titration**

Fluorescence Spectra of DOM samples (L-1, L-2 and L-3) showed marked changes when they were titrated with metal ions. Complexation spectra of 20 μM metal-DOM solutions are shown in Fig. 2 due to the clear spectral visibility. Figure 2 shows the changes in fluorescence spectra of DOM samples before and after complexation titrations. This was the result of complexation of metal with fluorophores present in DOM which can alter the DOM fluorescence intensity (Mostofa et al. 2013). Decreasing fluorescence intensities (in RU) after addition of metals were observed for each sample. Before titration the excitation – emission maxima in L-1 was at 250/338 nm but after titration with metals, it was shifted from shorter to longer wavelengths (red shift). For Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$ the shifts were at 250–260/338–342 nm, 250–255/338–342 nm and 250–420/338–490 nm, respectively. L-2 showed red shift in interaction with Pb$^{2+}$ and it was at 365–375/454–456 nm. L-3 also exhibited the red shift with the interactions with these three metal ions. The excitation- emission maxima of L-3 was at 355/436 nm and it was shifted at 355/436–442 nm, 355/436–446 nm and 355–450/436–526 nm for Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$, respectively. These red shifts might be contributed by the formation of metal complexes with fluorophores or functional groups (Mostofa et al. 2013) that also resulted in reduced fluorescence intensities.

On the other hand, L-2 showed blue shift (shift of excitation-emission wavelength maxima from longer to shorter wavelength regions) in interaction with Ni$^{2+}$ and Hg$^{2+}$. The excitation- emission maxima of L-2 were at 365/454 nm but they were shifted at 365–335/454–438 and 365–325/454–406 nm for Ni$^{2+}$ and Hg$^{2+}$, respectively (Fig. 2). This type of shift could be the results of the loss of fluorophores by precipitation, aggregation or flocculation as a consequence of physicochemical alteration of them caused by increased ionic strength (Sholkovitz 1976; Carlson and Mayer 1983; McCarthy et al. 1998). Similar kinds of spectral shifts were also found in complexation titrations of 1, 5, 10, 15 μM metal-DOM solutions but at slightly different wavelengths.

**Toxic metal binding affinity of DOM**

The fluorescence spectral patterns of all the samples were similar and all showed a decrease in fluorescence intensities as Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$ were added (Fig. 2). This was consistent with fluorescence quenching of DOM by Hg$^{2+}$, Cu$^{2+}$ and Pb$^{2+}$ in previous studies (Lu and Jaffe 2001; Saar and Weber 1980). Good linear correlations between $F_f/F_0 = F$ and $1/[M]$ were observed (indicated by $R^2$ values in Table 2) and the titration data were fitted to the modified Stern–Volmer equation to calculate Log$K$ and %f. Calculated binding parameters are presented in Table 2.

In Fig. 3a, the highest values in L-2 with all the metals indicated that suitable condition for DOM complexation by metal ions (Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$). Functional groups such as phenols (–OH) and amines (–NH$_2$) may play an important role in the complexation, which was similar to the study conducted Smith et al. (2002). Lower Log$K$ value in L-1 may due to the very unstable and changing composition and structure in DOM as well as lack of amines functional group. As L-1 was a very fresh sample, different physico-chemical reactions and biological decompositions might be occurred until a certain level of maturity which took several times that might curtail the stability of the DOM-metal complexes.

L-2 showed (Fig. 3b) the maximum binding capacity with Ni$^{2+}$, Pb$^{2+}$ and Hg$^{2+}$ and the values were 80.02, 83.63 and 68.97%, respectively, indicating higher number of ligands that interacted with metal ions. Similar type of result was found by Li et al. (2014b) in interaction between DOM and copper (II). Despite of higher SFI$_{350/450}$ than L-3, L-1 exhibited lowest %f with Ni$^{2+}$ and Pb$^{2+}$ (34.33% for Ni$^{2+}$ and 32.95% for Pb$^{2+}$). This might be due to the presence of high concentration of Ni and Pb.
(0.81 and 0.86 mg/L, respectively) in raw sample which already occupied the some binding sites of DOM and also due to the absence of adequate functional groups (C–N, N–H) that could act as metal binding sites. On the other hand, Hg\(^{2+}\) showed strong binding affinity to fresh DOM (L-1) and binding affinity decreased as the age of leachate increased. Pb\(^{2+}\) binding capacity of L-3 was higher than Ni\(^{2+}\) and Hg\(^{2+}\).

**Table 2** DOM-Ni\(^{2+}\), DOM-Pb\(^{2+}\) and DOM-Hg\(^{2+}\) binding parameters

| Sample | Ni\(^{2+}\) LogK | f (%) | R\(^2\) | Pb\(^{2+}\) LogK | f (%) | R\(^2\) | Hg\(^{2+}\) LogK | f (%) | R\(^2\) |
|--------|------------------|-------|--------|------------------|-------|--------|------------------|-------|--------|
| L-1    | 3.62             | 34.33 | 0.580  | 5.94             | 32.95 | 0.973  | 6.27             | 58.62 | 0.968  |
| L-2    | 6.97             | 80.02 | 0.970  | 6.73             | 83.63 | 0.987  | 6.73             | 68.97 | 0.923  |
| L-3    | 6.31             | 49.36 | 0.916  | 5.49             | 76.22 | 0.974  | 6.18             | 27.40 | 0.898  |

**Conclusion**

The physico-chemical properties and Ni\(^{2+}\), Pb\(^{2+}\), Hg\(^{2+}\) binding ability of leachate-derived DOM were investigated by spectroscopic techniques. The results suggested that leachate could be a potential pollutant in term of high organic load and biological pollution. Clear fluorescence...
spectral shifts were observed due to the addition of metal ions. Results also highlighted the higher %f and LogK of young DOM (L-2) sample indicating highest toxic metal binding affinity. On the other hand, binding affinity of Pb²⁺ was higher than the other metals (Ni²⁺ and Hg²⁺) with L-2. Binding affinity of DOM with metals showed the following pattern-Ni²⁺ binding affinity: L-2 > L-3 > L-1, Pb²⁺ binding affinity: L-2 > L-3 > L-1 and Hg²⁺ binding affinity: L-2 > L-1 > L-3. Overall, DOM in leachate samples showed variations in metal complexing capacity depending on DOM composition, structure and initial metal concentrations. As metal – DOM complexation plays a vital role in the toxicity and bioavailability of heavy metals in the environment, dissolved organic load of leachate should be treated effectively before discharge to avoid the potential threat of organic and heavy metal pollution on surrounding soil and aquatic environment.

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