ORIGINAL ARTICLE

Phytotoxicity of leachate from closed sanitary landfill on Mung Bean Seed *(Vigna radiata)*

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Abstract:

Decomposing waste at landfills will produce a highly polluted liquid called leachate which can pose a great risk to the environment and potentially be the source of soil, groundwater and water pollution. Therefore, in order to prevent the continual toxicity of landfill leachate, toxicity assessment of potential impact of leachate towards plants is a legal necessity in some nations. In this study, leachate was characterized in addition to phytotoxicity assessment using *Vigna radiata* seeds as an indicator. The test concentration of leachate used were 0% (control group) 3%, 5%, 10%, 25%, 50%, 75% and 100%. Seed germination and root elongation were measured and calculated after seven days exposure. All parameters were found to be within the standards limits after treatment however concentrations of zinc, iron, ammoniacal nitrogen and cyanide were observed to increase after the leachate treatment process. Germination rate, average root length and relative growth rate were observed to decrease with the increase in leachate concentration although all parameters measured were within standard limits.

Keywords: Landfill leachate,physicochemical, phytotoxicity, seed germination,

1. INTRODUCTION

Leachate production is a major problem for municipal solid waste landfills and is known to contain numerous contaminants such as organic matter, heavy metals, chemicals and nitrogen. The mass flux of transported contaminants has the potential to enter the human food chain through vegetation and aquatic sources [1-2]. In Malaysia, more than 10.4 million tons of municipal solid waste is disposed into landfill sites annually [3], and as a result, vast amounts of leachate are generated at a landfill as a product of waste decomposition.

The assessment of toxic effects of leachates using rapid, sensitive and cost-effective biological assays is more useful in assessing the risks as they measure the overall toxicity of the chemicals in the leachate [4]. The determination of landfill leachate toxicity using plants is known as phytotoxicity assessment and measures the delay of seed germination, inhibition of plant growth or any adverse effect on plants caused by specific substances called phytotoxins or growing conditions [5]. The usage of plants as a medium is suitable compared to other organisms as plants could be more sensitive towards a wide range of pollutants in addition to being simple, fast, reliable and cost effective instead of using aquatic animals [6].

Therefore, in order to reduce or prevent the effects and continual toxicity of landfill leachate, phytotoxicity assessment on potential impact of leachate towards specific plants has been made a legal necessity in some nations [7]. Even after a landfill site is closed, a landfill will continue to produce contaminated leachate and this process could last for 30-50 years [8]. In this study, the toxicity assessment is focused on plant bioassays of municipal landfill leachate from a closed operation landfill.

2. MATERIALS AND METHODS

Raw and treated leachate samples were collected from Air Hitam Sanitary Landfill located in Puchong, Selangor. The landfill received domestic waste from 1st April 1995 until 31st December 2006 and is currently undergoing post closure maintenance activity. All samples were collected using grab sampling method, preserved with either ice at 4°C, sulphuric acid or nitric acid to prevent chemical degradation and transported to the laboratory immediately. Samples that were not analyzed within one week were frozen at -80ºC [9]. pH, temperature, dissolved oxygen and turbidity were analyzed *in situ* whereas total suspended solid (TSS), nitrate, cyanide, ammoniacal nitrogen, chemical oxygen demand

(COD), biochemical oxygen demand (BOD), chromium, manganese, iron, zinc and nickel were analyzed in the laboratory [10-11].

Phytotoxicity bioassay was determined using *Vigna radiata* seeds also known as mung bean. Seeds were immersed in 200 mL deionized water to test their ability to sink. Seeds were soaked in 20 mL of the test solution for approximately 2 hours. Concentrations of test solutions used were 0% (control), 3%, 5%, 10%, 25%, 50%, 75% and 100% v/v of treated leachate [12]. Ten plump undamaged seeds of almost identical size were laid on two layers of filter paper (125 mm in diameter, Whatman No. 1) in a clean petri dish (20 mm x 120 mm); each dish contained 10 mL of test solution and all test concentrations were replicated. Dishes were incubated in the dark, at 24ºC (±2ºC) for seven days. Seed germination and root elongation were measured at the end of seven days.

The germination percentage and relative root growth were calculated using the following formulas:

Germination Percentage = (No of seeds germinated/ total seeds) X 100%

Growth inhibition = (average root length in sample/average root length in control) X 100%

3. RESULTS AND DISCUSSION

Traditionally, the assessment of hazards from landfill leachate is based on chemicals analysis and evaluation to determine the chemical composition. However, the toxic effects cannot be evaluated if only compositions data is obtained as the chemical compositions and toxicity level are integrated with each other [13]. Therefore, in this study the leachate characterizations to assess the toxicity level of leachate and physicochemical characteristics was integrated with phytotoxicity bioassay use *Vigna radiata*.

3.1. Leachate characterization

Leachate as summarize in Table 1, samples were collected and analyzed for various physicochemical parameters to estimate its pollution potential. The leachate from the Air Hitam Sanitary Landfill is characterized as "old" leachate in the methanogenic phase based on the concentrations of heavy metals, ammoniacal nitrogen, COD and pH [14]. It has been found that the treated leachate samples contain low concentrations of organic and inorganic constituents all within the permissible limits set under Environmental Quality (Control of Pollution from Solid Waste Transfer Station and Landfill) Regulations 2009 [15] except for ammoniacal nitrogen as summarized in Table 1.

The concentrations of heavy metals in the leachate did not show a significant trend or pattern. Nickel, chromium and manganese had lower concentrations after treatment however concentrations of zinc and iron increased after the leachate treatment process. In general, the strength of leachate decreases with time due to biological breakdown of organic compounds and precipitation of soluble elements such as heavy metals. Generally, heavy metals are substantially higher in younger leachate under acidic conditions and lower in older landfills and aged leachate under methanogenic phase [16].

Table 1. Physicochemical characteristics of landfill leachate

Parameter	Raw Leachate	Treated Leachate	Standard Limit
pH	7.84	6.43	6.0 – 9.0
Temperature (ºC)	24.7	24.2	40
Colour (Pt Co)	1964	498	100 ADMI
DO (mg/L)	0.0	9.24	N/A
Turbidity (NTU)	245.67	0.0	N/A
BOD_5 (mg/L)	60.75	5.07	20
COD (mg/L)	163.67	22.67	400
TSS (mg/L)	211	11.3	50
Nitrate (mg/L)	4.6	3.5	N/A
Ammoniacal nitrogen (mg/L)	0.256	14.6	5
Cyanide (mg/L)	0.003	0.004	0.05
Nickel (mg/L)	0.167	0.034	0.2
Chromium (mg/L)	0.03	0.026	0.05
Manganese (mg/L)	0.596	0.179	0.2
Iron (mg/L)	0.56	0.86	5.0
Zinc (mg/L)	0.04	0.23	2.0

There was a marked reduction in the values for color, turbidity, BOD, COD and TSS due to the aeration treatment process implemented. Consecutively this contributes to the process of conversion from acidogenic to methanogenic phase [9].

In contrast, the concentration of ammoniacal nitrogen increased significantly after treatment and was way over the allowable limit. The increasing value of ammoniacal nitrogen is due to the contribution of nitrogen fixation in the ammonification process where oxygen was introduced during aeration in the leachate pond [17].

3.2. Phytotoxicity assessment

The germination rate of *V. radiata* showed only slight reduction as the test solutions of treated leachate increased. It was observed that at 3%, 5%, 10% and 25% treated leachate solutions, all seeds in each petri dish germinated. While, seeds exposed to 50%, 75% and 100% treated leachate solutions germinated at 90%, 70% and 70% respectively. The germination rate of *V. radiata* exposed to raw leachate declined starting at 5% test solution and significantly reduced to 0% germination for test solution of 100% raw leachate. There was a significant difference in the germination rate of seeds for exposure to raw and treated leachate (Figure 1).

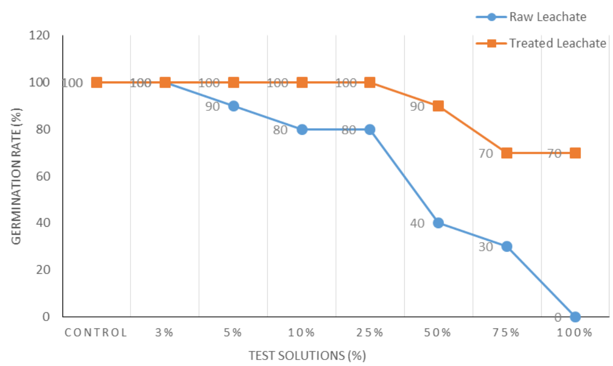

Figure 1. Germination rate of *V. radiata* exposed to raw and treated leachate

Root elongation results of *Vigna radiata* exposed to different concentrations of raw and treated leachate is shown in Figure 2. The root elongation lengths exhibit a decline trend with increase in test solution concentrations for both raw and treated leachate. The control seeds grew an average of 10.3 cm. Seeds exposed to leachate samples started to show a reduction in root length after exposure to the lowest concentration (3%). Seeds exposed to 100 % test solution grew an average of 2.58 cm for treated leachate, whereas seeds exposed to 100% raw leachate did not have any root length as all seeds did not germinate. There was a significant difference between root length of seeds exposed to treated and raw leachate ($p < 0.05$). Additionally, there was also significant differences between root lengths for varying concentrations of test solutions within the raw and treated exposure groups.

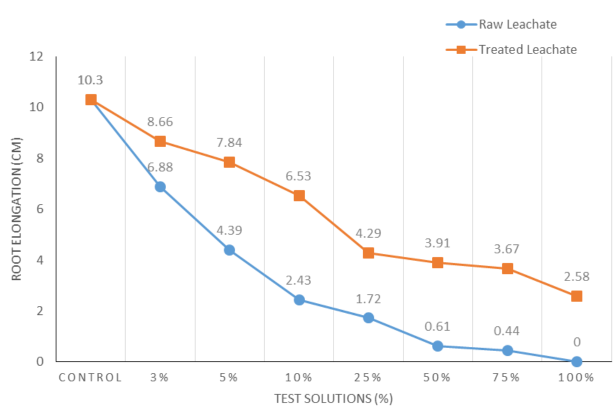

Figure 2. Root elongation of *V. radiata* exposed to raw and treated leachate

Growth inhibition of *Vigna radiata* after exposure to different concentrations of treated and raw leachate is shown in Table 2. The growth inhibition exhibited an increasing trend as the exposure concentrations increased. Both raw and treated leachate exposure showed an increase in growth inhibition after exposure to the lowest concentration (3%). Inhibition of 50% growth rate was observed with 5% raw leachate exposure and 25% treated leachate exposure. Treated leachate exposure showed less growth inhibition compared to raw leachate.

In preceding studies, seed germination was believed to be less sensitive in evaluating the toxicity of leachate [18], suggesting that root growth was a more reliable parameter with higher sensitivity in the assessment of toxicity using plant species [13]. As supported by this study, although all seeds in the treated exposure groups (3%, 5%, 10% and 25%) germinated, root growth was inhibited at 16%, 24%, 37% and 58% respectively. Previous studies have also reported root growth inhibition in *Lactuca sativa* (maximum 80%), *Cannabis sativa. L* (89.8%) and *Sinapis alba. L* (100%) [19]. Toxicity response for each plant species are varied and may possibly be due to the differences in species' attributes suchs as tissue permeability, differential uptakes and metabolism [20].

Table 2. Growth inhibition of *V. radiata* exposed to raw and treated leachate

Concentration (%)	Growth Inhibition (%)	
	Raw Leachate	Treated Leachate
0 (Control)	100	100
3	33	16
5	57	24
10	76	37
25	83	58
50	94	62
75	96	64
100	100	75

Leachate quality is an imperative factor in toxicity as low concentrations may stimulate germination, initial growth and cell division while growth inhibition is observed at higher concentrations [4]. Although findings in previous studies state that most metals could be the inhibitory factors of plant development and pose a threat to the ecosystem, copper, zinc and nickel are necessary micronutrients for plants in low concentrations but are noxious to organisms at high concentrations [21]. Additionally, zinc was found to not significantly affect the seed germination [22]. Hence, in this study we believe that mung bean seeds *V. radiata* exposed to treated leachate may have obtained a suitable micronutrient content compared to raw leachate where better seed germination and growth were seen.

Treated leachate has a better quality in terms of an improved composition, however efficacy of treatment technology should not only measure the physicochemical parameters against the standards requirements but also consider the environmental conditions on the dynamics of toxicity processes that may take place.

4. CONCLUSION

Although the quality of the aged leachate from the closed sanitary landfill was within the standards and under the methanogenic phase, seed germination rate and root elongation was significantly affected when seeds were exposed to higher concentrations of test solutions. This shows that the leachate still had an effect on plant growth after treatment. Analytical techniques can elucidate the chemical characteristics of an environmental sample, but give little information regarding its potential ecological effects. In Malaysia, no guidelines or standard procedures exist on phytotoxicity testing of landfill leachate and this study recommends that a new guideline be designed.

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