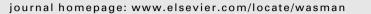
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Leachate pollution index as an effective tool in determining the phytotoxicity of municipal solid waste leachate

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ABSTRACT

Phytoremediation is a promising option for the treatment of municipal solid waste leachate. Combining the leachate pollution index with the phytotoxicity data will be useful in predicting the suitable concentration of leachate for the phytoremediation applications. Understanding the tolerant mechanisms of plants to leachate stress will further help to select the appropriate dose. The aim of the study was to investigate the effect of different concentrations of leachate on germination, growth, chlorophyll content and antioxidant enzyme activities in the plant *Vigna unguiculata*. The crude leachate has an LPI value of 31.99 with high concentration of organic matter, ammonia and dissolved solids. The results of the phytotoxicity study suggest that at lower concentrations the leachate enhanced the germination and promoted plant growth. Up to 5% concentration (v/v) of the leachate which had a LPI value of 11.84 the growth promotion was observed in *V. unguiculata*. This was made possible by the controlled modulation of reactive oxygen species through the enhanced antioxidant enzyme activities in leachate disrupt the enzyme activities and leads to the peroxidation of membrane lipids and significantly affected the plant growth. The study suggest that phytotoxic effects in plants are directly related to the LPI value and leachate with LPI values less than 10 are likely to promote plant growth and LPI values greater than 10 are likely to exert detrimental effect on the plant.

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1. Introduction

The proper management of municipal solid waste (MSW) continues to be one of the most challenging environmental issues in the world. Landfilling and open dumping are the widely used management practices in developing countries waste (Mangimbulude et al., 2009; Tränkler et al., 2005). Considering the high percentage of biodegradable constituents in the MSW and the environmental issues caused by the landfills/dumping yards, increasing efforts are made to divert the organic fraction of MSW (OFMSW) from landfills and treat it through other feasible techniques such as composting, gasification, incineration etc. This approach reduces the burden on existing waste disposal facilities, cost of waste disposal and the environmental impacts caused by the organic waste in landfills (Mokhtarani et al., 2012). Among the various treatment techniques, composting has the distinct advantage of low investment, ease of operation and requirement of less technical expertise. Moreover it produces compost - an organic fertilizer - as an end product. However the problem of

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http://dx.doi.org/10.1016/j.wasman.2017.07.012 0956-053X/© 2017 Published by Elsevier Ltd. leachate arising from these plants still persists and has to be treated properly before discharging into the environment.

Leachate produced during the decomposition of organic fraction of municipal solid waste (OFMSW) is a complex mixture of various pollutants such as organic compounds, inorganic salts, nutrients and heavy metals. The quality of leachate depends on various factors such as the composition of waste, biological and chemical processes occur during the degradation of waste, moisture content, rainfall, local climate, etc., thus it is site specific (Jones et al., 2006; Kumar and Alappat, 2005a). The leachate thus generated is considered as a major pollution threat to the surface and groundwater, soil, environment and human health. In many of the developing countries, the contamination issue is more serious since most of the municipal solid waste (MSW) management facilities and landfills do not have leachate collection and treatment systems. Moreover, in many cases, the MSW management facilities are located near to human settlements resulting into a number of leachate contamination of groundwater incidents in the past (Deshmukh and Aher, 2016; Fernández et al., 2014; Mor et al., 2006; Singh et al., 2016).

Several methods have been developed to treat landfill leachate. However, the inconsistency of leachate flow and characteristics have resulted into the failure of many treatment systems. In recent

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years phytoremediation has emerged as a low cost and environmentally sound technique for the onsite treatment of leachate. It is a versatile technology which can be utilized in several configurations, such as constructed wetlands (CW), nutrient film technique (NFT), or irrigation of vegetated land for the production of biomass, horticultural and energy crops (Abbasi and Ramasamy, 1999; Ignatius et al., 2014; Arunbabu et al., 2015). Under ideal conditions phytoremediation of leachate (raw or partly treated) provide an opportunity for closing the nutrient cycle loop and simultaneously reduce the risk of environmental pollution (Jones et al., 2006). However, the failure in many cases can be ascribed to excessive leachate application and poor management due to a fundamental lack of understanding of the plant soil system (Jones et al., 2006). Therefore the success of phytoremediation is critically dependent on whether the plants can tolerate or avoid the stress induced by leachate and maintaining the leachate concentration within the toxic threshold of the plants (Kalčíková et al., 2012).

Even though several studies reported the growth and chlorophyll inhibition in plants exposed to leachate, the information on oxidative stress and antioxidant status of plants treated with leachate are limited (Gupta and Rajamani, 2015; Sang et al., 2010). In this context, it is critical to understand the responses of plant systems to landfill leachate stress, especially the tolerant mechanisms of plant systems (Sang et al., 2010). Various studies have shown that leguminous crops are very responsive to leachate in terms of growth and seed germination (Gupta and Rajamani, 2015). Therefore, in the present study, the plant Vigna unguiculata which belongs to the legume family was selected to study the toxicity of leachate. To get more information on plant response to leachate, in our present study, the effect of leachate on germination, growth, chlorophyll content, lipid peroxidation, activities of superoxide dismutase (SOD), catalase (CAT), and peroxidase (POX) were analyzed under various leachate concentrations.

It has been reported in the literature that the various pollutants in the leachate exhibit synergistic, antagonistic and additive effects on the plants hence chemical analysis alone may not be sufficient to determine the toxicity of leachate. However, (Kumar and Alappat, 2005a) reported an index known as leachate pollution index (LPI) to quantify the leachate contamination potential of different landfills on a comparative scale. The LPI value represents the composite concentration of 18 parameters, including pH, total dissolved solids, biological and chemical oxygen demand, nitrogen, heavy metals (Fe, Cu, Ni, Zn, Pb, Cr, Hg and As), phenolic compounds, chloride, cyanide, and total coliform bacteria and provides a better insight on the strength of various pollutants and helps to report the leachate pollution in a quantitative manner (Kumar and Alappat, 2005b). Therefore the present study analyzed the relationship of various stress parameters of the plant with the calculated LPI value. Combining the LPI value with the phytotoxicity data will be useful in predicting the suitable concentration of leachate for the phytoremediation applications.

2. Materials and methods

2.1. Leachate collection and characterization

The present study was carried out using the leachate collected from the MSW management facility at Brahmapuram (Kochi), Kerala, India. The plant receives an average of 220 tonnes of biodegradable waste on a daily basis and aerobic composting method was used to treat the waste (Hridya et al., 2016). The plant doesn't have any facilities to treat the leachate produced during the degradation process. Fresh leachate from the composting plant was collected in November 2015 and immediately transported to the laboratory in ice boxes and allowed to settle for 12 h in the lab, the supernatant was used for experiments. The physicochemical properties of the leachate were analyzed according to standard methods (APHA, 1998). The pH, electrical conductivity and total dissolved solids were measured using respective electrodes (Systronics, India). The chemical oxygen demand (COD) and biological oxygen demand (BOD₅) were determined using dichromate oxidation method and 5-day BOD test respectively. Nutrients like phosphate and nitrate were analyzed colorimetrically following stannous chloride and brucine method respectively using a spectrophotometer (UVmini -1240, Shimadzu). Total Kjeldahl nitrogen and ammonia nitrogen were determined using a semi-automatic nitrogen analyzer (KEL Plus–Elite EX (VA), Pelican Equipments, India). The samples for TKN were digested in the Kjeldahl automatic sample digestion system (Pelican equipments, India) at 450 °C for 2 h before analysis.

The heavy metals in the leachate were analyzed using ICP MS (iCAP Qc, Thermo Scientific, USA) after microwave digestion of the samples with HNO₃. Samples were introduced into the ICP MS with the help of an auto sampler ASX520 (Cetac Technologies Inc., USA). The sample introduction system consists of a standard peltier cooled quartz cyclonic spray chamber, PFA concentric nebulizer and demountable quartz torch with a 2.5 mm ID quartz injector. Standard Ni sample and skimmer cones were also used. The instrument was operated in a single collision cell mode with kinetic energy discrimination (KED), using pure He as the collision gas. The instrument was calibrated using a series of multi-element standards before performing the analysis.

2.2. LPI calculation

LPI was calculated according to the procedure given by Kumar and Alappat (2005a). Out of the 18 parameters used for the calculation of LPI, phenolic compounds, cyanide and total coliform were not analyzed in the present study. Therefore the modified equation as described in Kumar and Alappat (2005a) was used to calculate the LPI.

$$LPI = \frac{\sum_{i=1}^{m} wipi}{\sum_{i=1}^{m} wi}$$

where

LPI = Leachate pollution index,

wi = Weight of the ith pollutant variable,

pi = Sub index score of the ith leachate pollutant variable,

m = Number of leachate pollutant variables used in calculating LPI.

2.3. Plant material

Cowpea (*Vigna unguiculata* (L.) Walp.), supplied by Kerala Agricultural University, Kerala, India was used as the test plant for the present study. It is a herbaceous annual plant with twining stem and trifoliate leaves and belongs to the Leguminosae family.

2.4. Germination study

The germination study was conducted in pre-cleaned Petri dishes lined with two layers of Whatman No. 42 filter paper. The filter papers were moistened with 5 ml of different concentrations of leachate (0%, 0.5%, 1%, 2%, 5%, 10%, 25%, 50% and 100% v/v). Dry seeds of uniform size and weight soaked in distilled water for overnight were placed in the Petri dishes containing leachate. Each Petri dish had 10 seeds and each treatment was repeated 5 times. The experiment was conducted at room temperature and the number of seeds germinated was recorded. The Emergence of radi

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