

# Removal of COD, ammoniacal nitrogen and colour from stabilized landfill leachate by anaerobic organism

Mohamad Anuar Kamaruddin · Mohd Suffian Yusoff ·  
Hamidi Abdul Aziz · Nur Khairiyah Basri

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**Abstract** This study was conducted to investigate the treatment performance of anaerobic organism for stabilized leachate under ambient air condition. The treatability of landfill leachate was tested under various influences including pH, dosage of anaerobic organism and contact time. Laboratory experiment revealed that anaerobic organism was in progressive state when the leachate in neutral condition. At this phase, the removal efficiency of chemical oxygen demand (COD), ammoniacal nitrogen ( $\text{NH}_3\text{-N}$ ) and colour of 76.84, 59.44 and 46.2 %, were experimentally attained. The experimental results also showed that the optimum variables condition was established at pH 7 of leachate sample, dosage of 100 mL of anaerobic organism and required 14 days to reduce the COD,  $\text{NH}_3\text{-N}$  and colour pollutants to 65.5, 60.2 and 46.3 %, respectively. The preliminary investigation shows that the anaerobic organisms were able to remediate leachate pollutants by the microbial activity in the leachate sample under ambient air condition.

**Keywords** Landfill leachate · Anaerobic organism · COD ·  $\text{NH}_3\text{-N}$  · Colour

## Introduction

Landfilling is the primary means of municipal solid waste disposal in many countries worldwide owing to its economic advantages and minimum technology being practiced. Contamination of surface and ground water through

leachate; soil contamination through direct waste contact or leachate; air pollution through the burning of wastes and the uncontrolled release of methane by anaerobic decomposition of waste (Aziz et al. 2010) are some of the effects of landfilling activities. In addition, landfill leachate produced from landfill sites consists of contaminated pollutants that is very difficult to deal with (Umar et al. 2010). If not properly treated and safely disposed, leachate can migrate to soil and subsoils which might cause severe damage to eco-system of land and receiving water. Landfill leachate contains pollutants that can be categorized into four groups [dissolved organic matter, inorganic macro-components, heavy metals, and xenobiotic organic compounds (Kjeldsen et al. 2002)] as consequence of the existing waste disposal practice. Organic content of leachate pollution is generally measured by chemical oxygen demand (COD) and biochemical oxygen demand ( $\text{BOD}_5$ ). Leachate is also rich in ammonia, halogenated hydrocarbons suspended solid, significant concentration of heavy metals and inorganic salts (Renou et al. 2008; Umar et al. 2010; Bashir et al. 2009; Aziz et al. 2010). According to Umar et al. (2010) for stabilized leachate, the COD content generally ranges between 5,000 and 20,000 mg/L. The  $\text{BOD}_5/\text{COD}$  ratio provides a good estimate of the state of the leachate and this ration for young leachate is generally between 0.4 and 0.5 (Kurniawan et al. 2006). The presence of concentrated pollutants in leachate has become one of the major issues to the landfill operator in obeying strict regulations for the safe disposal of leachate. Normally, the existence of high levels of pollutants such as ammonia in landfill leachate over a long period of time leads to a motivated algal growth, decreased performance of biological treatment systems, accelerated eutrophication, promoted dissolved oxygen depletion, and increased toxicity of living organisms in water bodies (Aziz et al. 2010).

M. A. Kamaruddin · M. S. Yusoff (✉) ·  
H. A. Aziz · N. K. Basri  
School of Civil Engineering, Universiti Sains Malaysia,  
14300 Nibong Tebal, Pulau Penang, Malaysia  
e-mail: suffian@eng.usm.my

Various process applications such as aerobic and anaerobic biological degradation, chemical oxidation, chemical precipitation, coagulation–flocculation, activated carbon adsorption and membrane processes have been reported by various authors (Aziz et al. 2010; Halim et al. 2009; Aghamohammadi et al. 2006; Deng and Englehardt 2006). Anaerobic treatment is one of the treatments in biological process which is ideally fitted for the pre-treatment of high strength wastewaters that are typical of many industrial facilities for the removal of the bulk of effluent containing high concentrations of COD, BOD and heavy metals. In favour of anaerobic treatment, biodegradation is carried out by the microorganisms, which can degrade organic compounds to carbon dioxide and sludge under aerobic conditions and to biogas (a mixture comprising chiefly  $\text{CO}_2$  and  $\text{CH}_4$ ) under anaerobic conditions (Ghasimi et al. 2010). Effective microorganism (EM) technology identified as one of the biological treatment that comprises the most effective application for heavy metal removal in industrial wastewaters (Zhou et al. 2008). Historically, EM was first discovered by Teruo Higa of the University of the Ryukyus in Japan (Higa and Parr 1994). Initially, Higa and Parr investigated the microorganisms that were useful in crop farming as they were seen as alternatives to the use of pesticides and fertilizers (Zhou et al. 2008). However, as far as the authors concern, technology of EM or anaerobic organism is mostly being used in pig, cattle, dairy, and poultry farming for purification of the wastewater (Higa and Parr 1994; Joo et al. 2006). The main activity in the anaerobic process is the creation of an antioxidant environment by anaerobic organisms, which is deemed crucial to the enhancement of the solid–liquid separation in cleaning water practice. However, its usage and the effectiveness on industrial wastewater such as leachate is very far limited and documented. Therefore, this study was carried out to determine the feasibility of anaerobic organism inoculation in biological treatment of stabilized landfill leachate. The main purpose of the existing research is to investigate the effect of leachate parameters removal namely COD,  $\text{NH}_3\text{-N}$  and colour by anaerobic organism cultures. The efficiencies of COD,  $\text{NH}_3\text{-N}$  and colour removal were tested via different pH of leachate sample, dosage and contact time of anaerobic organism via laboratory experimental set up.

## Materials and methods

### Anaerobic organism preparation

Anaerobic organism solution was procured from Seberang Perai Municipal Council (Malaysia). The preliminary experimental work for obtaining the optimum ratio of

brown sugar/fruit waste/distilled water conducted towards the removal of COD,  $\text{NH}_3\text{-N}$  and colour. The livelihood of the anaerobic organisms was also tested at the longest possible of preliminary experiment so that the microbial activity in the leachate sample lasts longer. To prepare anaerobic organism activated solution (AOAS), a ratio of 1:3:10 which was: 1 part of molasses/brown sugar; 3 parts of fruit waste; and 10 parts of distilled water were used. The mixture was amalgamated in a Teflon container and mixed thoroughly. The AOAS was fermented for 12 weeks. The survival of the microorganisms was largely dependent by the temperature of the environment. Therefore, the prepared mixtures were kept under minimum temperature fluctuations without direct sun light exposure. The off-gassing process occurred during the 7 days of fermenting with the released of carbon dioxide ( $\text{CO}_2$ ). The entire process of fermentation was strictly monitored. In addition, the cover of the tank was ensured tightly closed to avoid generation of mold. The pH value was observed at the final duration of fermentation. A pH value in the range of 3–4 indicated that the fermentation process has completed. Next, the anaerobic organisms were kept in an airtight container to keep it anaerobic prior to use. Fundamentally, the activated anaerobic organism suspension is a mixture of molasses (brown sugar) and anaerobic organism in free chlorinated water or rice rinse water which provides the minerals for the multiplication of the microorganisms (Mohamad Yatim et al. 2009). The activated anaerobic organisms were then fermented in an anaerobic environment for 7–10 days of incubation. Prior to the experimental works, each of the containers was rinsed with 10 % (v/v) HCL and then deionised with water to remove background effects of the medium.

### Identification of anaerobic organism

Identification of anaerobic organisms was carried out according to the manual (Gerhardt 1994) and the procedure recommended by effective microorganism (EM) Research Organization (Okinawa, Japan). Theoretically, anaerobic organisms are rich with selected species of microorganisms, which can be commonly found in many ecosystems, including lactic acid bacteria (LAB), yeasts, actinomycetes photosynthetic bacteria, and other types of organisms. The analyses were done by diluting fermented materials in phosphate buffer (pH 7.0) at appropriate levels and then spread-plated onto an appropriate agar media and incubated at 25–30 °C at least 1 week prior to observing the morphological and physiological characteristics. Initial laboratory investigation indicated that microbial groups of lactic acid bacteria, yeast and total bacteria count of  $1.6 \times 10^7$ ,  $1.0 \times 10^4$  and  $1.2 \times 10^7$  cfu/mL, respectively, exists in the solutions.

**Table 1** Solid waste characteristics at PBLs

Waste characteristics	Amount (%)
Food	40
Plastic	22
Paper	10.5
Metals	2.5
Glass	3.25
Textile	3.5
Others	18.25
Total	100

### Leachate sampling

The leachate used in the experiment was collected from Pulau Burung Landfill Site (PBLs), which is situated in the North West of Malaysia. PBLs started its operation in 1991 as a semi-aerobic system complying with Level II sanitary landfill standards by establishing a controlled tipping technique. The site receives 1,800 tonnes of domestic waste daily which origins from Penang Island and Perai district. Table 1 shows the characteristics of the solid wastes at PBLs. In 2001, the landfill was upgraded to a Level III sanitary landfill by employing controlled tipping with leachate recirculation (Aziz et al. 2010). The site has a natural marine clay liner with daily covers practices. Characteristically, PBLs has surpassed more than 20 years of operation which in the methanogenic phase, and the leachate produced is referred to as mature, “stabilized” leachate (Bashir et al. 2009). The dark-coloured liquid, with high concentrations of COD and ammonium, and a low BOD<sub>5</sub>/COD ratio is another indicator of stabilized leachate. The leachate sample was collected from September 2010 until March 2011. The samples were transported to the laboratory, and stored in a cold room at 4 °C to minimize biological and chemical changes prior to the experimental use (Bashir et al. 2010). The sampling procedure and preservation of samples were done according to the Standard Methods for the Examination of Water and Wastewater (APHA 2005). pH of the leachate sample was

measured on-site using portable pH meter (Hach, sens ion 1, USA). The characteristics of the raw leachate from the oxidation pond of PBLs are shown in Table 2.

### Experimental procedure

The study was focused on the preliminary determination of the condition that provides the optimum condition of anaerobic organisms in treating leachate. Prior to the experimental works, each of the containers was rinsed with 10 % (v/v) HCL and then deionised with water to remove the background effects of the medium. Three different aspects of the experimental procedure were as follows:

#### *Influence of pH*

To investigate the influence of pH by anaerobic organism in the leachate sample, six containers of 20-cm length and 10-cm height with a total volume of 4,000 mL made of Plexi glass were used. The containers were filled with 1,500 mL raw leachate sample and pH was adjusted from 5 to 9 for each of the containers by either 0.1 N hydrochloric acid (HCl) or 0.1 N sodium hydroxide (NaOH). Then, anaerobic organisms were added to leachate at ratio of 1:10 (150 mL of anaerobic organism) using measuring cylinder. The solution was stirred with a glass rod thoroughly to avoid chemical reaction with the leachate sample and the anaerobic organism activities in the container. A net grating was installed on the top of each container to allow anaerobic activity with the surrounding environment throughout the experimental period. The containers were kept for 24 days to ensure complete reaction between leachate sample and anaerobic organisms and the percentage removal of COD, NH<sub>3</sub>-N and colour were eventually measured.

#### *Influence of dosage of anaerobic organism*

Different dosage of anaerobic organisms at optimum pH was mixed in five containers each with the exact volume of

**Table 2** Raw leachate characteristics

Parameters	1	2	3	4	5	6	Average	DOE standard
pH	7.81	7.67	7.34	7.65	7.12	7.51	7.52	5.5–9.0
Ammoniacal nitrogen (mg/L)	1,335	1,374	1,554	1,207	1,274	1,187	1,322	5
BOD <sub>5</sub> (mg/L)	302	287	269	280	242	273	276	20
COD (mg/L)	1,420	1,374	1,271	1,429	1,238	1,457	1,365	400
BOD <sub>5</sub> /COD	0.21	0.21	0.21	0.20	0.20	0.19	0.20	0.05
Colour (ADMI)	1,568	1,547	1,761	1,644	1,543	1,455	1,586	200
Zinc (mg/L)	3.0	2.5	2.6	3.5	2.9	2.8	2.9	2.0
Iron (mg/L)	5.9	5.2	6.9	5.1	4.9	4.3	5.3	5.0
SS (mg/L)	258	237	249	298	301	168	250	100

1,500 mL leachate sample. Different dosage ratio of 1:25, 1:20, 1:15, 1:10, and 1:5 which were 60, 75, 100, 150, and 300 mL of anaerobic organisms were filled in five containers, respectively, and kept under ambient air for 24 days. A net grating was installed on top of each container to allow anaerobic activity with the surround environment. The containers were kept in normal air condition in the range of 25–30 °C and the percent removal of leachate parameters was measured for COD, NH<sub>3</sub>-N and colour, respectively.

#### *Influence of contact time*

To investigate the effect of removal efficiency of COD, by organic organisms, fixed amount of anaerobic organisms at optimum pH were prepared in 24 containers. A net grating was installed on top of each container to allow anaerobic activity with the surround environment. The sample was kept under normal air condition from day 1 to day 24 regimes. At the elapse of each set contact time, the treated leachate was filtered and analyzed for COD, NH<sub>3</sub>-N and colour, respectively.

#### *Analytical method*

Treated leachate samples were first filtered by using 0.45 μm GC-50 glass micro fibre filters (Advantec, Japan) to retain fines particles from passing through. COD was measured using Calorimetric Method (5220-D) whereby the concentration of NH<sub>3</sub>-N was measured based on the Nesslerization Method (Method: 8038) using UV spectrophotometer (HACH DR 2500, USA). Colour measurements were described as true colour, filtered using 0.45 μm GC-50 (Advantec, Japan) assayed at 455 nm using DR 2500 HACH spectrophotometer. Method No. 2120C reports colour in platinum–cobalt (Pt–Co), the unit of colour being produced by 1 mg platinum/L in the form of the chloroplatinate ion (Al-Hamadani et al. 2011).

## **Results and discussions**

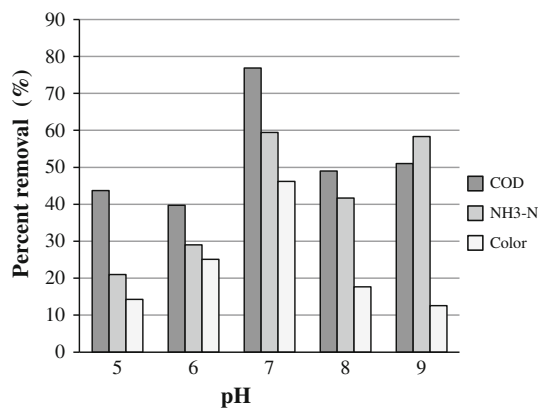
### **Raw leachate characteristics**

Table 1 illustrates some of the leachate characteristics at PBLs. Even though the samples were collected on different days or at different times, the leachate characteristics demonstrate similar fashion and were largely comparable for most of the parameters. The average pH value of the leachate samples is 7.52. According to Umar et al. (2010), the pH of young leachate was less than 6.5, while for old landfill, the leachate has pH higher than 7.5. Initial low pH was due to high concentration of volatile fatty acids

(VFAs). Similarly, stabilized leachate shows fairly constant pH with little variations and it may range between 7.5 and 9. The presence of significant amount of NH<sub>3</sub>-N (1,322 mg/L) in the leachate indicates degradation of soluble nitrogen due to the decomposed waste. As a result, the concentration of NH<sub>3</sub>-N increases with the increase in age of the landfill which was due to hydrolysis and fermentation of nitrogenous fractions of biodegradable refuse substrate (Umar et al. 2010). Higher concentration of NH<sub>3</sub>-N enhances algae growth and promotes eutrophication due to decrease in dissolved oxygen content. Further, nitrification also leads to motivated algal growth, decreased performance of biological treatment systems, accelerated eutrophication, promoted dissolved oxygen depletion and increased toxicity of living organisms in water bodies (Aziz et al. 2010). According to Kurniawan et al. (2006), ammoniacal nitrogen was ranked as a major toxicant to living organisms, as established by various toxicity analyses using bioassays such as *Salmo gairdneri* and *Oncorhynchus nerka*. The average values for BOD<sub>5</sub> and COD were 276 and 1,365 mg/L, respectively. Meanwhile, the calculated BOD<sub>5</sub>/COD ratio was in the ratio of 0:20. In general, The BOD/COD ratio indicates the changes in the amount of biodegradable compounds in the leachate (Warith 2002). Based on the ratio, it shows that the leachate was poorly biodegradable. The low BOD/COD ratio (0:20) also shows that the leachate was stable and difficult to be further degraded biologically (Foul et al. 2009). The high value of pH and low concentrations of COD, BOD<sub>5</sub> and heavy metals indicate that the leachate was in the phase of methane fermentation and classified as anaerobic phase (Salem et al. 2008). Normally, at the early stage of landfill operation, the presence of heavy metals are significant because of higher metal solubility whereby low pH caused by production of organic acids (Kulikowska and Klimiuk 2008). The colour concentration was reported as 1,586 mg/L, which was due to the low biodegradability of dissolved organic constituents in the leachate. Slow process of decomposition of buried waste has induced the percolation of liquid with the refuse. Therefore, dark-coloured liquid is an indicator of saturated organic content in the leachate. Overall, the leachate characteristics did not meet the permissible limit declared under Environmental Quality (Control of Pollution From Solid Waste Transfer Station and Landfill) Regulations 2009 (PU(A) 433) Second Schedule (Regulation 13) from Department of Environment, Malaysia.

### **Optimum pH**

Since the large majority of bacteria present growth optima at or near neutral pH values, most laboratory-based biodegradation studies have been carried out in this pH range. Figure 1 shows the variation of pH adjustment of leachate



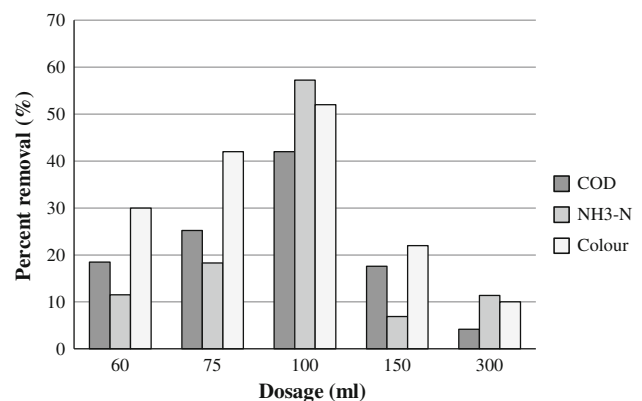
**Fig. 1** Influence of pH for the removal of COD, NH<sub>3</sub>-N and colour

sample to the leachate parameters removal. From the figure, it was shown that anaerobic organisms played a significant role in removal efficiency of leachate parameters. By lowering the pH of leachate sample, COD percentage removal was increased from 39.7 to 43.7 %. Under acidic condition, all organic compounds were fully oxidized to carbon dioxide. In this case, although anaerobic organisms are not an oxidizing agent, microorganisms in the solution dissolved thoroughly in leachate sample that are responsible for the degradation of the organic content. Furthermore, the reduction of COD was due to the organic strength of the leachate which is affected by the reduction of the methanogenic bacteria. Increasing pH from 8 to 9 would lead to a slight increment of the latter from 49 to 51 %. Lowering pH of leachate sample reduced the amount of dissolved oxygen needed by aerobic biological organisms in leachate sample to break down the organic material present, which was known as the worse condition of anaerobic organisms' survival. In contrast, normal pH of leachate leads to higher removal of COD which comprises of 76.8 %. It was assumed that anaerobic organisms would tolerate in the normal pH of leachate. Similar finding was observed for the case of NH<sub>3</sub>-N removal. Under acidic condition, NH<sub>3</sub>-N removal was observed below 29 %, whereas slight increment removal was recorded when pH increased from 8 to 9. According to Rashid and West (2007), most microorganisms exhibit optimal growth at pH values between 6 and 8 and most could not tolerate at pH levels above 9 or below 4. The phenomenon of free ammonia inhibition of ammonia oxidizing bacteria (AOB) and nitrite oxidizing bacteria (NOB) also were found affected by pH adjustment. High pH would increase the free ammonia concentration and inhibits nitrifying activity, especially when the NH<sub>4</sub><sup>+</sup>-N level was high (Zhang et al. 2007). Moreover, increasing the pH of leachate attributed to the higher concentration of NH<sub>3</sub>-N generated from biodegradation process due to parts of protein were biodegraded and some nitrogen species were transferred to the form of NH<sub>3</sub>

after bioreaction. These results suggest that the bacteria produce enzymes whose optimal activity occurs in different pH values. For the colour removal, the highest reduction was achieved when the leachate pH at 7 (46.2 %). As the pH of the leachate increased to alkaline state, further reduction was observed. This was due to condition of the organisms in the mixture of leachate which is susceptible when the pH of the leachate was normal. Since the present work intended to investigate the ability of anaerobic organisms to remove pollutants in a stationary condition of leachate sample, the excess negatively charged hydroxide ions have to compete with inorganic constituents available in the leachate. Therefore, colour removal was not favourable in this condition. This result was in agreement with the work reported by Oliveira et al. (2009).

### Optimum dosage

Figure 2 shows the influence of anaerobic dosage to the removal of leachate parameters. Briefly, removal of COD and NH<sub>3</sub>-N were directly proportional to the anaerobic organism dosage to a certain level. The presence of anaerobic organism in the leachate sample at ratio 1:15 (100 mL) could remove up to 42 % of the COD concentration followed by dosage of 75 mL (1:20) and 100 mL (1:15), respectively. Meanwhile, the anaerobic organism dosage of 150 mL (1:10) and 300 mL (1:5) could only remove COD concentration at 17.6 and 4.2 %, respectively. Therefore, based on the observation from the figure, the highest removal of COD concentration achieved when 100 mL of anaerobic organism was introduced in the leachate sample. Generally, as the dosage of anaerobic organism increased, the removal of COD concentration was increased due to the increasing of active bacteria and organic acid in anaerobic organism including *Bacillus subtilis* which decomposes and digests the organic residuals in leachate. Meanwhile, the lactic acid bacteria increase the carbohydrate metabolism



**Fig. 2** Influence of dosage for the removal of COD, NH<sub>3</sub>-N and colour



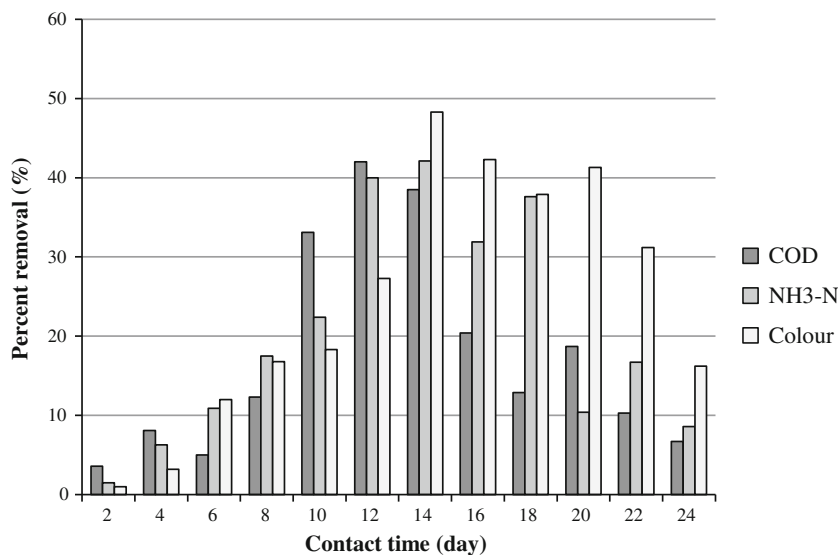
by producing lactic acid and other antimicrobial products. Those antimicrobial products having antibacterial properties that inhibited the growth of pathogens and other non-beneficial microorganisms during the biodegradation of organic particle in leachate sample. From the figure, the removal efficiency of  $\text{NH}_3\text{-N}$  was reduced when incrementing the amount of anaerobic organisms was introduced. At dosage of 100 mL, highest removal of  $\text{NH}_3\text{-N}$  was achieved at 57.21 %. In contrast, reducing dosage of anaerobic organisms resulted low removal of  $\text{NH}_3\text{-N}$ . Low removal in  $\text{NH}_3\text{-N}$  concentration leads to motivated algal growth, accelerated eutrophication and rendered the livelihood of the microorganisms without food availability. These reductions could be attributed to the microbial utilization of these nutrients. Inorganic nitrogen variation should be similar to COD, since COD is consumed to oxidize the inorganic chemicals. The removal pattern for colour was observed similar to COD and  $\text{NH}_3\text{-N}$ , respectively. The highest colour reduction was observed when the anaerobic organism dosage was 100 mL (52 %). As the dosage of the anaerobic organism increases, lower removal of the colour was observed at dosage 1:5. Initially, the culture mixtures available were adequate during the microbial activity in the leachate mixture. Consequently, addition of anaerobic organisms was found to attribute mainly towards the stabilized biological activity of the leachate sample (Zouboulis et al. 2001). This would hinder the breakdown process of dissolved organic constituents which deplete the microbial activity in the leachate that leads to formation of dark liquid in the container.

#### Optimum contact time

The removal efficiency of COD and  $\text{NH}_3\text{-N}$  is shown in Fig. 3. At initial interval of treatment, slight removal was

observed for COD and  $\text{NH}_3\text{-N}$  where only 1.5 and 8.1 % reductions were recorded. The data show that the treatment process provides a consistent high efficiency of COD removal until day 12 accounted for 42 %. Similar observation was also recorded in the case of  $\text{NH}_3\text{-N}$  where the removal percentage was increased from 1.5 to 42.1 % at day 14. Despite the fluctuations of COD and  $\text{NH}_3\text{-N}$  concentrations on the first 14 days of treatment, the removal pattern shows a gradual reduction for the case of COD. Hence, a slow dropped off percent removal of  $\text{NH}_3\text{-N}$  occurred at day 18. Increasing the contact time of anaerobic organisms allowed the microorganism in the leachate sample mixed assisted by acclimatize with the indigenous microbial population in the leachate. As the period of leachate sample prolonged, COD and  $\text{NH}_3\text{-N}$  removal was observed reduced significantly. The differences in reducing abilities of each leachate may be attributed to differences in bacteria species, bacterial biomass concentration or organic type, as well as landfill conditions such as waste content and landfill age. Similarly, microorganisms are often sensitive to their environment. Likewise, high concentration of  $\text{NH}_3\text{-N}$  increased nitrification reduced the rate of biodegradation of leachate. According to Umar et al. (2010), higher concentrations of ammonia are also known to enhance algal growth, promote eutrophication due to decreased dissolved oxygen. Hence, microbial activity was further depleted over time. In addition, longer contact time of anaerobic organisms and leachate sample will reduce the ability of different microorganism to react efficiently with the leachate pollutants, since more oxygen depleted in the containers. The mechanism involved suggests that as anaerobic organism was a mixed culture of many species of microorganisms, some of which can transform from  $\text{NH}_4^+$

**Fig. 3** Influence of contact time for the removal of COD,  $\text{NH}_3\text{-N}$  and colour



**Table 3** Optimum variable condition of anaerobic organisms

Optimum condition			Percent removal		
pH	Dosage	Contact time (day)	COD	NH <sub>3</sub> -N	Colour
7	100 mL	14	65.5	60.2	46.3

to NO<sub>3</sub><sup>-</sup>, thereby decreasing the potential for N-fraction (Javaid 2010). Meanwhile, it was observed that the highest colour removal was recorded at 14 days (48.3 %). Although the reduction of colour was fluctuated after 14 days, the anaerobic organisms continue to decolorize until the 24 days of experiment at slow rate. The possible justification of this behaviour was governed by the rejuvenation of microbial activity in the leachate sample. When the period of incubation increases, longer duration of active organisms in the leachate sample with the surround environment which promotes the active cell of microorganism activity entirely.

#### Optimum variable condition

In assessing the optimum variable conditions of the anaerobic organisms, the evaluation of the preliminary experiment was prerequisite. Since the variable conditions produce different removal efficiency, the selection of the variables was based on the highest removal of COD, NH<sub>3</sub>-N and colour. Table 3 shows the optimum condition of anaerobic organism preparation. It can be observed that the optimum condition of the anaerobic organism achieved at pH 7, dosage of 100 mL and required 14 days to reduce the COD, NH<sub>3</sub>-N and colour pollutants to 65.5, 60.2 and 46.3 %, respectively. The final concentrations of the pollutants are: COD, 470 mg/L; NH<sub>3</sub>-N, 526 mg/L; colour, 851 ADMI. We can observe that the final concentration of the treated leachate sample still exceeded the permissible discharge limit regulated by Malaysia DOE (COD, 400 mg/L; NH<sub>3</sub>-N, 5 mg/L; colour, 200 ADMI).

#### Conclusions

In this study, biological treatment of leachate by anaerobic organisms under ambient condition was investigated. The biological treatment process was found to be feasible for treating the leachate from stabilized landfill. COD, NH<sub>3</sub>-N and colour-removal efficiencies under neutral pH of leachate sample were found in the range of 76.8, 54.4 and 46.2 %, respectively. Increasing the dosage of anaerobic organisms led to a higher removal of COD from 18.5 to 42 %, respectively. In addition, when equal dosage was tested for NH<sub>3</sub>-N, up to 57 % removal was recorded. As

for the colour removal, similar pattern was observed for the removal efficiency. In this case, 100 mL dosage of anaerobic organism was found optimum with 52 % colour reduction. However, further increasing of anaerobic organism resulted in drastic reduction of these parameters between 4.2 and 11.4 %, respectively, for COD, NH<sub>3</sub>-N and colour. Meanwhile, removal pattern of COD, NH<sub>3</sub>-N and colour increased from day 2 and reached maximum at day 14. However, fluctuation pattern was observed starting at day 16 until day 24 for COD and NH<sub>3</sub>-N, respectively. Meanwhile, the optimum condition of the anaerobic organism achieved at pH 7, dosage of 100 mL and required 14 days to reduced COD, NH<sub>3</sub>-N and colour pollutants to 65.5, 60.2 and 46.3 %, respectively. It also indicates that the actual contact time was reduced from 24 to 14 days when the replication of the experiment was conducted, which proves that anaerobic organism could shorten the process of microbial activity in leachate sample. As a result, the application of anaerobic organisms was effective for biological treatment of landfill leachate that simultaneously reduces high concentration of pollutants from landfill. However, the optimal conditions of AOAS were not able to reduce the concentration of leachate sample to a stipulated permissible discharge limit enforced by Malaysia DOE. The partial treatment system such as aeration process can be employed to improve oxidation state of organic content in the leachate sample provided that ammonia stripping is designated after the AOAS treatment to assist denitrification of ammonia content. Since AOAS is a biological process, the application of this treatment can be designated in the pre-treatment of leachate. In this case, the AOAS treatment can be conducted in a reactor supplied with sprinkler system. At a certain time of period, the AOAS will be emitted in the reactor by the sprinkler system and the microbial activity occurs when the aerated leachate flows into the reactor. Conclusively, the AOAS is considered as partial treatment system for leachate remediation process considering its ability to reduce leachate contents. However, more investigation should be conducted to explore the ability of AOAS in full-scale reactor system so that the ability of this microbial system can be optimized.

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