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Effects of activated sludge on the degradation of chlorate in soils under varying environmental conditions

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ABSTRACT

Incubation experiments were conducted to examine the effects of activated sludge on degradation of chlorate in soils. The results show that application of activated sludge could significantly promote the decomposition of soil chlorate though the degradation rate of chlorate did not necessarily increase with increasing application rate of the sludge. The effectiveness of activated sludge on soil chlorate degradation was significantly affected by temperature, moisture content and pH. There is a tendency that the rate of chlorate decomposition increased with increasing temperature and moisture content until optimal values of temperature and moisture content were reached. This can be attributed to the enhanced activity of chlorate-reducing microorganisms in hot and more reducing soil conditions. Soil pH also had important controls on the decomposition of chlorate. The experimental results demonstrate that neutral pH more favoured the degradation of soil chlorate, compared to either acidic or alkaline pH. While soil organic matter content could affect chlorate decomposition, its impact on the effectiveness of activated sludge on chlorate degradation was minor. This study has implications for developing cost-effective techniques for remediating chlorate-contaminated soils, particularly in the longan-producing countries.

1. Introduction

Chlorate ($ClO_3{}^{-1}$) is a powerful oxidant which poses toxic effects on a diverse range of organisms [1–5]. Due to its potential threat to human health, chlorate is listed by the World Health Organization as a chemical of health significance in drinking water [6]. Globally, chlorate is produced on a large scale by the chemical industry and used in a wide range of applications. It is manufactured for the production of chlorine dioxide which is used as a bleaching agent in the pulp and paper industry and in oxidative treatment of drinking water. Chlorate also has wide applications in agriculture, e.g. being used as a weed controller (herbicide), defoliant and soil sterilant [7]. Yen et al. [8] discovered that the application of potassium chlorate ($KClO_3$) can induce off-season flowering in longan (*Dimocarpus longan*, *Lour.*), a very popular fruit tree in southern China and Southeast Asia. Since then, potassium or sodium chlorate has been increasingly used for off-season longan production in many longan-producing countries [9,10]. Manochai et al. [11] reported that the most effective method to apply chlorate to longan trees was soil drenching. However, this application method may lead to massive accumulation of residual chlorate in the soils and subsequently have adverse impacts on the soil ecosystems, as well as the growth of longan [12]. It is also likely that substantial amounts of chlorate stored in soils are exported to aquatic environments during heavy rainfall events. Therefore, appropriate methods and techniques are needed for *in situ* remediation of chlorate-contaminated soils in order to minimize its environmental impacts.

Biodegradation provides a possible way for reducing or eliminating the potentially environmental hazards from improper uses of chlorate. Chlorate-reducing bacteria (CRB) are known to exist in natural environments under anaerobic conditions [13–15]. CRB can obtain energy for growth by simultaneously degrading chlorate and organic substances in anaerobic conditions [16]. Although a few CRB have been identified and isolated [14,17–21], they are not commercially available for large-scale field applications.

Germgard [22] examined the effects of organic fertilizer on chlorate degradation in the soils of a longan plantation where potassium chlorate was applied for over 3 years. He found that there was a significant difference in residual chlorate between the treated and untreated soils. Pathipan et al. [23] and Sutigoolabud et al. [24] also

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Table 1
Major physical and chemical characteristics of the three soils used in this study

Soil	pH	OM (%)	Soil texture	MB (mg kg^{-1})	CEC (cmol kg^{-1})	TC (mg kg^{-1})	KN (mg kg^{-1})
Zhanjiang	5.4	0.5	Sand	5.2	6.7	1895	44.6
Guangzhou	5.3	2.1	Sandy loam	98.1	15.4	2780	73.5
Maoming	5.6	3.5	Clay	175.0	18.2	3500	82.1

OM: organic matter content; MB: microbial biomass; CEC: cation exchange capacity; TC: total carbon; KN: Kjeldahl nitrogen.

observed an accelerated degradation of chlorate in a contaminated soil by application of molasses. Hunter [25,26] showed that addition of vegetable oil to a bioreactor consisting of a gravel column had a significant effect on the degradation of chlorate in groundwater.

Many of the above organic materials are either too costly or not present in substantial amounts that allow large-scale application. Activated sludge is an abundant waste/by-product from municipal wastewater treatment. This material frequently contains substantial organic matter and a diverse range of microorganisms. It is possible that some of the activated sludge-borne bacteria are capable of reducing chlorate [19]. Therefore, application of activated sludge may offer an opportunity to develop cost-effective techniques for remediating large-scale chlorate-contaminated soils. The objective of this study was therefore to examine the effects of activated sludge application on the degradation of chlorate in soils under varying intensity of environmental parameters, including temperature, soil moisture, pH and organic matter content. This knowledge is needed for optimizing the reaction conditions for chlorate degradation in soils, which is an important step toward developing effective methods for remediation of chlorate-contaminated soils using activated sludge as a major soil amendment.

2. Materials and methods

2.1. Materials

2.1.1. The soils

The soils used for this study were collected from the longan tree plantations located at three locations in the southern China region: (1) Guangzhou (latitude: 23°08′N; longitude: 113°14′E), (2) Maoming (latitude: 21°26′N; longitude: 111°20′E) and (3) Zhanjiang (latitude: 21°11′N; longitude: 110°24′E). No chlorate has been applied in these longan plantations prior to the collection of the soil samples. The major soil characteristics are given in Table 1.

2.1.2. The activated sludge

The activated sludge used in this study was taken from a local municipal sewage plant located near the Guangzhou City. After collection, the sludge sample was stored at 4 °C prior to the experiment. Some major chemical characteristics of this activated sludge can be seen from Table 2.

2.1.3. Chlorate and artificial chlorate-contaminated soils

Analytical grade potassium chlorate was used to formulate the artificial chlorate-contaminated soils. The chlorate concentration of all the artificial chlorate-contaminated soils used in this study was set at a fixed value of 300 mg chlorate kg^{-1} soil by thoroughly mixing an appropriate amount of potassium chlorate powder into the soils.

2.2. Experimental design

Five experiments were conducted to examine the effects of (1) varying quantity of the added sludge, (2) varying soil temperature, (3) varying soil moisture content, (4) varying soil pH, and (5) varying soil organic matter content on the degradation of chlorate in the soils.

2.2.1. Experiment 1: varying quantity of the added sludge

This experiment was performed in 500 mL sterile Erlenmeyer flasks; each contained 100 g of the Maoming soil adjusted to pH 7.0 with 1 M NaOH solution. One control and three treatments (each in three replicates) were set for this experiment: (1) control: containing no added sludge; (2) treatment 1: containing 0.07% added sludge; (3) treatment 2: containing 0.7% added sludge; and (4) treatment 3: containing 1% added sludge. The soils in the Erlenmeyer flasks were supersaturated with deionized water and then incubated at room temperature for 20 days. The incubated soils were agitated at 400 rpm for 5 min once a day to accelerate the reaction. Each week, an appropriate amount of sterile deionized water was added to the samples to compensate for water loss by evaporation. An appropriate amount of soil sample was taken from each Erlenmeyer flask at the following times: 5, 8, 10, 15 and 20 days after the commencement of the experiment. The samples were used for determination of chlorate concentration.

2.2.2. Experiment 2: varying soil temperature

This experiment was performed in 500 mL sterile Erlenmeyer flasks; each contained 200 g of the Maoming soil adjusted to pH 7.0 with 1 M NaOH solution. The application rate of activated sludge was fixed to obtain a uniform soil with sludge content of 0.7%. Five treatments (each in three replicates) were set for this experiment: (1) treatment 1: at 5 °C; (2) treatment 2: at 10 °C; (3) treatment 3: at 20 °C; (4) treatment 4: at 30 °C; and (5) treatment 5: at 40 °C. The soils for various treatments were pre-incubated at 4 °C overnight before being transferred to various incubators with set temperature at 5, 10, 20, 30 and 40 °C, respectively. The moisture content of the soils was maintained at a similar level during the entire period of the experiment by daily addition of sterile deionized water into the incubated soils. An appropriate amount of soil sample was taken from each Erlenmeyer flask at the following times: 5, 8, 10, 15 and 20 days after the commencement of the experiment. The samples were used for determination of chlorate concentration.

Table 2
Major physical and chemical characteristics of the activated sludge used in this study

Parameter	Value
Water content (%)	27
pH	6.95
Organic matter content (%)	38
Total carbon (mg kg^{-1})	5600
Kjeldahl nitrogen (mg kg^{-1})	25000
C:N	100:446
Arsenic (mg kg^{-1})	2.3
Cadmium (mg kg^{-1})	<1
Chromium (mg kg^{-1})	15
Lead (mg kg^{-1})	20
Zinc (mg kg^{-1})	75
Copper (mg kg^{-1})	63
Nickel (mg kg^{-1})	41

2.2.3. Experiment 3: varying soil moisture

This experimental apparatus and procedure for soil formulation were the same as described in Section 2.2.2. Four treatments (each in three replicates) were set for this experiment: (1) treatment 1: at 15%; (2) treatment 2: at 30%; (3) treatment 3: at 40%; and (4) treatment 4: at 50% of the water holding capacity (WHC, ca. −1500 kPa water potential). The soils for various treatments were pre-incubated at 4 °C overnight and then incubated at 20 °C for 20 days. An appropriate amount of soil sample was taken from each Erlenmeyer flask at the following times: 5, 8, 10, 15 and 20 days after the commencement of the experiment. The samples were used for determination of chlorate concentration.

2.2.4. Experiment 4: varying soil pH

This experimental apparatus and procedure for soil formulation were the same as described in Sections 2.2.2 and 2.2.3, except that soil pH was not adjusted to a fixed value of 7.0. Moisture contents of the soils were adjusted to 30% of the WHC. Three treatments (each in 3 replicates) were set for this experiment: (1) treatment 1: at pH 5.4; (2) treatment 2: at pH 6.8; and (3) treatment 3: at pH 8.3. The soils for various treatments were pre-incubated at 4 °C overnight and then incubated at 20 °C for 20 days. An appropriate amount of soil sample was taken from each Erlenmeyer flask at the following times: 5, 8, 10, 15 and 20 days after the commencement of the experiment. The samples were used for determination of chlorate concentration.

2.2.5. Experiment 5: varying soil organic matter content

In this experiment, three soils collected from different locations (i.e. Maoming, Guangzhou and Zhanjiang) were used. There soils had varying organic matter content as shown in Table 1: 3.5% for the Maoming soil, 2.1% for the Guangzhou soil, and 0.5% of the Zhanjiang soil. They were used to examine the influence of abundance of naturally occurring soil organic matter on the degradation of chlorate. The experiment was performed in 500 mL sterile Erlenmeyer flasks. For each soil type, 200 g of soil were used (each in three replicates). Appropriate application rate was applied to obtain a sludge content of 0.7%. Moisture contents of the soils were adjusted to 30% of the WHC. An appropriate amount of soil sample was taken from each Erlenmeyer flask at the following times: 5, 8, 10, 15 and 20 days after the commencement of the experiment. The samples were used for determination of chlorate concentration.

2.3. Laboratory and statistical analysis

Chlorate was determined by a five-step iodometry [27]. The statistical significance of difference between treatment means was determined by one-way analysis of variance (ANOVA).

3. Results

3.1. Quantity influence of activated sludge

Changes in residual chlorate concentration for the control and different treatments are shown in Table 3. There is a trend that residual chlorate concentration decreased with increasing incubation time for the control and the treatments. However, treatments tended to have a greater magnitude of decrease in the concentration of residual chlorate, compared to the control. Statistical analysis indicates that at the 5th day of incubation, there was no statistically significant difference in residual chlorate concentration between the control and the treatments, except for treatment 2; from the 10th day onward, the residual chlorate concentration was always significantly higher in the control than in the treatments at each sampling occasion. At the end of the experiment (the 20th day), the residual chlorate concentration was only 22.7 mg kg^{-1} (less than 7.6% of the originally added chlorate) and 46.4 mg kg^{-1} (less than 16% of the originally added chlorate) in the soils with the sludge concentration of 0.7% (treatment 2) and 1% (treatment 1), respectively.

3.2. Effects of temperature

When the temperature was below 10 °C, no significant drop in residual chlorate concentration was observed at the end of this experiment. Increase in incubation temperature to 20 °C led to a significant drop in residual chlorate concentration and at the end of the experiment, the residual chlorate concentration was below 200 mg kg^{-1}. Further increase in temperature to 30 °C brought about a more significant concentration drop and a nearly linear relationship can be established between the residual chlorate concentration and the incubation time; at the end of the experiment, the residual chlorate concentration dropped to below 50 mg kg^{-1}. There is no marked difference between the pattern of trendlines for 30 and 40 °C incubation treatments (Fig. 1).

3.3. Effect of soil moisture

When the soil moisture content was at 15% of WHC, residual chlorate concentration decreased gently with increasing length of incubation and at the end of experiment the residual chlorate concentration was still above 200 mg kg^{-1}. Increase in soil moisture content led to a decreased residual chlorate concentration and at the end of the experiment, the residual chlorate concentration was 35.6, 75.7 and 50.9 mg kg^{-1} for the soils with moisture content of 30, 40 and 50% of WHC, respectively (Fig. 2).

3.4. Effect of pH

It can be seen from Fig. 3 that concentration drop of residual chlorate during the period of experiment was more significant in

Table 3
Variation on chlorate concentration in soil amended with varying amounts of activated sludge during the entire course of incubation experiment

Treatment	Soil chlorate (mg kg^{-1}) at different times				
	5 days	8 days	10 days	15 days	20 days
Control	295.5 ± 1.6 a	283.4 ± 3.7 a	258.6 ± 5.9 a	226.1 ± 6.2 a	198.7 ± 4.3 a
Treatment 1	283.4 ± 3.6 a	265.4 ± 3.0 a	243.8 ± 2.2 b	157.8 ± 3.9 b	85.2 ± 3.4 b
Treatment 2	274.7 ± 5.9 b	234.2 ± 6.5 c	152.7 ± 3.6 d	78.3 ± 4.9 d	22.7 ± 2.1 d
Treatment 3	287.5 ± 11.2 a	249.7 ± 4.8 b	186.4 ± 5.0 c	97.7 ± 5.0 c	46.4 ± 3.1 c

Each value is the mean of three replicates. Different letters (a–d) behind the values indicate significant difference at P = 0.05 among treatments based on LSD test, one-way ANOVA (n = 3).

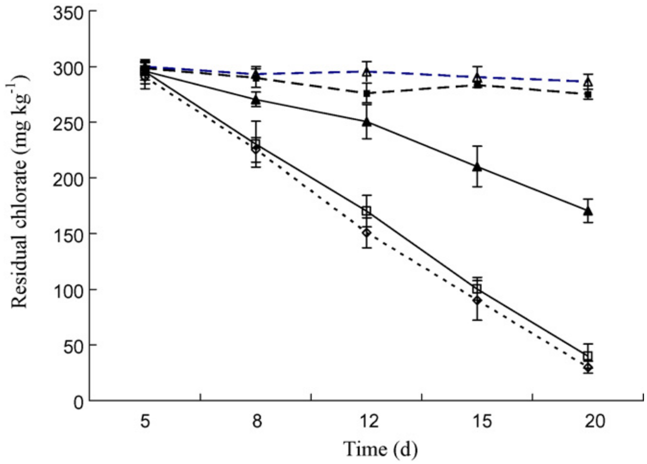

Fig. 1. Variation on chlorate concentration in the artificially chlorate-contaminated soil amended with activated sludge under varying temperatures during the entire period of incubation experiment (symbols in the figure denote temperature at 5 °C (△), 10 °C (■), 20 °C (▲), 30 °C (□) and 40 °C (◊); control adjustment is made for each data point which is the difference between a treatment and the control; each value is the mean of three replicates; error bars represent standard deviation of the mean).

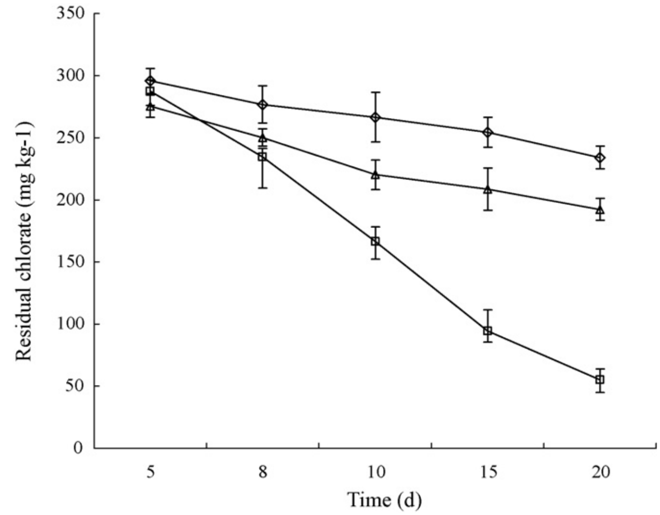

Fig. 3. Variation on chlorate concentration in the artificially chlorate-contaminated soil amended with activated sludge under varying soil pH during the entire period of incubation experiment (symbols in the figure denote pH at 5.4 (◊), 6.8 (□) and 8.3 (△); control adjustment is made for each data point which is the difference between a treatment and the control; each value is the mean of three replicates; error bars represent standard deviation of the mean).

the soil with pH 6.8 than in the soil with either a higher or lower pH. Less than 10% of the initial dose was recovered at the end of the experiment for the soil with pH 6.8 while more than 75% of the initially added chlorate was recovered at the same time for the soil with pH 5.4 or 8.3. There was no significant difference ($P = 0.05$) in residual chlorate between the soil with pH 5.4 and the soil with pH 8.3 though residual chlorate concentration was slightly higher in the former than in the latter on each sampling occasion.

3.5. Effect of soil organic matter content

Chlorate concentration versus incubation time pattern was very similar to each other for the all three soils with different organic matter contents (Fig. 4). However, except at the 5th day of the experiment, residual chlorate concentration was in the following decreasing order at any sampling occasions: Zhanjiang > Guangzhou > Maoming. Since the organic matter content of the soils was in an opposite order, i.e. Maoming > Guangzhou > Zhanjiang (refer to Table 1), Therefore, this reflects that residual chlorate concentration in the activated sludge-amended soils decreased with increasing organic matter content originally contained in the soils.

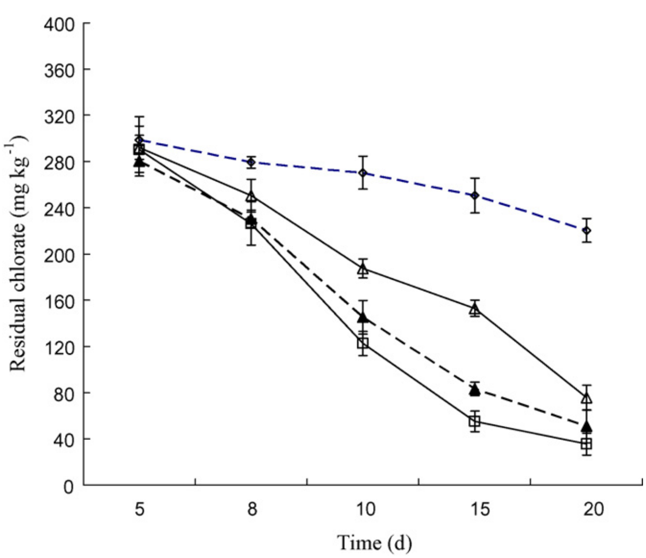

Fig. 2. Variation on chlorate concentration in the artificially chlorate-contaminated soil amended with activated sludge under varying soil moisture contents during the entire period of incubation experiment (symbols in the figure denote moisture content at 15% (◊), 30% (△), 40% (▲) and 50% (□) of the water holding capacity; control adjustment is made for each data point which is the difference between a treatment and the control; each value is the mean of three replicates; error bars represent standard deviation of the mean).

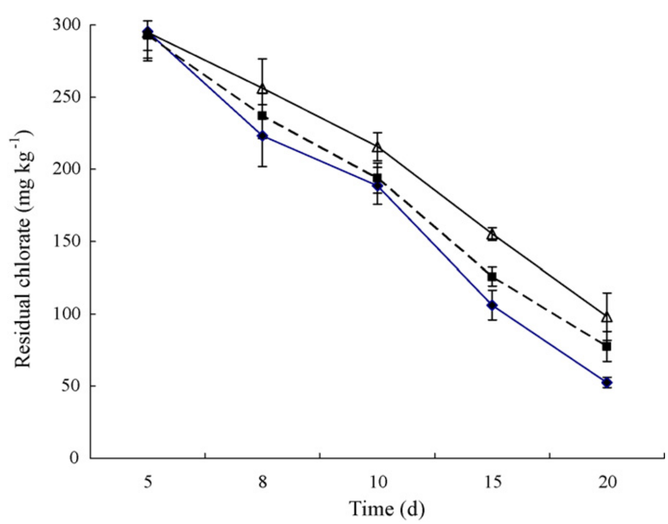

Fig. 4. Variation on chlorate concentration in the three artificially chlorate-contaminated soils that were formulated separately from three different original soils with differing organic matter contents during the entire period of incubation experiment; these soils were amended with the same amount of activated sludge and under the same temperature, moisture and pH conditions (symbols in the figure denote the soil sample collected from Maoming (♦), Guangzhou (■) and Zhanjiang (△); control adjustment is made for each data point which is the difference between a treatment and the control; each value is the mean of three replicates; error bars represent standard deviation of the mean).

4. Discussion

The results obtained from this study suggest that although chlorate undergoes degradation in the soil that was not amended with activated sludge, addition of activated sludge into chlorate-contaminated soils can significantly accelerate the degradation of chlorate. At an application rate of about 0.7 g sludge kg^{-1} soil (treatment 1), significant effects of the added sludge on promoting chlorate degradation could be observed after the 10th day of incubation; when the application rate was increased to about 7 g sludge kg^{-1} soil (treatment 2), significant effects of the added sludge on promoting chlorate degradation could be observed after the 5th day of incubation; further increase in application rate to about 10 g sludge kg^{-1} soil (treatment 3) did not do better than treatment 2 but on the contrary, it caused a slower degradation of chlorate, compared to treatment 2. Activated sludge can become 'souring' under anaerobic conditions which favour the conversion of particulate organic molecules into soluble organic acids [28]. The higher sludge application rate could result in increased production and accumulation of organic acids [29,30]. The production of organic acids and CO_2 due to anaerobic decomposition of organic matter is expected to cause pH drop which may inhibit the growth of CRB. This may explain the reduced rate of chlorate degradation in treatment 3 where a high rate (10 g sludge kg^{-1} soil) was applied.

The current work confirms that the activated sludge is also capable of degrading chlorate in soils. It is likely that the presence of certain organic matter forms promotes the activity of chlorate-reducing microorganisms. The optimal ratio of carbon to nitrogen for the microbial proliferation was generally considered to be 10:1 [31]. The C:N ratio of the Maoming soil used for this experiment was about 42:1 (refer to Table 1), which does not favour microbial proliferation. The activated sludge had a C:N ratio of 1:4.5 (refer to Table 2). The addition of sludge to soil may enable an adjustment of C:N ratio toward a more optimal level for the proliferation of chlorate-reducing microorganisms and consequently enhance the degradation of chlorate. However, over-treatment of chlorate-contaminated soils with activated sludge may have opposite effects on the degradation of chlorate.

The effects of activated sludge on chlorate degradation in soils also depend on various environment factors. Temperature plays an important role in this aspect. At low temperature (<10 °C), chlorate degradation is extremely slow despite the presence of activated sludge. This is probably related to the depressed microbial activity under low temperature conditions. Under the experimental conditions set for this study, the optimal temperature for chlorate degradation in the presence of activated sludge was 30 °C. A higher temperature than 30 °C did not further enhance the degradation of chlorate to a significant degree. Chlorate degradation was not weakened even at a temperature as high as 40 °C, compared to that at 30 °C. This indicates that the sludge borne-CRB are of tolerance to high temperature that frequently encountered in the tropical and subtropical area where longan trees are grown.

The effects of activated sludge on chlorate degradation in soils are also affected by soil moisture content. The observed increase in chlorate degradation rate as a result of increased moisture content can be attributed to the intensified reducing conditions as the soil pores are increasingly filled by water. Chlorate decomposition through reduction reactions is mediated by chlorate-reducing microorganisms which are basically anaerobic [13,32]. Under the experimental conditions set for this study, if a soil moisture content was maintained between 30 and 50% of the WHC, satisfactory chlorate degradation rate can be achieved.

Clearly, either acidic or alkaline condition does not favour chlorate degradation even in the presence of activated sludge. Cove [33] also found that chlorate degradation could be retarded in acid soils. Probably, either acidic or alkaline pH can inhibit the growth of CRB and subsequently the degradation of chlorate. Under the experimental conditions set for this study, the optimal pH for chlorate degradation in the presence of activated sludge was around 7, i.e. neutral conditions.

Organic-rich soils may experience rapider chlorate degradation than soils with low organic matter content when they are amended with the same amounts of activated sludge. However, it appears that soil organic matter content only plays a minor role in the process. In this study, even for the Zhangjiang soil which had an organic matter content of only 0.5%, marked degradation of soil chlorate could still be achieved after 20 days incubation when it was amended with activated sludge.

The research results obtained from this study have implications for remediation of chlorate-contaminated soils, especially in longan plantations. Sewage sludge is an abundant material and can be found in many municipal regions. Therefore activated sludge has a higher availability, compared to many other forms of organic materials. Manochai et al. [11] compared the effectiveness of different methods for potassium chlorate application in longan plantation and found that soil drenching was the most effective one among the investigated methods. Therefore, the soil chlorate can be treated by adding appropriate amounts of activated sludge as soil drenches with pH and soil moisture content being controlled at levels that favour chlorate degradation. It must be realized that the current experimental results can only provide a generic guide for soil conditioning. Field experimentation is needed to optimize the actual field conditions for effective chlorate elimination.

5. Conclusions

Under the experimental conditions set in this study, application of activated sludge could significantly enhance the degradation of chlorate in soils. However, the degradation rate does not necessarily increase with increasing amounts of added sludge. Increase in added sludge could result in increased amounts of organic acids and CO_2 being produced through decomposition of organic matter under anaerobic conditions and this may lead to soil acidification which in turn inhibits the growth of CRB that catalyzes chlorate degradation.

Environmental factors such as temperature, moisture and pH have great controls on the rate of soil chlorate decomposition. Degradation of soil chlorate could be significantly depressed when the temperature is below 10 °C. Increase in temperature tends to more favour the degradation of soil chlorate until the optimal temperature (around 30 °C) is reached. Chlorate decomposition rate increases with increasing soil moisture content at least until 50% of WHC is attained. Soil pH also had important controls on the decomposition of chlorate. Soils with neutral pH is more favourable than acidic or alkaline soils for the degradation of chlorate. Although organic matter content that the soils originally contained (prior to activated sludge application) had some influences on chlorate decomposition, its impact on the effectiveness of activated sludge on chlorate degradation was smaller, compared to the above three soil environmental factors.

The research findings obtained from this study have implications for developing cost-effective techniques for the remediation of chlorate-contaminated soils, particularly in the longan-producing countries of the tropical and subtropical zones.

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References

[1] F.S. Woodiwiss, G. Fretwell, The toxicities of sewage effluents, industrial discharges and some chemical substances to brown trout (Salmo trutta) in the Trent River authority area, Water Pollut. Control 73 (1974) 396–405.
[2] A.G. Brownlee, H.N. Arst, Nitrate uptake in *Aspergillus nidulans* and involvement of the third gene of the nitrate assimilation gene cluster, J. Bacteriol. 155 (1983) 1138–1146.
[3] L.M. Dosdall, L.R. Goodwin, R.J. Casey, L. Noton, The effect of ambient concentrations of chlorate on survival of freshwater aquatic invertebrates, Water Qual. Res. J. Can. 32 (1997) 839–854.
[4] D.J. van Wijk, S.G.M. Kroon, I.C.M. Garttener-Arends, Toxicity of chlorate and chlorite to selected species of algae, bacteria, and fungi, Ecotoxicol. Environ. Saf. 40 (1998) 206–211.
[5] R.C. Anderson, S.A. Buckley, L.F. Kubena, L.H. Stanker, R.B. Harvey, D.J. Nisbet, Bactericidal effect of sodium chlorate on *Escherichia coli* O157:H7 and *Salmonella typhimurium* DT104 in rumen contents in vitro, Food Prot. 63 (2000) 1038–1042.
[6] World Health Organization, Guidelines for Drinking-water Quality, 3rd edition, WHO, Geneva, 2004, 335, 392, 414.
[7] B.E. Logan, A review of chlorate and perchlorate respiring microorganisms, Bioremed. J. 2 (1998) 69–79.
[8] C.R. Yen, C.N. Chau, J.W. Chang, J.C. Tzeng, Effects of chemicals on flowering in longan, Chin. Soc. Horticult. Sci. 47 (2001) 195–200 (in Chinese, with English abstract).
[9] S. Subhadrabandhu, C. Yapwattanaphun, Lychee and longan production in Thailand, Acta Hortic. 558 (2001) 49–58.
[10] S. Subhadrabandhu, C. Yapwattanaphun, Regulation off-season flowering of longan in Thailand, Acta Hortic. 558 (2001) 193–198.
[11] P. Manochai, P. Sruamsiri, W. Wiriyaalongkorn, D. Naphrom, M. Hegele, K. Bangerth, Year around off season flower induction in longan(*Dimocarpus longan*, *Lour.*) trees by $KClO_3$ applications: potentials and problems, Sci. Hortic. 104 (2005) 379–390.
[12] J.H. Zhu, H.X. Peng, Technique of inducing longan flowering with chemicals, Guangxi Trop. Agric. (in Chinese) 3 (2002) 12–14.
[13] C.G. van Ginkel, C. Plugge, C. Stroo, Reduction of chlorate with various energy substrates and inocula under anaerobic conditions, Chemosphere 31 (1995) 4066–4075.
[14] J.D. Coates, U. Michaelidou, R.A. Bruce, S.M. O'Connor, J.N. Crespi, L.A. Achenbach, Ubiquity and diversity of dissimilatory (per)chlorate-reducing bacteria, Appl. Environ. Microbiol. 65 (1999) 5234–5241.
[15] J. Wu, R.F. Unz, H. Zhang, B.E. Logan, Persistence of perchlorate and the relative numbers of perchlorate- and chlorate-respiring micro-organisms in natural waters, soils and wastewater, Bioremed. J. 5 (2001) 119–130.
[16] N.C. Tan, W. van Doesburg, A.A. Langenhoff, A.J. Stams, Benzene degradation coupled with chlorate reduction in a soil column study, Biodegradation 17 (2006) 113–119.
[17] V. Stepanyuk, G. Smirnova, T. Klyushnikova, N. Kanyuk, L. Panchenko, T. Nogina, V. Prima, New species of the *Acinetobacter* genus *Acinetobacter thermotoleranticus* sp. nov, J. Microbiol. (English translation of Mikrobiologiya) 61 (1992) 347–356.
[18] A. Malmqvist, T. Welander, E. Moore, A. Ternstrom, G. Molin, I.M. Stenstrom, *Ideonella dechloratans* gen. nov., sp. nov., a new bacterium capable of growing anaerobically with chlorate as an electron acceptor, Syst. Appl. Microbiol. 17 (1994) 58–64.
[19] G.B. Rikken, A.G.M. Kroon, C.G. van Ginkel, Transformation of (per)chlorate in chloride by a newly isolated bacterium: reduction and dismutation, Appl. Microbiol. Biotechnol. 45 (1996) 420–426.
[20] K. Stenklo, H.D. Thorell, H. Bergius, R. Aasa, T. Nilsson, Chlorite dismutase from *Ideonella dechloratans*, J. Biol. Inorg. Chem. 6 (2001) 601–607.
[21] H.D. Thorell, K. Stenklo, J. Karlsson, T. Nilsson, A gene cluster for chlorate metabolism in *Ideonella dechloratans*, Appl. Environ. Microbiol. 69 (2003) 5585–5592.
[22] U. Germgard, Chlorate discharges from bleach plants: how to handle a potential environmental problem, Pap. Puu. 71 (1989) 255–260.
[23] S. Pathipan, S. Keishi, O. Somchai, Decontamination of chlorate in longan plantation soils by bio-stimulation with sugar amendment, Soil Sci. Plant Nutr. 50 (2004) 249–256.
[24] P. Sutigoolabud, K. Senoo, S. Ongprasert, T. Mizuno, T. Mishima, M. Hisamatsu, H. Obata, Decontamination of chlorate in longan plantation soils by bio-stimulation with molasses amendment, Soil Sci. Plant Nutr. 51 (2005) 583–588.
[25] W.J. Hunter, Use of vegetable oil in a pilot-scale denitrifying wall, Contam. Hydrol. 53 (2001) 119–131.
[26] W.J. Hunter, Bioremediation of chlorate or perchlorate contaminated water using permeable barriers containing vegetable oil, Curr. Microbiol. 45 (2002) 287–292.
[27] USEPA, Perchlorate Environmental Contamination: Toxicological Review and Risk Characterization (External Review Draft), NCEA-1-0503, U.S. Environmental Protection Agency, Washington, DC, Office of Research and Development, 2002.
[28] N. Ferreiro, M. Soto, Anaerobic hydrolysis of primary sludge: influence of sludge concentration and temperature, Water Sci. Technol. 47 (2003) 239–246.
[29] M.C. Wu, K.W. Sun, Y. Zhang, Influence of temperature fluctuation on thermophilic anaerobic digestion of municipal organic solid waste, J. Zhejiang Univ. Sci. B 7 (2006) 180–185.
[30] J.R. Banu, S. Kaliappan, I.T. Yeom, Treatment of domestic wastewater using upflow anaerobic sludge blanket reactor, Int. J. Environ. Sci. Technol. 4 (2007) 363–370.
[31] USEPA, How to Evaluate Alternative Cleanup Technologies for Underground Storage Tank Sites: A Guide for Corrective Action Plan Reviews 510-R-04-002, U.S. Environmental Protection Agency, Washingon, DC, 1995.
[32] A. Malmqvist, T. Welander, L. Gunnarsson, Anaerobic growth of microorganisms with chlorate as an electron acceptor, Appl. Environ. Microbiol. 57 (1991) 2229–2232.
[33] J.D. Cove, Chlorate toxicity in *Aspergillus nidulans*. Studies of mutants altered in nitrate assimilation, Mol. Gen. Genet. 146 (1976) 147–159.