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Enhanced removal of *Chironomus kiiensis* larvae in conventional water treatment process by pre-oxidation

Sun Xing-bin, Jiang Yi-feng and Cui Fu-yi

ABSTRACT

Chironomus kiiensis larvae, which cannot be exterminated by conventional disinfection processes, propagate prolifically in eutrophic water bodies, therefore posing a potential problem for drinking water quality. In this work, quantitative experimental studies were carried out on inactivation of *Chironomus kiiensis* larvae using conventional oxidants of chlorine (Cl_2), chlorine dioxide (ClO_2) and ozone under conditions of different oxidant dosages, organic precursor concentration and pH values. The results showed that three oxidants used could inactivate *Chironomus kiiensis* larvae to some extent. Chlorine and chlorine dioxide were proved to be the least and most efficient oxidant for inactivating *Chironomus kiiensis* larvae, respectively. In addition, the synergic removal effects with pre-oxidation followed by conventional water treatment processes were also evaluated. The results showed that pre-oxidation could lead to enhanced removal of the inactivated organism by the subsequent coagulation process while lowering the dosage of oxidant. In view of the need to secure drinking water quality, it is necessary to combine oxidant inactivation with conventional processes for removing *Chironomus kiiensis* larvae from micro-polluted water sources.

Key words | *Chironomus kiiensis* larvae, chlorine dioxide, inactivation, ozone, pre-oxidation, water treatment

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INTRODUCTION

Chironomus kiiensis, from the dipteran family Chironomidae, is widely distributed in the northern hemisphere at temperate latitudes. They can be found in both lentic and lotic environments, and especially in organically enriched water bodies. Thus, densities of *Chironomus kiiensis* usually increase rapidly in water sources such as reservoirs and fresh water lakes with eutrophic characteristics (Gtirgy & Judit 1983), which therefore induce the risk of extra first instar of larvae to be removed in drinking water treatment process for it is plankton (Bay 1993; Michael 1997; Sun *et al.* 2007). Although there are no indications that these organisms pose a threat to public health (WHO 1996), their presence is not appreciated because most people associate the organisms with poor hygiene.

Owing to its motility, the first instar larvae can easily penetrate the sand filter and enter waterworks reservoirs and municipal service pipes. However, recent studies have revealed that *Chironomus kiiensis* larvae are difficult to remove in conventional water treatment processes by flocculation, sedimentation and subsequent filtration steps (Zhou *et al.* 2003; Van Lieverloo *et al.* 2004; Sun *et al.* 2007). Therefore, inactivation or weakening of *Chironomus kiiensis* larvae with chemical oxidants is believed to be crucial for the complete removal of these larvae from water sources. The promising combination of chemical pre-oxidation with conventional water treatment processes is thus put forward in this paper. The inactivation effects with currently available oxidants were thereby evaluated comparatively, with more

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attention paid to chlorine, chlorine dioxide and ozone. In addition, the synergic removal of *Chironomus kiiensis* larvae in such a hybrid system is clarified in detail, as a useful reference for other waterworks.

MATERIALS AND METHODS

The preparation of oxidants

The stock solutions of chlorine were prepared by diluting a commercial solution of sodium hypochlorite (NaOCl, 9% active chlorine). Chlorine dioxide stock solution with a purity of 99% was made using the method described by Ruffell *et al.* (2000). The stock chlorine dioxide solution was usually diluted to obtain a concentration of about 1 g l^{-1} in order to facilitate the addition of low concentrations to the water samples. Diluted chlorine dioxide stock solutions were stored in head-free 50-ml amber vials at 4°C in the dark. The concentration was measured with the N,N-diethyl-*p*-phenylenediamine (DPD) methods (*Standard Methods* 1998). Ozone was generated from oxygen carrier gas, and sparged into cold Milli-Q water no longer than 1 h before use. It was stored under dark, refrigerated conditions prior to use. Ozone concentration in the stock solution ranged from 10 to 20 mg l^{-1}. Ozone from the stock solution was transferred to batch reactors using a syringe attached with Nalgene tubing so that the ozone could be injected into the bottom of the reactor. Ozone residual in the batch reactors was measured by the indigo method.

Culturing of *Chironomus kiiensis* larvae

Egg masses of *Chironomus riparius* were initially obtained from a population collected in the Shenzhen reservoir (Guangdong, China) and maintained in the laboratory for several generations. *Chironomus kiiensis* larvae were cultured in aerated 25-l glass aquaria filled with aerated tap water. A 5 cm thick artificial sediment layer consisting of washed siliceous sand and cellulose was introduced at the bottom of the aquarium. Adult midges were confined using wooden cages covered with 1 mm mesh size metal net. Aquaria were placed under constant temperature (20°C) and photoperiod (14 h light/10 h dark). In order to obtain homogeneous samples (size and age), egg masses were transferred from rearing aquaria into 2-l glass experimental tanks filled with aerated tap water. Egg masses were left for 24 h in these tanks and non-hatched eggs were then removed. All bioassays were conducted using first instar larvae.

Experimental procedures

The experiments were carried out in two stages. In the first stage, inactivation of *Chironomus kiiensis* larvae with three different oxidants of chlorine, chlorine dioxide and ozone was investigated experimentally; and in the second stage, pre-oxidation was combined with a clarification process for further investigation of the synergic removal mechanism of *Chironomus kiiensis* larvae.

Distilled water solution was used in control assays. Three 1-l heat resistant glass beakers were used as reaction vessels. The beakers were immersed in a recirculating water bath to control temperature. The first larval stage organisms used in each experiment were obtained by transferring egg ropes from the culture to glass vessels containing culture water. The hatched animals were then transferred randomly to each test jar with a glass pipette until each reactor contained 15 larvae. At the desired time points, larvae were regarded as dead if unable to make a sustained and coordinated response (at least two consecutive sinuate movements involving 25% or more of total body length) when grasped with a pair of fine forceps. In the experiment, the pH value of the water sample was regulated with diluted acetic acid solution or sodium hydroxide solution; the organic substance in the water sample was composed of fulvic acid, which was added according to the experimental demand. Mortality data were pooled and corrected against the mean control mortality using Abbott's formula. For each treatment group, four replicates were undertaken and no oxidant was added to the control jar. The differences in mortality for each treatment were statistically compared using Fisher's least significant difference (LSD) multiple comparison test.

Chlorine dioxide, ozone and chlorine were selected as preferable oxidants for comparison. Then the inactivation effects on *Chironomus kiiensis* larvae by conventional processes alone and in conjunction with pre-oxidation were the subject of further experiments.

RESULTS AND DISCUSSION

Comparison of inactivation effects of different oxidants on *Chironomus kiiensis* larvae

Influence of oxidant dose on inactivation of *Chironomus kiiensis* larvae

Various amounts of oxidants were added into the given volume of distilled water solution free of other extraneous oxidant-demand substances except *Chironomus kiiensis* larvae. Dosages of ozone and chlorine dioxide ranged from 0 to 2.0 mg l^{-1} and that for chlorine from 0 to 10 mg l^{-1}. Figure 1 shows the oxidation effects after 30 min reaction under the conditions of pH 7.0 and 25°C, indicating that the inactivation rate was improved gradually as the oxidant dose was increased. In the case of ozone and chlorine dioxide, 100% of *Chironomus kiiensis* larvae may be inactivated by a lower dose of oxidants and the inactivation capacities of these three oxidants may be ranked as follows: $O_3 > ClO_2 > Cl_2$.

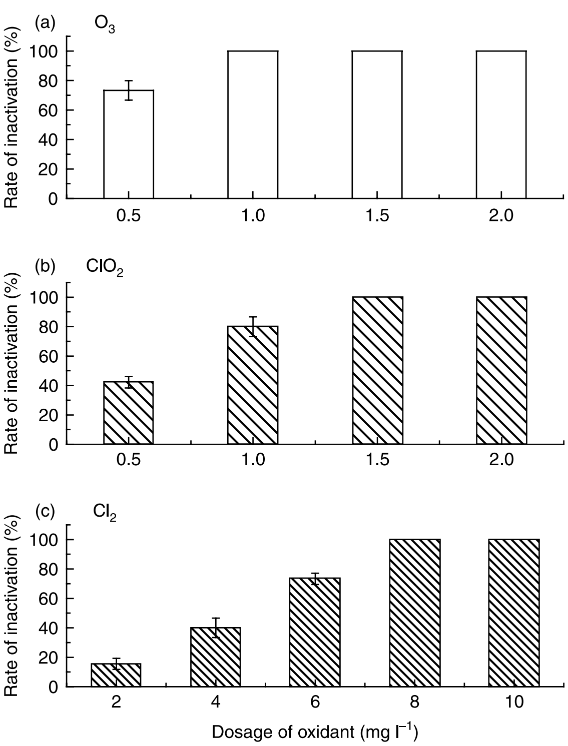

Figure 1 | Inactivation effects of different oxidants on *Chironomus kiiensis* larvae at pH 7.0 and 25°C.

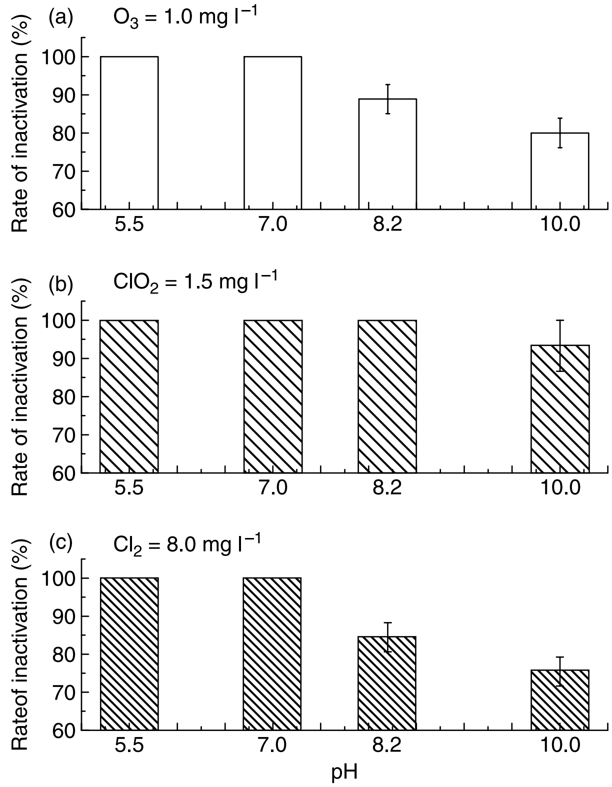

Figure 2 | Effect of pH on inactivation of *Chironomus kiiensis* larvae with different oxidants at 25°C after reaction for 30 min.

Influence of pH value on the inactivation of *Chironomus kiiensis* larvae

To observe the influence of pH value on the inactivation of *Chironomus kiiensis* larvae, the distilled water solution with *Chironomus kiiensis* larvae was selected as the test object. The dosages of chlorine, chlorine dioxide and ozone were 8.0 mg l^{-1}, 1.5 mg l^{-1} and 1.0 mg l^{-1}, respectively, according to the required amount of each oxidant at pH neutral condition with 100% inactivation effect attained (see Figure 1). As shown in Figure 2, the influence of pH on the inactivation rate with chlorine dioxide was not obvious, but the other oxidants were much more sensitive. In the pH range 5.5–8.2, *Chironomus kiiensis* larvae may be killed effectively by chlorine dioxide, whereas pH 10 resulted in an approximate 10% decrease in inactivation rate. Several researchers have speculated that the decreased efficiency of chlorine dioxide at pH 9.0 might be due to the disproportionate reaction of chlorine dioxide occurring under the basic condition (Equation (1)) (Huang *et al.* 1997).

$$2ClO_2 + 2OH^- = ClO_2^- + ClO_3^- + H_2O \quad (1)$$

Similar results of lower inactivation rate of *Chironomus kiiensis* larvae under a higher pH value are presented in this study. With regard to ozone and chlorine, *Chironomus kiiensis* larvae could be killed effectively in the range of pH 5.5–7.0 and the inactivation rate declined rapidly when the pH increased from 7 to 10. The change in the oxidant species present from predominant HOCl at lower pH value to predominant OCl^- at higher pH value, results in the drop of inactivation rate with increased pH (Huang *et al.* 1997). *Chironomus kiiensis* larvae may be effectively inactivated by ozone only at lower pH conditions, which is the direct result of the higher decomposition rate of ozone at higher pH (Facile *et al.* 2000). At pH values of 5.5–10, the adaptability of these methods with respect to pH value may be ranked as follows: $ClO_2 > O_3 > Cl_2$.

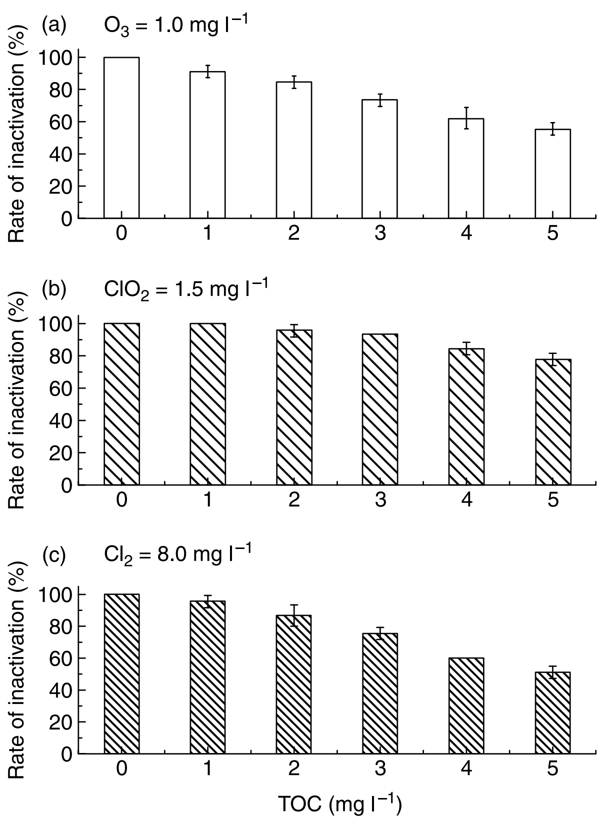

Figure 3 | Effect of TOC on inactivation of *Chironomus kiiensis* larvae with different oxidants at 25°C and pH 7.0 after reaction for 30 min.

Influence of organic matter on the inactivation effect

As shown in Figure 3, the inactivation rate of each oxidant declined when the organic precursor concentration increased, and the influence was most visible in the chlorine tests in comparison with ozone and chlorine dioxide, respectively. The inactivation rate dropped sharply from 100% to 53.3% when TOC concentration ranged from 0 to 5 mg l^{-1} with a chlorine dosage of 8.0 mg l^{-1}. In the chlorine dioxide test, the inactivation rate was steady at around 80% when the organic matter concentration increased to 5 mg l^{-1}. The presence of organics therefore decreased the potential for oxidants to destroy *Chironomus kiiensis* larvae and lowered the inactivation rate. In addition, organic matter in the water not only depleted the available amount of ozone but also accelerated the decomposition efficiency of ozone, thus shortening the virtual reaction times and bringing about a rapid decrease in inactivation rate. According to the result above, the adaptability of these methods for organic content may be ranked as follows: $ClO_2 > O_3 > Cl_2$.

Full-scale study on *Chironomus kiiensis* larvae removal by pre-oxidation

Removal of *Chironomus kiiensis* larvae by conventional water treatment processes

Actual raw water taken from the Shenzhen Water Treatment Plant was used as the test samples. The water quality was as follows (average value): water temperature 24.5°C, turbidity 6.5 NTU, COD_{Mn} 4.36 mg l^{-1}, TOC 3.81 mg l^{-1} and pH 7.1. *Chironomus kiiensis* larvae were added according to the experiment demand. A standard jar test apparatus was used in the coagulation tests; 2 mg l^{-1} of $AlCl_3$ were dosed into each sample as chemical coagulant. After 1 min of rapid mixing at 200 rpm, 10 min of slow mixing at 50 rpm was provided, followed by 20 min of settling. The removal effect on first instar larvae was studied by observing the supernatant fluid, and then the supernatant water after the jar test was filtrated. The filtration velocity was 9 m h^{-1}, the time was 8 h, and the results are given in Table 1, showing that the coagulation and sedimentation process was more efficient for *Chironomus kiiensis* larvae removal than the filtration method.

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Table 1 | Removal results of *Chironomus kiiensis* larvae by conventional water treatment processes

Water treatment process		Original number of larvae (ind. l^{-1})	Final number of larvae (ind. l^{-1})	Removal rate of larvae (%)	Average removal rate of larvae (%)
Coagulation and sedimentation	Group 1	15	4	73.3	
	Group 2	15	4	73.3	75.5
	Group 3	15	3	80	
Filtration	Group 1	4	2	50	
	Group 2	4	1	75	54.5
	Group 3	3	2	33.3	
Combination	Group 1	15	2	86.7	
	Group 2	15	1	93.3	88.9
	Group 3	15	2	86.7	

The removal rate of 75.5% was quite close to that for the combination processes (88.9%), which means that coagulation and sedimentation play the main role in removing *Chironomus kiiensis* larvae in conventional water treatment processes.

Removal of *Chironomus kiiensis* larvae by pre-oxidation in conjunction with conventional processes

In the experiment on pre-oxidation in conjunction with conventional processes, the samples were also assembled in the lab and the water quality was the same as before. The oxidant was added to the water together with the coagulant and the experimental method was the same as before. As shown in Figure 4, the removal rates of *Chironomus kiiensis* larvae improved gradually with the increase in dosage of oxidant regardless of the type of oxidant. Chlorine dioxide proved to be more efficient for *Chironomus kiiensis* larvae removal than ozone. When 100% removal rate of *Chironomus kiiensis* larvae was achieved, the dosage of chlorine dioxide was only $0.6\,mg\,l^{-1}$, whereas $0.9\,mg\,l^{-1}$ was required for ozone. Compared with chlorine dioxide, the influence of organic matter on the inactivation effects of ozone was more visible (see Figure 3). Therefore, ozone could react more easily with many extraneous oxidant-demanding substances in raw water, such as organic compounds, algae, bacteria and protozoan, ahead of inactivating *Chironomus kiiensis* larvae. The extraneous substances were in competition with *Chironomus kiiensis* larvae for ozone, which results in a lower inactivation rate.

Based on the results mentioned above, *Chironomus kiiensis* larvae with stronger vitality could not be completely removed at the lower oxidant dosage, so that the conventional process still could not capture these *Chironomus kiiensis* larvae. Therefore, it was important to inactivate or weaken *Chironomus kiiensis* larvae with adequate available oxidant in order to remove it completely from the water treatment system. In view of the importance of securing drinking water quality, it is necessary to combine oxidant inactivation with conventional processes for removing *Chironomus kiiensis* larvae from raw water, which also reduces the required dosage of the oxidant.

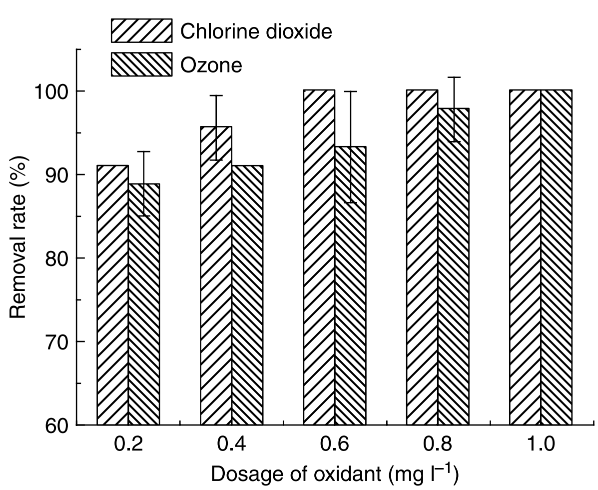

Figure 4 | Removal rate of pre-oxidation in conjunction with conventional processes.

CONCLUSIONS

Chlorine dioxide and ozone proved to be efficient oxidants for *Chironomus kiiensis* larvae inactivation in view of their more visible and stable oxidation effects under variable experimental conditions. This therefore led to enhanced removal of *Chironomus kiiensis* larvae in conventional water treatment processes. To assure the security of drinking water quality, as well as to reduce the dosage of oxidant, it is necessary to combine pre-oxidation with conventional processes when removing *Chironomus kiiensis* larvae from micro-polluted raw water.

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