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Application of Diluted Chlorine Dioxide to Radish and Lettuce Nurseries Insignificantly Reduced Plant Development

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The possible toxicity of a commercial chlorine dioxide preparation (Halox E-100) was evaluated on radish and lettuce seedlings growing in pots under controlled conditions. A single application of various dilutions to radish seedlings growing in a sterile or nonsterile commercial plant substrate only slightly decreased plant dry weight. At the end of the experiments, the plants appeared unaffected by the treatments. Other common plant parameters (root and stem length, number of true leaves) were unaffected or even enhanced. Halox did not reduce the total level of sod bacteria even after four consecutive applications at any dilution rate. In nonsterile soil, high Halox dilution (1:1000) significantly decreased plant dry weight, and the other concentrations (1:10,000; 1:50,000, and 1:100,000) had no apparent effect on the size of the plants. In sterile sod, high concentrations of Halox (1:1000 and 1:10,000) significantly decreased plant growth, but higher dilutions produced no significant reduction in plant dry weight. For radish plants growing in organic matter-free sand only, dilution of 1:10,000 reduced plant growth. On lettuce plants, dilutions from 1:5000 to 1:25,000 did not reduce plant growth. High levels of Halox (1:1000) were toxic to both radish and lettuce seedlings growing in sand and resulted in chlorosis and significant depression of plant growth. Further dilutions of Halox (equivalent to the level used in water disinfection) significantly decreased toxicity for both plant species. Low concentrations of Halox (>1:50,000) had no apparent effect on the appearance of both plant species. In conclusion, this study suggests that chlorine dioxide-treated drinking water can be considered safe for growing plants; this treatment should be further evaluated using other plant species under more realistic growth conditions. ©Academic Press. inc.

INTRODUCTION

Chlorine ions are generally toxic to plants at relatively low concentrations and may cause irreversible damage to plant development (Welch, 1995). Yet, numerous chlorine compounds are used for horticultural and agricultural procedures such as seed disinfection for greenhouse trials (Bashan, 1986a,b; 1991; 1994; Bashan *et al.*, 1978; Bashan and Okon, 1981 b; Dubrovsky *et al.*, 1994), disinfection of plant organs designated for tissue cultures and/or inoculation with beneficial bacterial (Alcaraz-Melendez *et al.*, 1994; Toledo *et al.*, 1995) or for pathogen inspections (Puente and Bashan, 1994; Sharon et al., 1982; Soroker *et* al., 1984), disinfection of working surfaces in fruit packinghouses or in mushroom production facilities (Bashan and Platt, 1980; Brown and Schubert, 1987; Brown and Wardowski, 1985; Roberts and Reymond, 1994), controlling surface phytopathogenic microorganisms in stored plant products (Costilow *et* al., 1984; Hendrix, 1991; Punja and Gaye, 1993; Sholberg and Owen, 1990, 1991; van Doom *et al.*, 1990; Wyatt and Lund, 1981), or as direct agents against phytopathogenic microorganisms (Bashan and Okon, 1981a; Boyette *et* al., 1993; Grech and Rijkenberg, 1992).

The chlorination of drinking water, sewage effluents, or . irrigation water is the most common water disinfection method in the world. It is indisputable that drinking water chlorination represents one of the biggest successes of sanitation ever achieved (Strobel and Dieter, 1990). Commonly used ions such as NaOCI or CaOCI (popular disinfectants in the cleaning industry) are being slowly phased out of the drinking water market because they produced trihalomethanes (THM), various hazardous compounds, and offensive odors when interacting with organic matter (Levi and Jestin, 1988; Suh and Abdel-Rahman, 1985). Still, water disinfection by chlorination is obligatory for potable water, gardening, and sanitary uses (disinfection of fresh vegetables and fruits) (Roberts and Reymond, 1994; Strobel and Dieter, 1990). One of the popular alternatives to common chloride is chlorine dioxide (Bull, 1982; Wilson *et al.*, 1989). It is less toxic, can be buffered, produces no THMs when interacting with organic matter, and has no particular odor or taste (Dietrich *et al.*, 1992; Wondergem and van Dijk-Looijaard, 1991). It is a highly efficient disinfectant against many bacteria which are pathogenic to humans, animals, marine animals, and plants (Berg *et al.*, 1986; Brown and Schubert 1987; Datnoff *et al.*, 1987; Foegeding *et al.*, 1986; Pedersen and Jahromi,1993; Rutala *et al.*, 1991; Tanner, 1989; Wyatt and Lund, 1981), viruses (Harakeh, 1987; Taylor and Butler, 1982), and human parasites (Pesters *et al.*, 1989). It controls microorganisms on reverse osmosis membranes (Adams,

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1990) and biofouling in heat exchanger tubing (Botzenhart *et al*., 1993; Mayack et al., 1984), but may cause corrosion in the stainless-steel tubing used in the dairy industry (Bohner and Bradley, 1991). Still, there are minor health concerns (effects that are not always detected), especially with ingestion of large amounts of chlorine dioxide and/or its metabolites, chlorite and chlorate (Abdel Rahman et al., 1985; Ames and Stratton, 1987; Bercz *et al*., 1982; Couri et al., 1982; Richardson et al., 1994; Tuthill et al., 1982; Zoetman *et* al., 1982), or possible toxicity against marine animals (Chien and Chou, 1989; Hose *et* al., 1989; Puente *et al*., 1992).

The possible harmful by-products of chlorine dioxide are fewer than those when standard chlorination is used (Strobel and Dieter, 1990). Health concerns may be reduced to a minimum by using chemicals to remove chlorine dioxide residues from the disinfected water or reducing the by-products to chloride before water distribution (Griese *et al*., 1991; Lykins *et al.*, 1990; Michael et *al.*, 1981). Currently, commercial chorine dioxide preparations are used on a relatively large scale for the disinfection of drinking water in some countries (Dietrich *et al.*, 1992; Medema *et a1*, 1991; Wilson *et al.*, 1989; Wondergem and van Dijk-Looijaard, 1991). Although it is not its intended purpose, this water is also frequently used to irrigate golf courses, public parks, street vegetation, home gardens, home ornamental plants, and small-scale commercial greenhouses for flowers and vegetables (Bisessar and Mcllveen, 1992). To the best of the authors' knowledge, there is little information on whether this water adversely affects plant growth (Bugbee, 1985; Vijayan and Bedi, 1989).

Halox E-100 (Halox Co., Eugene, OR) is one of the commercial buffered preparations of chlorine dioxide (de Guevara, 1955). It is being used commercially on polluted water in the United States and Mexico at concentration of 1:20001:200,000, as tropical fish medication, and also been used by chiropractors, alternative veterinarians, and many smallscale uses through health-food stores (S. Schulz, personal communication).

Due to shortages in irrigation water in many parts of the world, effluents from water treatment plants are being used for agriculture (Narkis and Kott, 1992; Rav Acha *et al.*, 1995). This water is chlorinated after other conventional water treatments (such as oxidation, mechanical and chemical adsorption of particles) mainly for nonedible crops such as cotton, fiber plants, and flowers. This water clogs modem drip irrigation systems; thus, one of the solutions is to treat it with chlorine dioxide (Rav Acha *et al.*, 1995) which subjects the plants to low levels of chlorine dioxide for prolonged periods. The nonchemical alternative for this water (or even for drinking water) by large-scope microbial filtration or heat disinfection is not cost effective or practical for agriculture.

The aim of this study was to reveal whether water treated with Halox E-100 has any adverse effects on plant growth. This was conducted by evaluating the effect of irrigating two plant species susceptible to chlorine toxicity (radish and lettuce) with diluted Halox E-100.

MATERIALS AND METHODS

Plant Material

Radish (*Raphanus sativus*) cv. Champion (Westar Seed International, El Centro, CA) and lettuce (*Lactuca sativa*) cv. Great Lakes 118 (Westar Seed International) was used for all experiments.

Plant Growth Conditions and Soil Mixtures

All experiments were carried out in a growth chamber (Biotronette Mark III Environmental Chamber, Lab-Line, Melrose Park, IL) or in an indoor, large growth room in pots. The environmental conditions were temperatures of 30 ± 2 or 24 ± 2°C (according to the experiment; Table 1), with light intensity ranging from 71.5 to 121.8 nmol/m^2/sec (according to the various locations of the pots in the growth chambers). Both growth chambers were operating at 12hr photoperiod under constant, noncontrolled, ventilation of fresh air.

Seedlings were grown in black, plastic, rectangular 200mí pots, each containing either 50 g of commercial plant growth mixture or 60 g of soil or 102.5 g of sand-vermiculite mix. The following plant growth substrates were used-(i) a commercial potting mixture (Sunshine Mix 3, Special fine, Fisons Horticulture, Mississauga, Ontario); (ii) agricultural sandy soil (Haplic Xerosol plus Eutric Rhegosol with course texture) obtained from a commercial field near La Paz airport which contained the following percentages: nitrogen, 0.14; phosphorus, 1.68; potassium, 0.3; clay, 10.53; silt, 53.3; sand, 36.2; water field capacity 13.5; CaC03. 0.86; organic matter 2.49, pH 7.5 and conductivity of 0.5 mS/cm; (iii) freshwater-washed beach sand from a public beach 17 km north of La Paz, Baja California Sur, Mexico, treated as follows: sieved to < 1-mm particle size, washed by hand in brackish tap water (2560 µS/cm) 10 times. The sand (approx 10-kg portions) was covered with 10 liters of 1% HCl solution overnight at room temperature (28-33°C). The mixture was then washed 10 times with distilled water (10 kg sand per 10 liters water each time). Different batches of sand were combined when the pH of the last wash was around 7.0 ± 0.5. The wet sand was dried overnight in an oven at 60°C. Then, it was mixed with ground fine vermiculite milled in a household blender at the rate of 100:2.5 (sand:vermiculite, w/w). Prior to the mixing, the vermiculite was similarly treated with acid. The sand originally contained the following: 100% course sand, no N, P, K, clay,

TABLE 1
Setup of the Different Experiments Used to Evaluate the Possible Toxicity of Halox to Plants

Expt	Plant species	Growth substrate	Sterile or nonsterile substrate	Halox dilutions and quantity	Dilutions in PBS buffer	Number of Halox applications	No. of replicates	Duration of the experiment (days)	Temperature (±2°C)	Number of seeds per pot
1	Radish	Sunshine Mix	Nonsterile	1:1000–1:100,000; 25 ml	None	1	10	10	30	2
2	Radish	Sunshine Mix	Sterile and nonsterile	1:1000–1:100,000; 25 ml	None	1	10	10	30	2
3	Radish	Sunshine Mix + agricultural soil	Sterile and nonsterile	1:100–1:100,000; 50 ml + 10 ml each irrigation	None	5	14	18	24	2
4	Radish	Sunshine Mix + agricultural soil	Sterile	1:1000–1:100,000; 50 ml + 10 ml each irrigation	None	5	10	14	24	2
5	Radish + lettuce	Sand + vermiculite	Sterile	1:5000–1:25,000; 25 ml + 10 ml each irrigation	Yes	5	10	15	24	5 (lettuce), 3 (radish)
6	Radish + lettuce	Sand + vermiculite	Sterile	1:5000–1:25,000; 25 ml + 10 ml each irrigation	Yes	5	10	15	24	5 (lettuce), 3 (radish)

silt, or measurable amounts of organic matter, water field capacity of 9.8%, $CaCO_3$, 4.29%, pH of 73 and conductivity of 0.77 mS/cm. The water-holding capacity of the sand:vermiculite mixture was 24.39%; (iv) a mixture of the above agricultural soil with Sunshine Mix 3 (described above) at the rate of 3:1 (w/w).

The different plant growth substrates used were either nonsterilized or sterilized by tyndelization procedure. The substrate was placed in plastic bags containing approximately 2 kg each. Then, they were autoclaved three times for 1 hr each at 15 psi. Between each sterilization they were incubated for 24 hr at ambient temperature. Because the pots were not steam resistant, they were first washed with commercial dish detergent, thoroughly rinsed with drinking water, and then surface disinfected by spraying with 70% (in water) of technical alcohol. The sterile substrate was placed in the pots in a disinfected room (disinfected by spraying with 70% technical alcohol on all surfaces). Pots were immediately sown with seeds.

Radish seeds containing the commercial fungicide treatment (Thiram) were used without any treatment or, alternatively, they were washed 10 times for I min each under constant agitation with distilled water. The last two washings were performed with sterile distilled water. Radish seeds were sown without drying.. Lettuce seeds were sown without any special treatment.

Chlorine Dioxide Application

A commercial preparation of chlorine dioxide (Halox E100, Halox Co.) which is stable in freshwater (Halox Co., unpublished data) and seawater (Puente et al., 1992) was used. It contains approximately 26 mg/ml of active chlorine (Hager, unpublished data) and is used mainly for drinking and sewage water disinfection.

Halox preparation of chlorine dioxide was applied either after dilution with distilled water (Arrowhead, Los Angeles, CA) (Experiments 2-4) without controlling the pH [which varied within the range of 5.42 (no Halox) to 9.85 (1:10 dilution)], or after dilution with a mixture containing 0.0165 M phosphate buffer supplemented with 0.15 MNaCI (PBS) and 1/5 Hoagland's plant nutrient solution (Experiments 5-6). In Experiment 1, the Shieve's plant nutrient solution (Salisbury and Ross, 1985) was used instead of Hoagland's. The pH of all mixtures varied according to the Halox concentration and was within the range of 5.28 (no Halox) to 6.1 (highest concentration). The conductivity of the solutions was 0.25 to 200 (for water dilutions) and from 114 to 130 $\mu mhos/cm^2$ (for buffered solutions). Dilutions used ranged from 1:1 to 1:100,000 corresponding to 13,000 to 0.26 µg active chlorine/ml in the preparation. Halox was applied to plants by direct irrigation on the "soil" surface. Although the chlorine level could be measured in the dilutions by a "chlorine test kit" (Hach, Loveland, U.S.A.), after application to soils, it was

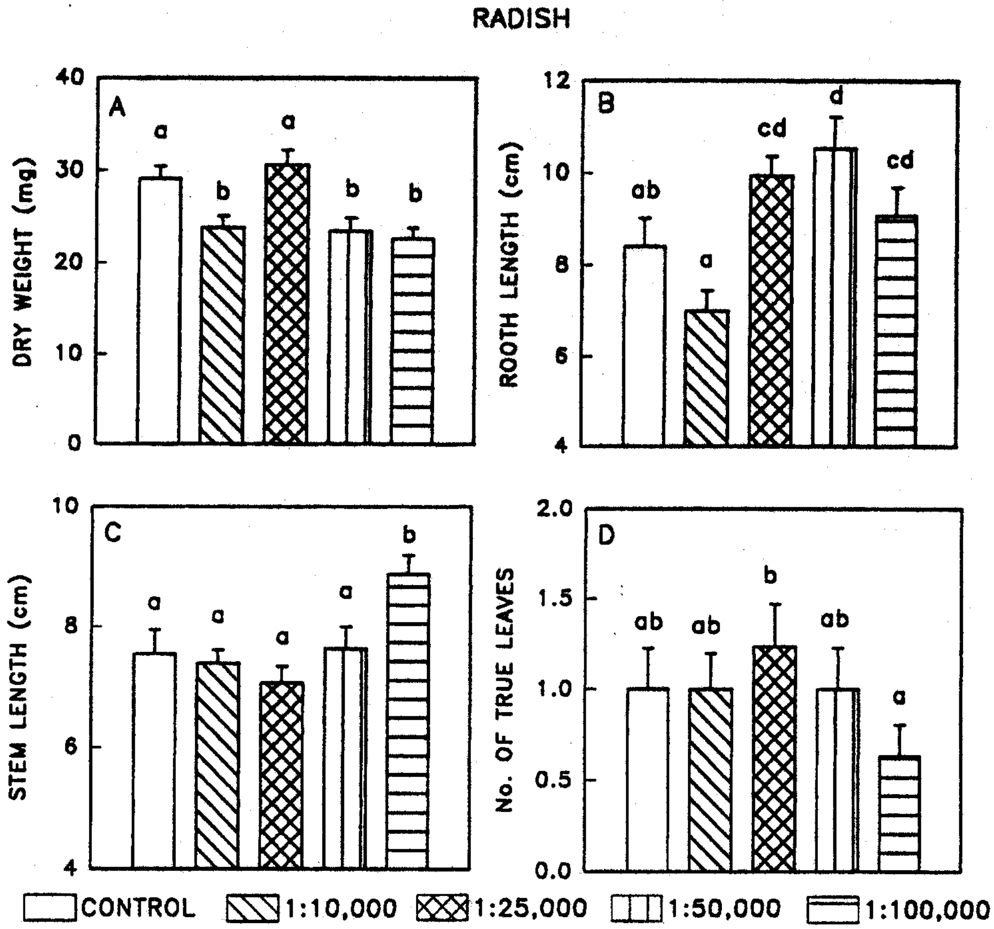

FIG. 1. Effect of a single application of Halox on radish seedlings growing in nonsterile plant growth substrate. A, dry weight; B, root length; C, stem length; D, number of true leaves. Bars represent SE. Columns denoted by a different letter in each subfigure differ significantly at $P \leq 0.05$ by one-way ANOVA.

impractical to measure residual chlorine because of its different adsorption to the organic matter in each soil batch. A single application of Halox was used in two experiments. In other experiments, Halox was applied in 3-day intervals and up to five applications.

Toxicity Index

In many cases the toxic effect of Halox did not have an effect on the plant dry weight, but some discoloration could be detected by the naked eye on some of the plants. A quantitative analysis of these effects was done by a visual index where:

0 = No apparent effect; healthy plant

1 = Slightly affected, 1-2 leaves yellowing

2 = Yellow leaves, damage to the stem, visible reduction in plant size

3 = Significant reduction in plant size

4 = Dead plants, very deformed plants or almost a complete inhibition of plant growth.

Each seedling of the entire population was analyzed separately, and the average cumulative values of all the plants in a particular treatment represent the "toxicity index" of the treatment.

Bacterial Count from Soil after Halox Treatments

Each soil sample was diluted to a level of 10^{-6} in 0.85% NaCI. The first soil dilution contained 1 g of soil and 99 ml of a saline solution in a flask with 20 glass boiling beads (3 mm in diameter, VWR, Los Angeles, CA). The mixture was shaken for 10 min at 200 rpm in an orbital shaker and then decimally diluted in saline without letting the soil particles to settle.

Bacteria were counted on soil extract agar (SEA; Wollum, 1982) by a conventional plate agar method (mixing 1 ml sample with 15 ml of SEA medium maintained at 48°C and immediately spreading the mixture on a petri plate). Each sample was duplicated and incubated for 48 hr at 30 ±2°C. Colonies were counted by a colony counter (Manostat, New York, NY). SEA medium contained 1000 ml soil extract, 1 g/liter glucose, 5 g/liter yeast extract, 0.2 g/liter K_2HPO_4, 15 g/liter agar. Final pH was 6.8-7.0. Soil extract for this medium was prepared as follows: 500 g of soil was thoroughly mixed in 1000 ml of tap water. The mixture was autoclaved for 1 hr and then filtered through a Whatman 5 filter paper, and the volume was adjusted to 1 liter with tap water. To this, ail the other ingredients were added and the new mixture was autoclaved for 20 min.

Experimental Design and Statistical Analysis

All experiments were designed by a random fashion and contained 10 replicates each. Each experiment was repeated at least twice. A replicate was considered a single pot containing several seedlings as detailed in Table 1. Controls for the experiments, although varying between the experiments, included irrigation with nutrient solution, water, or mixtures of buffers and plant nutrient solution.

All results were statistically analyzed by one-way analysis of variance (ANOVA) at $P \leq 0.05$ using Statgraphics software 5.1 (Graphic Software Systems, Rockville, MD) and were drawn by Sigma Plot (Jandel, San Rafael, California) software 5.0. Results presented are from representative experiments demonstrating similar trends.

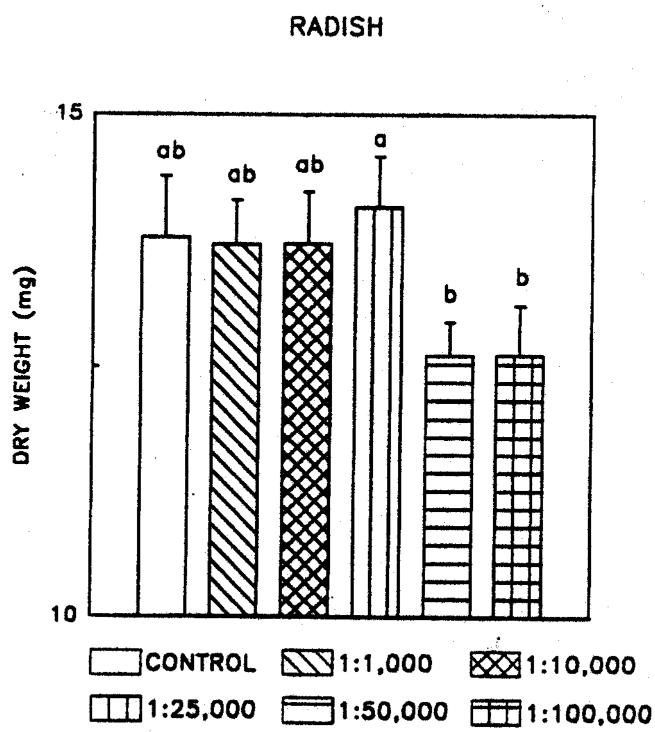

FIG. 2. Effect of a single application of Halox on the dry weight of radish seedlings growing in sterile plant growth substrate. Bars represent SE. Columns denoted by a different later differ significantly at $p \leq 0.05$ by one-way ANOVA.

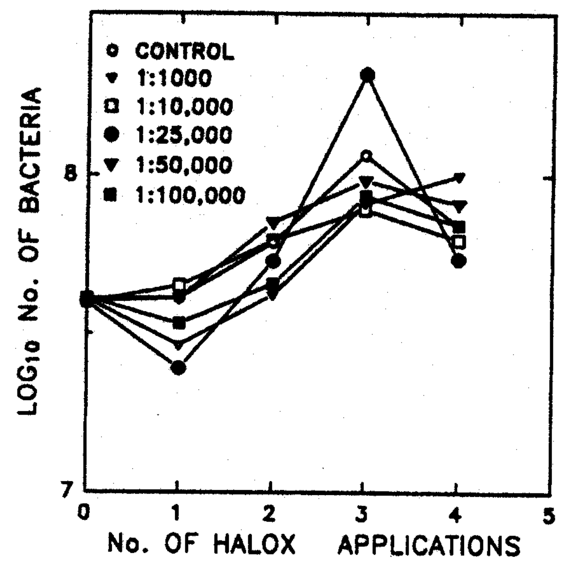

FIG. 3. Effect of multiple applications of various dilutions of Halox on the total number of soil bacteria in agricultural soil.

RESULTS

Effect of a Single Application of Halox on Radish Seedlings Growing in Nonsterile Plant Growth Substrate (Experiment 1, Table I)

A single application of various dilutions of Halox (1:10,000-1:100,000) to radish seedlings growing in a commercial plant substrate only slightly, but significantly, decreased plant development. One dilution (1:25,000) slightly, but nonsignificantly, increased plant growth (about 4% increase). At the end of the experiment the plants looked unaffected (Fig. IA). Other measured plant parameters were as follows: root length, while not negatively affected at 1:10,000 dilution, significantly increased at lower Halox levels (dilutions >25,000) (Fig. 1B). Stem length, although nonaffected by any concentration of Halox, significantly increased in the lowest concentration (1:100,000) (Fig. 1 C). The number of true leaves was not negatively affected (Fig. 1D).

Effect of a Single Application of Halox on Radish Seedlings Growing in Sterile Substrate (Experiment 2, Table I)

Similar to Experiment 1, Halox had only a marginally decreasing effect on the development of radish seedlings growing in sterile commercial plant growth substrate (Fig. 2). Dilution of 1:25,000 did not decrease plant growth (but a nonsignificant increase of about 2% was observed).

Effect of Multiple Applications of Halox on Radish Seedlings Growing in Sterile and Nonsterile Agricultural Soil (Experiment 3, Table 1)

Halox did not reduce the level of soil bacteria after even four consecutive applications at any dilution rate (Fig. 3).

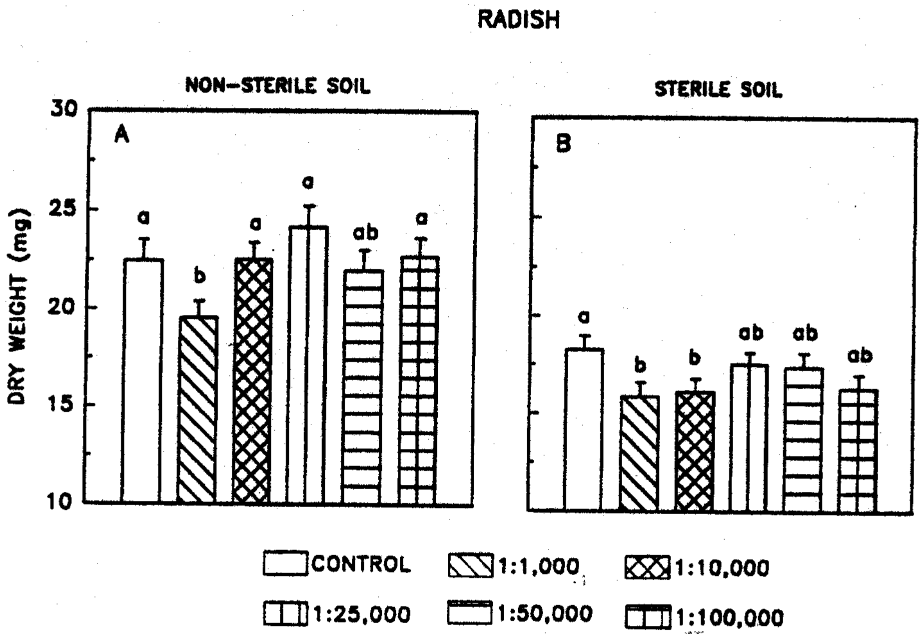

FIG. 4. Effect of multiple applications of Halox on radish seedlings growing in sterile and nonsterile agricultural soil. Bars represent SE. Columns denoted by a different letter in each subfigure separately, differ significantly at $P \leq 0.05$ by one-way ANOVA.

In nonsterile soil, Halox dilution of 1:25,000 increased plant growth, but only by about 8%. High Halox dilution (1:1000) significantly decreased plant growth, and the other concentrations (1:10,000; 1:50,000, and 1:100,000) had no apparent effect on the dry weight of the plants (Fig. 4A). In sterile soil (Experiment 4, Table 1) high concentrations of Halox (1:1000 and 1:10,000) significantly decreased plant growth, but higher concentrations had no significant reduction on plant development (Fig. 4B).

Effect of Multiple Applications of Halox on Radish and Lettuce Seedling Dry Weight Growing in Sterile Sand without Organic Matter (Experiments 5 and 6, Table I)

To avoid interference of organic matter interaction with chlorine ions, the experiments were conducted in pure sand and vermiculite mixtures. Application of Halox to radish reduced plant dry weight only in the 1:10,000 dilution. Other dilutions and also application of Halox on lettuce plants in dilutions ranging from 1:5000 to 1:25,000 did not reduce plant dry weight (Figs. 5A and 5B).

Effect of Halox on the Appearance of Radish and Lettuce Seedlings

High concentrations of Halox (1:1000) were toxic to both radish and lettuce seedlings and resulted in chlorosis and significant depression of plant size (Figs. 6A and 6B). Dilutions of Halox significantly decreased toxicity in both plant species. Yet, when a large plant population was analyzed, small adverse toxicity was observed. Low concentrations of Halox (> 1:50,000) had no apparent effect on the appearance of both plant species (data not provided).

DISCUSSION

Agriculture is the biggest freshwater consumer on Earth, overwhelming all other uses. In California, for example, agriculture consumes 78% of all available water leaving domestic, industrial, and environmental needs to vie for the remainder (Conniff and Rickman, 1993). In less-developed countries this proportion is even higher (Graves, 1993). There is a clean water crisis in the world, where 40% of its population experiences chronic water shortage (about 1 billion people live without pathogen-free drinking water), and 10 million annual deaths are related to diseases from contaminated water (Hager, 1995). Fortunately, the majority of the population enjoys sanitary water, largely due to the use of chlorine compounds (Strobel and Dieter, 1990). Water for agricultural irrigation does not need to be disinfected because plants are not affected by human- and animal-related pathogens. Nevertheless, large amounts of plants are irrigated with chlorinated drinking water. Furthermore, because the use of chlorinated urban and industrial sewage water is growing in agriculture (Narkis *et al.*, 1995; Rav Acha *et al.*, 1995), more plants are being exposed to chlorine compounds. Surprisingly, the literature on water purification does not deal with related effects, even though there is sufficient knowledge about the various adverse effects of chlorine compounds on plant material (Welch, 1995).

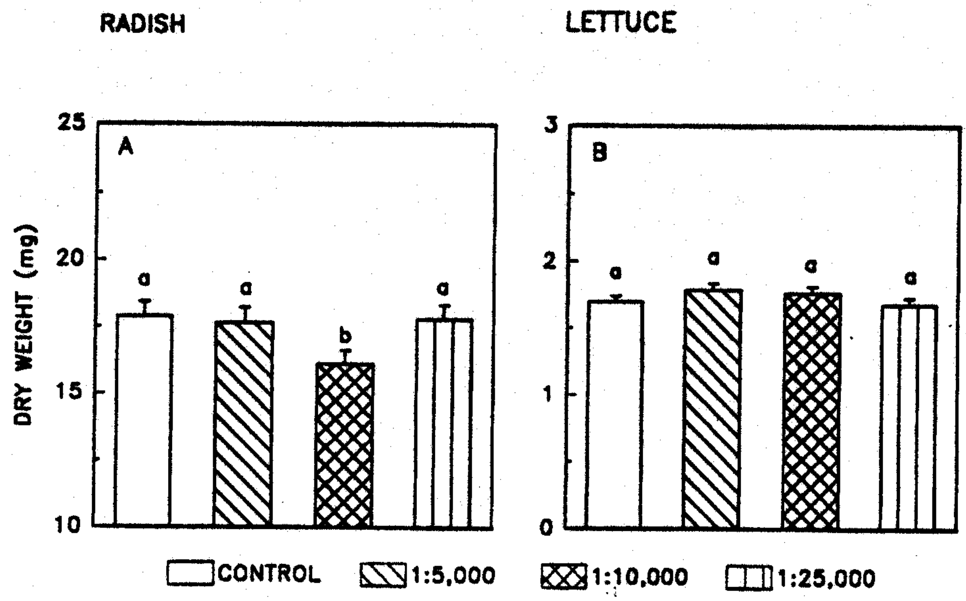

FIG. 5. Effect of multiple applications of Halox on radish and lettuce seedling dry weight growing in sterile sand without organic matter. Bars represent SE. Columns denoted by a similar letter in each subfigure separately, do not differ significantly at $P \leq 0.05$ by one-way ANOVA.

Chlorine dioxide is being considered for water treatment in Mexico; therefore, it is inevitable that some of this water will be used to irrigate plants in urban zones and in rural agricultural area where economical constrains require that one water system serves both needs. Thus, the aim of this study was to assess possible risks for plant development after irrigation with chlorine dioxide-treated water.

Using two chlorine-susceptible plant species, it was found that the plants were not able to tolerate high levels of Halox in the irrigation water. However, the plants did tolerate low levels corresponding to the actual dilutions used for disinfecting drinking water. The higher the dilution, the smaller the effect that could be detected in the plants.

From these results alone, one cannot draw final conclusions on

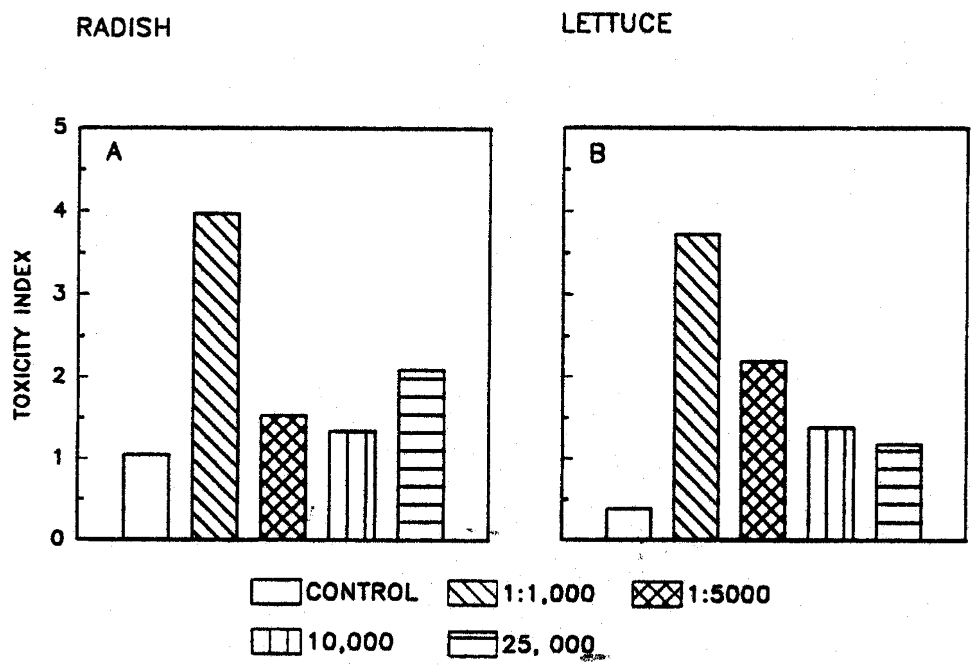

FIG. 8. Effect of Halox on radish and letuce seedlings appearance. Results are the average of two experiments. Dilution 1:1000 in radish represents results from a single experiment.

the general safety of Halox application on plants. (i) The model plants were chosen more for their susceptibility to the disinfectant rather than their economic importance (which is marginal in Mexico). There are numerous, more economically important plants to be tested such as beans, corn, peppers, and tomatoes. (ii) The experiments were conducted under controlled conditions and not under field conditions. (iii) Halox was applied by manual irrigation using clean, organic matter-free equipment, and not through the commonly dirty agricultural irrigation systems which may deactivate it. (iv) In these experiments Halox was not exposed to the field UV irradiation that might eliminate or reduce its effects on plants. (v) Only seedlings were used, and the long-term effects (if any) on more mature plants were not studied. (vi) The experiments, despite their repetitions and many replications, were, in fact, on a small scale. Despite the above reservations, this study demonstrates that Halox-treated drinking water is not hazardous to plants.

CONCLUSION

In conclusion, this study revealed that chlorine dioxide-treated drinking water should be considered safe for growing plants. For more conclusive recommendations, more commercial plant species and long-term effects should be tested under greenhouse and field conditions with agricultural hardware.

ACKNOWLEDGMENTS

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REFERENCES

Abdel-Rahman, M. S., Couri, D., and Bull, R J. (1985). Toxicity of chlorine dioxide in drinking water. J. *Environ Pathol Toxicol. Oncol. Of. Organ.* **6**, 105-113.

Adams, W. R (1990). The effects of chlorine dioxide on reverse osmosis membranes. *Desalination* **78**, 439-453.

Alcaraz-Melendez, L., Real-Cosio, S., and Bashan, Y. (1994). Domistication of micropropagated plants of the spice damiana *(Turnera diffusa). Plant Cell Rep.* **13**, 679-683.

Ames, R G., and Stratton, J. W. (1987). Effect of chlorine dioxide water disinfection on hematologic and serum parameters of renal dialysis patients. *Arch. Environ. Health* **42**, 280-285.

Bashan, Y. (1986x). Enhancement of wheat roots colonization and plant development by *Azospirillum brasilense* Cd following temporary depression of the rhizosphere microflora. *Appl. Environ. Microbiol.* **51**, 1067-1071.

Bashan, Y. (1986b). Alginate beads as synthetic inoculant carriers for the slow release of bacteria that affect plant growth. *Appl. Environ. Microbial.* **51**, 1089-1098.

Bashan. Y. (1991). Air-borne transmission of the rhizosphere bacterium *Azospirillum. Microb. Ecol.* **22**, 257-269.

Bashan, Y. (1994). Symptom expression and ethylene production in leaf blight of cotton caused by *Alternaria macrospora* and *Alternaria alternata* alone and in combination. *Can. J. Bot.* **72**, 1574-1579.

Bashan, Y., and Okon, Y. (1981 a). Integrated control of bacterial blotch in Israel. *Mushroom J.* **97**,29-33.

Bashan, Y., and Okon, Y. (1981b). Inhibition of seed germination and development of tomato plants in soil infested with *Pseudomonas tomato. Ann. Appl. Biol.* **98**, 413-417.

Bashan, Y., Okon, Y., and Henis, Y. (1978). Infection studies of *Pseudomonass tomato*, causal agent of bacterial speck of tomato. *Phytoparasitica* **6**, 135-145.

Bashan, Y., and Platt, M. (1980). Increased yield of the cultivated mushroom in Israel by artificial misting. *HortScience* **15**, 263-264.

Bercz, J. P., Jones, L., Garner, L., Murray, D., Ludwig, D. A., and Boston, J. (1982). Subchronic toxicity of chlorine dioxide and related compounds in drinking water in *the nonhuman* primate. *Environ. Health Perspect* **46**, 47-55.

Berg, J. D., Roberts, P. V., and Martin, A. (1986). Effect of chlorine dioxide in selected membrane fractions of *Escherichia coli. J. Appl. Bacterial.* **60**, 213-220.

Bisessar, S., and Mcllveen, W. D. (1992). Effects of swimming pool sanitizing chemicals on turf grass. *Bull. Environ Contain. Toxicol.* **49**, 295-299.

Bohner, H. F., and Bradley, R L. (1991). Corrosivity of chlorine dioxide used as sanitizer in ultrafiltration systems. *J. Dairy Sci.* **74**, 3348-3352.

Botzenhart, K., Tarcson, G. M., and Ostruschka, M. (1993). Inactivation of bacteria and coliphages by ozone and chlorine dioxide in a continuous flow reactor. *Water Sci. Technol. J.* **27**, 363-370.

Boyette, M. D., Ritchie, D. F., Carballo, S. 1., Blankenship, S. M., and Sanders, D. C. (1993). Chlorination and postharvest disease control. *Hort-Technology* **3,** 395-400.

Brown, G. E., and Schubert, T. S. (1987). Use *of Xanthomonas campestris* pv. *vesicatoria* to evaluate surface disinfectants for canker quarantine treatment *of* citrus fruit *Plant Dis.* **71**, 319-323.

Brown, G. E., and Wardowski, W. F. (1985). Use *of* chlorine and chlorine dioxide in Florida citrus packinghouses to reduce inoculum of dacay pathogens. *Proc. Ann. Meet Fla. Stat Horticult. Son* **97**, 97-100.

Bugbee, G. J. (1985). Does chlorine in tape water cause damage to houseplants. *Front. Plant Sci. Conn. Agric. Exp. Stn. New Haven* **38**, 6-7.

Bull, R. J. (1982). Health effects of drinking water disinfectants and disinfectant by-products. *Environ. Sci. Technol.* **16**, A554-A559.

Chien, Y. H., and Chou, Y. H. (1989). The effects of residual chlorine, temperatures and salinities on the mortality of larval milkfish *Chanos chanos. J. Fish Son Taiwan* **16**, 271-280.

Conniff, R., and Rickman, R (1993). California: Desert in disguise. In Water, the Power, Promise, and Turmoil of North America's Fresh Water (W. Graves, Ed.), 13th special ed., pp. 38-53. National Geographic Society, Washington DC.

Costilow, R N., Uebersax, M. A., and Ward, P. J. (1984). Use of chlorine dioxide for controlling microorganisms during the handling and storage of fresh cucumbers. *J. Food* Sci. 49, 396-401.

Couri, D., Abdel-Rahman, M. S., and Bull, R J. (1982). Toxicological effects of chlorine dioxide, chlorite and chlorate. *Environ. Health Perspect* **46**, 13-18.

Datnoff, L. E., Kroll, T. K., and Lacy, G. H. (1987). Efficacy of chlorine for decontaminating water infested with resting spores of *Plasmodiophora brassicae. Plant Dis.* **71**, 734-736.

de Guevara M. L. (1955). Aqueous chlorine dioxide antiseptic compositions and production thereof. U.S. Patent 2 701 781.

Dietrich, A. M, Orr, M P., Gallagher, D. L., and Hoehn, R. C. (1992). Tastes and odors associated with chlorine dioxide. *Am. Water Works Assoc. J.* **84**, 82-88.

Dubrovsky, J. G., Puente, M. E., and Bashan, Y. (1994). *Arabidopsis thaliana* as a model system for the study of the effect of inoculation by *Azospirillum brasilense* Sp-245 on root hairs growth. *Soil Biol. Biochem.* **26**, 1657-1664.

Foegeding. P. M., Hemstapat, V., and Giesbrecht, F. G. (1986). Chlorine dioxide inactivation of Bacillus and *Clostridium* spores. *J. Food Sci.* **51**, 197-201.

Graves, W. (1993). Introduction. In *Water, the Power, Promise, and Turmoil of North America's Fresh Water (W.* Graves, Ed), 13th special ed., p. 1. National Geographic Society, Washington DC.

Grech, N. M., and Rijkenberg, F. H. J. (1992). Injection of electrolytically generated chlorine into citrus microirrigation systems for the control of certain waterbome root pathogens. *Plant Dix.* **76**, 457-461.

Griese, M H., Hauser, K., Berkemeier, M., and Gordon, G. (1991). Using reducing agents to eliminate chlorine dioxide and chlorite ion residuals in drinking water. *Am. Water Works Assoc. J.* **83**, 56-61.

Hager, M. (1995). Nor any drop to drink. *Newsweek* **August 14**, 52.

Harakeh, S. (1987). The behavior of viruses on disinfection by chlorine dioxide and other disinfectants in effluent. *FEMS Microbial. Lett.* **44**, 335-341.

Hendrix, F. F., Jr. (1991). Removal of sooty blotch arid flyspeck from apple fruit with a chlorine dip. *Plant Dis* **75**, 742-743.

Hose, J. E., Di-Fiore, D., Parker, H. S., and Sciarrotta, T. (1989). Toxicity of chlorine dioxide to early life stages of marine organisms. *Bull. Environ. Contam. Toxic.* **42**, 315-319.

Levi, Y., and Jestin, J. M. (1988). Offensive tastes and odors occurring after chlorine addition in water treatment processes. *Water Sci. Technol* **20**, 269-274.

Lykins, B. W., Goodrich, J. A., and Hoff, J. C. (1990). Concerns with using chlorine dioxide disinfection in the USA. *Aqua (Oxford)* **39**, 376-386.

Mayack, L. A., Soracco, R, Wilde, E. W., and Pope, D. H. (1984). Comparative effectiveness of chlorine and chlorine dioxide biocide regimes for biofouling control. *Water Res.* **18**, 593-599.

Medema, G. J., Wondergem, E., van Dijklooyaard, A. M., and Havelaar, A. H. (1991). Effectivity of chlorine dioxide to control *Aeromonas* in drinking water distribution systems. *Water Sci. Technol. J.* **24**, 325-326.

Michael, G. E., Miday, R K., Bercz, J. P., Miller, R. G., Greathouse, D. G., Kraemer, D. F., and Lucas, J. i3. (1981). Chlorine dioxide water disinfection: A prospective epidemiology study. *Arch. Environ. Health* **36**, 20-27.

Narkis, N., Armon, R., Offer, R, Orshansky, F., and Friedland, E. (1995). Effect of suspended solids of wastewater disinfection efficiency by chlorine dioxide. *Water Res.* **29**, 227-236.

Narkis, N., and Kott, Y. (1992). Comparison between chlorine dioxide and chlorine for use as a disinfectant of wastewater effluents. *Water Sci. Technol J.* **26**, 1483-1492.

Pedersen, K., and Jahromi, N. (1993). Inactivation of bacteria with SMAC-A stable solution of chlorine dioxide in water. *Vatten* **49**, 264-270.

Peeters, J. E., Mazas, E. A., Masschelein, W. J., Vill acorta Martinez de Maturana, I., and Debacker, E. (1989). Effect of disinfection of drinking water and ozone or chlorine dioxide on survival of *Cryptosporidium parvum* oocysts. *Appl Environ. Microbial.* **55**, 1519-1522.

Puente, M.- E., and Bashan, Y. (1994). The desert epiphyte *Tillandsia recurvata* harbours the nitrogen fixing bacterium *Pseudomonas stutzeri*. *Can. J. Bot* **72**, 406-408.

Puente, M E., Vega-Villasante, F., Holguin, G., and Bashan, Y. (1992). Susceptibility of the brine shrimp Artemis, and its pathogen *Vibrio parahemolyticus* to chlorine dioxide in contaminated sea-water. J. AWL *Bacteriol* **73**, 465-471.

Punja, Z. K., and Gaye, M. M (1993). Influence of postharvest handling practices and dip treatments on development of black root rot on fresh market carrots. *Plant Dis.* **77**, 989-995.

Rav Acha, C., Kummel, M., Salamon L, and Adin, A. (1995). The effect of chemical oxidants on effluent constituents far drip irrigation. *Water Res.* **29**, 119-129.

Richardson, S. D., Thruston, A. D., Jr., Collette, T. W., Patterson, K. S., Majetich, G., and Zhang, Y. (1994). Multispectral identification of chlorine dioxide disinfection byproducts in drinking water. *Environ. Sci. Technol.* **28**, 592-599.

Roberts, R G., and Reymond, S. T. (1994). Chlorine dioxide for reduction of postharvest pathogen inoculum during handling of tree fruits. Appl. Environ. Microbial. **60**, 2864-2868.

Rutala, W. A., Cole, E. C., Wannamaker, N. S., and Weber, D.1. (1991). Inactivation of Mycobacterium tuberculosis and Mycobacterium bovis by 14 hospital disinfectants. Am. J. Med. **91**, 2675-2715.

Salisbury, F. B., and Ross, C. W. (1985). Mineral nutrition. In *Plant Physiology*, pp. 96-113. Wadsworth, Belmont, CA.

Sharon, E., Okon, Y., Bashan, Y., and Henis, Y. (1982). Detached leaf enrichment: a method for detecting small numbers of Pseudomonas syrin*gae pv.* tomato and *Xanthomonas campestris* pv. *vesicatoria* in seeds and symptomless leaves of tomato and pepper. *J. Appl Bacteriol.* **53**, 371-377.

Sholberg, P. L., and Owen, G. R (1990). Populations of propagules of *Penicillium* spp. during immersion dumping of apples. *Can. Plant Dis* Surv. **70**, 11-14.

Sholberg, P. L., and Owen, G. R (1991). Populations of propagules of *Mucor* spp. during immersion dumping of Anjou pears. *Can. Plant Dis Surv.* **71**, 33-35.

Soroker, E., Bashan, Y., and Okon, Y. (1984). Reproducible induction of cavity spot in carrots and physiological and microbial changes occurring during cavity formation. *Soil Biol. Biochem.* **16**, 541-548.

Strobel, K., and Dieter, H. H. (1990). Toxicological risk/benefit aspects of drinking water chlorination and of alternative disinfection procedures. *Z. Wasser Abwasser Forsch.* **23**, 152-162.

Suh, D. H., and Abdel-Rahman, S. (1985). Mechanism of chloroform formation by chlorine and its inhibition by chlorine dioxide. *Fundam. Appl. Toxicol.* **5**, 305-313.

Tanner, R. S. (1989). Comparative testing and evaluation of hard-surface disinfectants. *J. Ind Microbial.* **4**, 145-154.

Taylor, G. R, and Butler, M. (1982). A comparison of tire virucidal properties of chlorine, chlorine dioxide, bromine chloride and iodine. *J. Hyg.* **89**, 321-328.

Toledo, G., Bashan, Y., and Soeldner, A. (1995). In vitro colonization and increase in nitrogen fixation of seedling roots of black mangrove inoculated by a filamentous cyanobacteria. *Can. J. Microbial.* **41**, 1012-1020.

Tuthill, R W., Giusti, R A., Moore, G. S., and Calabrese, E. J. (1982). Health effects among newborns after prenatal exposure to $C1O_2$-disinfected drinking water. *Environ Health Perspect.* **46**, 39-45.

van Doom, W. G., de-Witte. Y., and Perik, R R J. (1990). Effect of antimicrobial compounds on the number of bacteria in stems of cut rose flowers. J. Appl. Bacteriol. **68**, 117-122.

Vijayan, R, and Bedi, S. J. (1989). Effect of chlorine pollution on three fruit tree species at Ranoli near Baroda, India *Environ.* Pollut. **57**, 97-102.

Welch, R M. (1995). Micronutrient nutrition of plants. Crit *Rev. Plant* Sci. **14**, 49-82.

Wilson, A. E., Boyd-Jones, I., and Bedford, G. K (1989). Chlorine dioxide disinfection of potable water for taste and odour control. in *water in the 1990s*: *Annual Conference, 1989*, pp. 115-138. New Zealand Water Supply and Disposal Association, Hamilton, New Zealand.

Wollum, A. G. (1982). Cultural methods for soil microorganisms. In *methods of soil analysis*. Part 2, chemical and microbiological properties (A. L. Page, Ed.), 2nd ed., pp. 781-801. American Society of Agronomy , Madison, WI.

Woondergem E., and van Dijk-Looijaard, A. M. (1991). Chlorine dioxide as a post-disinfectant for Dutch drinking water. Sci. *Total Environ.* **102**, 101-112.

Wyatt, G. M., and Lund, B. M. (1981). The effect of antibacterial products on bacterial soft rot of potatoes. *Potato Res.* **24**, 315-329.

Zoeteman B. C. J., Hrubec, J., de-Greef, E., and Kool, H. J. (1982). Mutagenic activity associated with by-products of drinking water disinfection by chlorine, chlorine dioxide, ozone and UV-irradiation. *Environ. Health* Perspect. **46**, 197-206.