Efficiency Comparison between Chlorine and Chlorine Dioxide to Control Bacterial Regrowth in Water Distribution System

Yoon-jin Lee[†]

Department of Environmental Engineering, Cheongju University, 36, Naedok-dong, Sangdang-gu, Cheongju, Chungbuk 360-764, Korea (Received May 19, 2006/Accepted August 9, 2006)

ABSTRACT

This study investigated the inactivation of the total coliform, an indicator organism in chlorine and chlorine in order to control microbial regrowth for water distribution systems and select an appropriate disinfection strategy for drinking water systems. The disinfection effects of chlorine and chlorine dioxide with regard to the dosage of disinfectant, contact time and DOC was investigated experimentally. In spite of the consistency of chlorine residuals at approximately 0.2 mg/l, bacteria regrowth was detected in the distribution system and it was confirmed by the scanning electron microscope results. The influence of organic carbon change on the killing effect of chlorine dioxide was strong.

Keywords: chlorine, chlorine dioxide, water distribution system, total coliform

I. Introduction

Chlorine has traditionally been the most effective guard against waterborne pathogenic organisms due to its advantages.¹⁾ Thus, chlorine has been the primary disinfectant used in Korea. Despite the fact that newly emerging pathogenic protozoa, such as *Giardia* cysts and *Cryptosporidium oocysts*, are clearly resistant to the conventionally used dose of chlorine, chlorine remains the most commonly used disinfectant.^{2,3)}

In 1974, when chlorination of portable water was found to produce THMs (trihalomethanes) and one of these by-products, chloroform, resulted in an increase in the incidence of tumors, the safety of chlorine disinfection was questioned. In order to comply with the regulations of trihalomethane⁴⁾, many portable water utilities were forced to alter their treatment methods. These utilities had the option to use alternative disinfectants, such as chlorine dioxide, or ozone. When either chlorine dioxide or ozone is used, trihalomethanes are not produced, and there are also fewer nonvolatile

halogenated organics produced. As active chemicals, these disinfectants may react with aquatic organic substances to form other by-products. The interest in using chlorine dioxide and ozone to disinfect portable water has increased, along with the desire to investigate how it affects taste, odor control, and oxidation of iron and manganese.⁵⁾

To safely supply tap water requires a reliable distribution system, especially due to the risk of pipe corrosion and contamination by microorganisms as the purified water runs through the system.^{6,7)} To ensure the supply of quality water, systematic control of the distribution system is required. This process involves not only the process of water treatment and quality control of source water, but also the process of transporting water from the treatment plant to the consumer's tap.^{8,9)} Many reports indicate that the excessive presence of bacteria in tap water, which was not detected at the water treatment, is caused by the regrowth of bacteria after the water has left the plant.¹⁰⁻¹²⁾

The sudden increase in the number of organisms coming into the portable water through distribution systems from the water treatment plant is known as "re-growth" or "after-growth" of microorganisms.¹³⁾ Brazos and O'Conner (1989) define re-growth as the recovery of bacteria in water supply pipes as a

[†]Corresponding author: Department of Environmental Engineering, Cheongju University

Tel: 82-43-229-8571, Fax: 82-43-229-8571

E-mail: yjlee@cju.ac.kr

result of the damage to microorganisms by the disinfectant.¹⁴⁾ They define after-growth as a proliferation of settled microorganism on the surface of pipe. Re-growth of microorganisms occurs under specific favorable conditions of growth within the distribution systems, direct inflow from the outside, or when bio-films form on the inner wall of the water supply pipes, all of which are not inactivated by the sterilization process.^{15,16)}

The goal of the research presented was to evaluate feasible disinfectants that could be used to effectively and practically control microorganisms in drinking water distribution. To achieve the goal of identifying suitable chemical disinfectants that could be used, it was determined and compared the inactivation of total coliform as the indicator organism when chlorine, chlorine dioxide are used.

II. Material and Methods

1. The Simulated Distribution System

To test the application of disinfectants under chemical and hydraulic conditions similar to those of a portable water distribution network, the simulated distribution system was constructed in

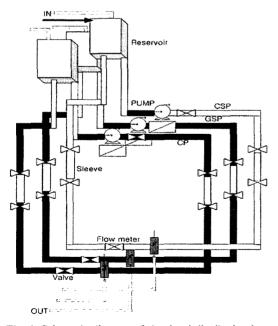


Fig. 1. Schematic diagram of simulated distribution loop system.

the laboratory. The schematic diagram of a simulated distribution loop system is presented in Fig. 1.

The model system was 7.4 m long and composed of 10 cm sections of carbon steel, galvanized, and copper pipe joined together with quick-fit PVC coupling to provide easy access to the interior of the pipe system for analysis of biofilm populations. Sampling sites were located at the inlet and outlet of the pipe network throughout the system.

Tap water at the University of *Konkuk*, which entered the system in the simulated clear well, served as the source water for the pipe system. To maintain a constant water volume, a float valve was utilized. A temperature circulator was used to control the water temperature. This device ensured that the source water was properly mixed with disinfectants. The pH of the water was maintained using a pH controller.

For this study, a stock solution of free chlorine and chlorine dioxide was prepared using 2 percent sodium hypochlorite and 0.8 percent chlorine dioxide. The water was pumped from the clear well through the pipe network at a flow velocity of 1.5 m/sec using a second digital peristaltic pump.

2. Analysis of Pipe System

At the initial stage of each experiment, the pipe used in the system was inoculated with an indigenous coliform population obtained from the sewer manhole adjacent to *Konkuk* University, as previously mentioned. Microorganisms were allowed to develop in the pipe interior for four weeks prior to application of the disinfectant.

Samples for suspended bacteria enumeration were collected at 1, 5, 15, 30, 60 and 120 min in the pipe system after applying the disinfectant. The aliquots were neutralized with sodium thiosulfate at various time intervals.

Samples indicating fixed bacteria on pipes were extracted with a phosphate solution using ultra sonification. After being incubated at 35°C for 24 hr, total coliform bacteria were enumerated using the membrane filter technique. A turbidimeter was used to determine turbidity. Total organic carbon (TOC) was analyzed with an organic carbon analyzer. Three times a week, flow rates of the peristaltic pumps were calibrated and subsequently adjusted.

3. Microscopical Examinations

A Leitz phase microscope capable of magnification up to 1000× was used for qualitative characterization of biofilms. Pieces of coupons were cut out for scanning electron microscopy of 5 × 5 mm under water. The specimens were washed in distilled water, and then dehydrated in a critical point dryer. Finally, the samples were mounted on brass stubs using carbon paint and sputter coated with 10-12 nm of gold. These specimens were observed using a Leika stereo scan 440 scanning electron microscope.

4. Culture Conditions and Microorganisms Analysis

The samples used to prepare stock inoculums were obtained from a sewer manhole adjacent to Konkuk University, which is located in the eastern area of Seoul. They were filtered through 11 µm membrane filters. A 10 ml filtrate was placed on a M-endo medium in a petridish and incubated for 24 h at 35°C. The colonies formed were introduced on the solution of nutrient broth and incubated at 25°C for 24 hr. The coliform bacteria were isolated using centrifugation at $5 \times 10^3 G$ for 5 min, then washed twice and resuspended in the buffer solution free from chlorine demand at a pH level of 7. The coliform bacteria isolated from the media were mixed using a vortex mixer. All experiments used the fresh cell suspensions in chlorine-demand-fee solution. The density of cells used varied between 2×10^6 and 8×10^8 cfu/100 ml in total coliform. Total coliform was enumerated in accordance with a membrane filtration procedure outlined by the US standard method 9222B.¹⁷⁾ The samples containing the coliform bacteria were filtered through the Millipore membrane sterilized to 0.45 µm in porosity. Every station in the filtration system was sterilized. A membrane was placed on the apparatus under stable conditions. Then, the aliquots of 1, 10 and 100 ml were filtered. Next, the membrane was placed on an agar M- endo medium in a petridish and incubated for 24 hr at 35 ± 1 °C. Results are expressed as coliform colony per 100 ml.

III. Result and Discussion

1. Bactericidal Effects of Disinfectants on Suspended Coliforms

Fig. 2 presents the results obtained from the

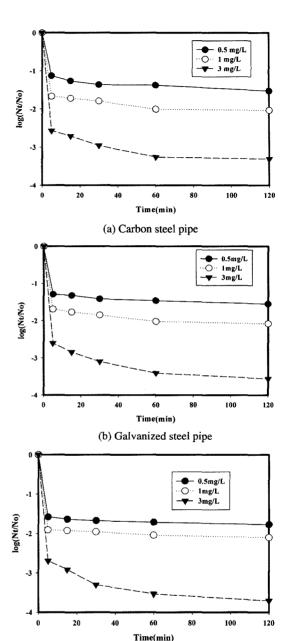


Fig. 2. Effect of chlorine concentration dosed to disinfect of suspended coliforms in simulated distribution system.

(c) Copper pipe

suspended coliforms inactivation experiments in simulated distribution systems, which depended on a free chlorine concentration at pH 7, 20°C. The rate of inactivation for suspended coliforms

using three different types of pipe (i.e. carbon steel, galvanized steel and copper pipes) increased as free chlorine increased after post-chlorination. Thus, it is clear that a large proportion of coliforms inactivated within 5 min.

Pipe materials can also affect the efficiency of disinfectant. When using free chlorine for carbon steel pipe, suspended coliforms are gradually reduced after a chlorine injection. The log survival ratio at carbon steel pipe was -1.13, -1.67 and -2.57 for 0.5, 1 and 3 mg/l after 5 min, respectively. The log survival ratio at carbon steel pipe was -1.36, -1.79, and -2.96 for 0.5, 1 and 3 mg/l after 30 min, respectively. When the contact time was extended up to 120 min., the inactivation rate of suspended coliform was 97.04, 99.08 and 99.95%, respectively.

In the galvanized pipe, the log survival ratio for coliforms reflects an increase as the injection concentration of free chlorine increases. Under 1 mg/l of free chlorine, inactivation reaction was almost completed in galvanized pipe during the first 5 min. At a level of 3 mg/l of free chlorine, the total coliform reduction achieved in galvanized pipe is achieved after exposing it continuously for 120 min. The inactivation efficiencies were 97.20, 99.17 and 99.97% for 0.5, 1, 3 mg/l after 120 min.

The rate of coliforms inactivation with chlorine in a copper pipe was higher than the values obtained using the other pipes. The log survival ratio for copper pipe was -1.58, -1.91 and -2.69 for 0.5, 1 and 3 mg/l after 5 min, respectively. Consequently, the inactivation rate was 97.36, 98.77 and 99.80%. The log survival ratio for copper pipe was -1.67, -1.96 and -3.30 for 0.5, 1 and 3 mg/l after 30 min. When the contact time was extended to 120 min, the inactivation rate of suspended coliform was 98.30, 99.21 and 99.98%, respectively. Results for free chlorine indicated that the type of pipe material slightly influences the inactivation of coliform. The inactivation rate of coliforms appeared in the lowest order of carbon steel, galvanized and copper pipe.

Fig. 3 presents the chlorine dioxide inactivation data obtained with coliforms at 20°C and pH 7 in simulated distribution system. The shapes of the inactivation curves were similar to those previously presented for the disinfection of chlorine. Using a

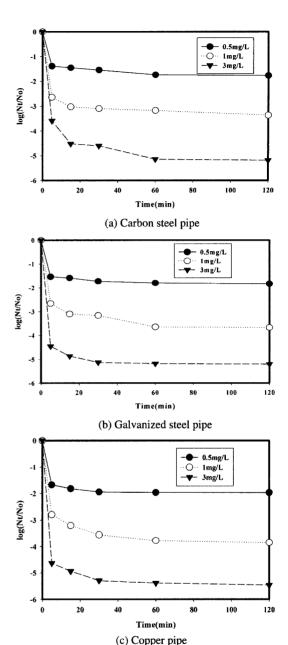


Fig. 3. Effect of chlorine dioxide concentration dosed to disinfect of suspended coliforms in simulated distribution system.

carbon steel pipe, the log survival ratio was -1.39, -2.64 and -3.59 for 0.5, 1 and 3 mg/l after 5 min, respectively. The log survival ratio at 30 min was -1.54, -3.02 and -4.54 for 0.5, 1 and 3 mg/l, respectively. When the contact time was extended

to 120 min, the log survival ratio was -1.77, -3.37 and -5.19 for 0.5, 1 and 3 mg/l, respectively. It followed that the inactivation efficiencies of suspended coliforms were 99.29, 99.96 and 99.99%. The log survival ratio at carbon steel pipe for 120 min of chlorine dioxide was 1.2, 1.7 and 1.6 times higher than that of free chlorine.

Using galvanized pipe, the log survival ratio was -1.53, -2.65 and -4.45 for 0.5, 1 and 3 mg/l after 5 min, respectively. Thus, inactivation efficiencies of suspended coliforms were 99.29, 99.96 and 99.99% for 0.5, 1 and 3 mg/l. The log survival ratio was -1.84, -3.67 and -5.20 for 0.5, 1 and 3 mg/l after 120 min, respectively. The log survival ratio for the galvanized pipe subjected to 120 min of chlorine dioxide was 1.2, 1.8 and 1.5 times higher than that of free chlorine.

Chlorine dioxide reacted quickly with microorganism in the first 5 min, using copper, and the inactivation efficiencies of suspended coliforms for 5 min were 99.88, 99.83 and 99.99% for 0.5, 1 and 3 mg/l. The log survival ratio was -1.94, -3.55 and -5.29 for 0.5, 1 and 3 mg/l at 30 min. The log survival ratio for 120 min was -1.97, -3.85 and -5.47 for 0.5, 1 and 3 mg/l, The log survival ratio for a copper pipe subjected to 120 min of chlorine dioxide was 1.1, 1.8 and 1.5 times higher than that of free chlorine.

In Fig. 4, coliforms inactivation is shown expressed as a function of dissolution of organic carbon for free chlorine. As DOC decreased, the bactericidal effects of the disinfectants were significantly decreased. It is well known that organic materials consume chlorine. As DOC is reduced by 3 mg/l with chlorine 1 mg/l, the coliform inactivation rate after 5 min can be increased by 1.8 times in the carbon steel pipe. For the majority of contact time, a significant coliform inactivation response to DOC is evident. The log survival ratio for 60 min was determined to be -2.01 and -1.31 for DOC 1.2 and 4.2 mg/l.

The inactivation of tap water is influenced by the addition of humic acids, which affect the rate of chlorine decay in galvanized steel pipe. A low DOC concentration results in a high rate of coliform inactivation. The log survival ratio for 5 min was determined to be -1.69 and -0.93 for DOC 1.2 and 4.2 mg/l. As DOC is reduced by 3 mg/l, the

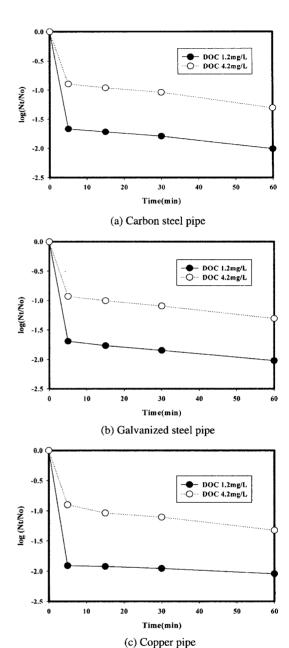


Fig. 4. Effect of DOC to disinfect of suspended coliforms with free chlorine in simulated distribution system.

coliform inactivation for 5 min can be increased by 1.8 times in galvanized steel pipe. The log survival ratio for 60 min was determined to be -2.02 and -1.31 for DOC 1.2 and 4.2 mg/l. The log survival ratio for 60 min of chlorine exposure at DOC 1.2 mg/l was 1.55 times higher than that

of DOC 4.2 mg/l. After 60 min of contact time, the killing effect percentage was 99.05 and 95.06% for DOC 1.2 and 4.2 mg/l.

Coliforms were also determined to be higher during chlorination with DOC 4.2 mg/l in the copper

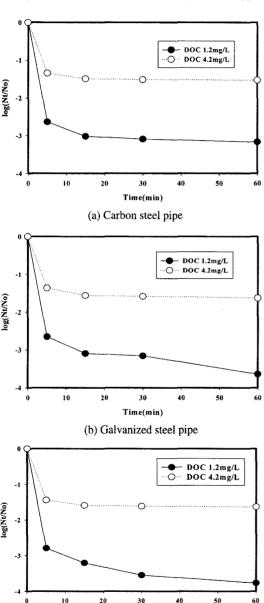


Fig. 5. Effect of DOC to disinfect of suspended coliforms with chlorine dioxide in simulated distribution system.

Time(min)

(c) Copper pipe

pipe. The log survival ratio for 5 min was determined to be -1.91 and -0.90 for DOC 1.2 and 4.2 mg/l. As DOC is reduced by 3 mg/l, the coliform inactivation for 5 min increases by approximately 2.12 times. The log survival ratio for 60 min was determined to be -2.64 and -1.33 for DOC 1.2 and 4.2 mg/l. The log survival ratio for 60 min of chlorine exposure at DOC 1.2 mg/l was 1.54 times higher than that of DOC 4.2 mg/l. After a contact time of 60 min, the killing effect percentage was 99.10 and 95.27% for DOC 1.2 and 4.2 mg/l.

Fig. 5 depicts the inactivation rate of coliforms after chlorine dioxide treatment. Significant decreases in bacterial counts were observed after the addition of humic acids. The simulated distribution samples presented -2.65 and -1.34 log inactivation of total coliforms for DOC 1.2 and 4.2 mg/l at 5 min. As DOC is descended by 3 mg/l, the coliform inactivation for 5 min increases by approximately 1.96 times in the galvanized steel pipe. The log survival ratio for 60 min was determined to be -3.17 and -1.53for DOC 1.2 and 4.2 mg/l. The log survival ratio for 60 min of exposure to chlorine at DOC 1.2 mg/l was 2.07 times higher than that of DOC 4.2 mg/l. After a contact time of 60 min, the killing effect percentage was 99.93 and 97.06% for DOC 1.2 and 4.2 mg/l.

For the galvanized pipe, a decrease in bactericidal effectiveness, which was attributed to the presence of DOC, was observed in all of the test solutions at DOC 1.2 and 4.2 mg/l. The 2.65 log₁₀ inactivation of coliform achieved by using a 1 mg ClO₂/L at DOC 1.2 mg/l exceeded, even at only 5 min of contact time, the amount of coliforms inactivation achieved after 60 min of contact time at DOC 4.2 mg/l. The percentage of killing effects after a contact time of 5 min was 99.78 and 95.67% for DOC 1.2, and 4.2 mg/l. The log survival ratio for 60 min was determined to be -3.64 and -1.63 for DOC 1.2, 4.2 mg/l. The log survival ratio for 60 min of chlorine at DOC 1.2 mg/l was 2.24 times higher than that of DOC 4.2 mg/l. The percentage of killing effects after a contact time of 60 min was 99.98 and 97.64% for DOC 1.2 and 4.2 mg/l.

There was a significant effect on the DOC in the copper pipe. The log survival ratio for 5 min was determined to be -2.79 and -1.44 for DOC 1.2 and 4.2 mg/l. The log survival ratio for 5 min of

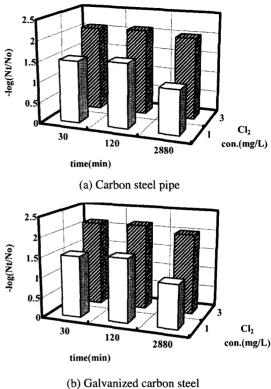
exposure to chlorine dioxide at DOC 1.2 mg/l was 1.94 times higher than that of DOC 4.2 mg/l. The percentage of killing effects after a contact time of 5 min was 99.84 and 97.66% for DOC 1.2 and 4.2 mg/l. The percentage of killing effects after a contact time of 30 min was 99.97 and 97.55% for DOC 1.2 and 4.2 mg/l. The log survival ratio for 60 min was determined to be -3.77 and -1.63 for DOC 1.2 and 4.2 mg/l. The log survival ratio for 60 min of chlorine at DOC 1.2 mg/l was 2.31 times higher than that of DOC 4.2 mg/l. The percentage of killing effects after a contact time of 60 min was 99.98 and 96.38% for DOC 1.2 and 4.2 mg/l. Thus, inactivation effects were presented in the lowest order of carbon steel, galvanized steel and copper pipe when injecting chlorine dioxide.

2. Bactericidal Effects of Disinfectants on Fixed Coliforms

To evaluate the inactivation effects of biofilm on various pipe surfaces, chlorination of fixed bacteria was performed at different initial free chlorine concentrations. Fig. 6 presents the results of these experiments. For carbon steel, galvanized steel, and copper, application of free chlorine residual facilitated greater biofilm inactivation than 1 log at a dosage of 1 mg/l. Reduced fixed coliforms exceeded 2 log unit survival ratios at a free chlorine dose of 3 mg/l. After a period of 24 hr dosage of 3 mg/l, there was no increase in the fixed coliforms. A dosage of 3 mg/l free chlorine, was effective for fixed bacteria. After 24 hours, a decrease in free chlorine residuals in distribution systems resulted in a subsequent decrease in coliforms inactivation.

The log survival ratio for 120 min was determined to be -2.10, -2.15 and -2.22 for carbon steel, galvanized steel, and copper pipe when a dose of free chlorine 3 mg/l was injected. The inactivation effects were observed in the lowest order of carbon steel, galvanized steel, and copper pipe when free chlorine was injected. Free chlorine effectively controlled biofilm grown on pipe surfaces at a dose of 3 mg/l.

Fig. 7 depicts the densities of biomass fixed in materials in the presence of water containing chlorine dioxide. The measured fixed biomass was practically equal in all three types of pipes. Application of a chlorine dioxide resulted in little higher



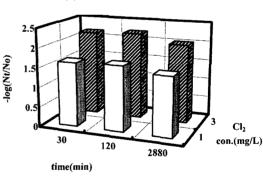


Fig. 6. Effect of initial chlorine to disinfect of fixed coliforms in simulated distribution system.

(c) Copper pipe

inactivation than did free chlorine. The log survival ratio for 120 min at chlorine dioxide 1 and 3 mg/l was 1.6 and 1.2 times higher than that of free chlorine. Chorine dioxide dosed at 3 mg/l reduced fixed coliforms by more than a 2.5 log unit survival ratio after 120 min.

The log survival ratio for 24 hr was determined to be -0.7, -0.73 and -0.74 for carbon steel,

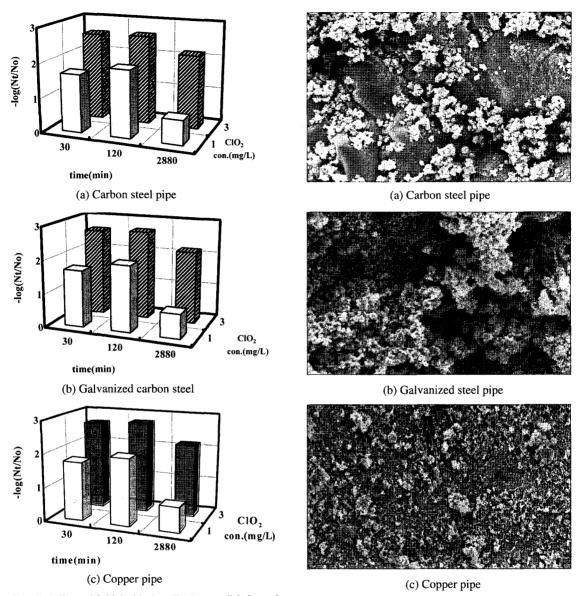


Fig. 7. Effect of initial chlorine dioxide to disinfect of fixed coliforms in simulated distribution system.

galvanized steel, and copper pipe when a dose of chlorine dioxide 1 mg/l was injected. These results suggest that chlorine dioxide failed to maintain residual disinfectant after 24 hr, and consequently failed to control the regrowth of microorganisms.

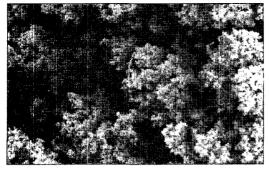
3. Pipe Wall Observation by SEM

To observe fixed bacterial growth in a simulated system, coupon was extracted from different disin-

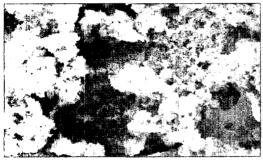
Fig. 8. The SEM photos of pipe at chlorinated system.

fectant treated pipelines after 20 days of operation. Before the variously treated pipes were allowed to attach bacterium through the test pipes, tap water containing a high density of coliforms was circulated to create biofilm through all the systems. Disinfectant residual ranged from 0.05 to 0.20 mg/l. Tubercles and crevices were observed in pipe walls, as shown in Fig. 8.

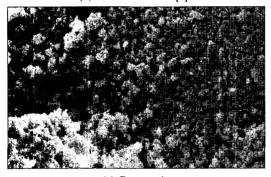
As presented in Fig. 8, the layer of patchy



(a) Carbon steel pipe



(b) Galvanized steel pipe



(c) Copper pipe

Fig. 9. The SEM photos of pipe at chlorine dioxide treatment system.

biofilm was decreased in the chlorinated system. However, the chlorine disinfected copper pipe system did not demonstrate any potential for biofilm formation. This fact does not correlate with a study by Characklis (1981). In this study an experimental Roto Torque System with a free-chlorine residual between 0.05 and 0.2 mg/l, concluded that the number of planktonic cells and biofilm bacteria was about equal to the non-disinfected control water. However, Lund and Ormerod(1984) determined

that the very low residual free-chlorine concentration sufficiently prevented the attachment of bacteria, and biofilm formation.¹⁹⁾

Water treated with chlorine dioxide disinfectant revealed also tubercles production. The results indicate that the use of chlorine dioxide may retard the formation of bacterial activity to some extent but it is still observed biofilm and tubercles formation in pipe coupons after chlorine dioxide dosing.

IV. Summary

This research provides developed discussion, which can be used to assess the applicability of chlorine and chlorine dioxide and determines an appropriate disinfectant for drinking water systems. The following conclusions were drawn from this study:

- 1. Pipe materials affected the efficiency of disinfectant. The inactivation rate of coliforms was observed at the lowest rate for carbon steel, followed by galvanized and copper pipe in the simulated distribution system.
- 2. The log survival ratio for three sorts of pipes of chlorine dioxide was higher than that of free chlorine for suspended coliform. Application of a chlorine dioxide for controlling fixed microorganism in distribution system resulted in little higher inactivation than did free chlorine.
- 3. There was a significant effect for DOC condition on the total coliform disinfection in the pipe system. Especially, it showed the effect of DOC for the inactivation with chlorine dioxide was higher than that with chlorine.
- 4. In spite of the maintenance of chlorine residuals at approximately 0.2 mg/l in pipes, regrowth of bacteria was observed. A scanning electron microscope confirmed these results. The SEM results of the pipe sample revealed that the inner wall of the pipe was not smooth, and bacteria were grown although disinfectant was applied such as chlorine and chlorine dioxide in pipe systems. It showed disinfectant treatment should not be absolute method for controlling the microbial problem in distribution system.

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