INTRODUCTION:

Drinking Water Distribution System Managers face a myriad of challenges as they work to provide safe and aesthetically pleasing water to customers. These challenges include minimization of corrosion and tubercule formation within distribution lines, control of microbial regrowth, and execution of effective flushing programs. Interestingly, these three issues are interrelated. Tubercules and corrosion within pipes yield sheltering homes for bacteria to breed and form microbial colonies (biofilms).

Research has shown that biofilms have the potential of harboring Coliform bacteria. Of a more serious concern, laboratory investigations have revealed that biofilms can contain microbial organisms of a pathogenic nature. Although biofilm organisms have not been linked to specific waterborne disease incidents, public health concerns lead one to conclude that distribution systems should be properly maintained in order to minimize the risk from microbial regrowth. To this end, guidance has been developed to assist public water systems in evaluating and controlling biological regrowth in distribution systems.

Although flushing and residual disinfection can be effective at controlling biofilms, there are many operational considerations that preclude their effectiveness. A major problem area for many municipal water systems is flushing small, water distribution lines. These lines are typically too small to flush with the high flow rates required to scour pipe walls. Therefore, corrosion and tubercule formation can occur at higher rates than are typically seen in larger capacity water mains. This circumstance also allows microbial regrowth to occur.

Sugden Development, LLC has developed an approach (“The Sugden Process”) to enhance the flushing process typically used on distribution system lines. This approach involves injecting CO$_2$ into the distribution system line to lower the pH while flushing takes place. The US Environmental Protection Agency (USEPA) Office of Research and Development’s Drinking Water Research Division has documented that this process is an effective way of removing biofilm organisms at the bench- and pilot-scale level. The effect of CO$_2$ on biofilm organisms has also been documented in a report written by Micor-Ambient Corporation. In order to demonstrate the effectiveness, practicality and viability of pH adjusted flushing under field conditions, a research project was conducted during December 2000 in Asheville, North Carolina. This study was conducted with the assistance of the City of Asheville’s Water Department on a segment of active distribution line that had recently been disconnected.
from individual customers. The pH was adjusted to lower levels through the addition of carbon dioxide to the distribution water used in the flushing process.

The objectives of this study were to provide information on the removal of biofilm organisms in distribution lines and to document the collateral effects while flushing at lower than normal pH levels. The specific aims of the study were to measure the reduction and/or removal of naturally occurring biofilm organisms in a drinking water distribution line while flushing at low pH levels and to evaluate the effect of this practice on calcium, iron, lead, copper, suspended solids, heterotrophic plate count organisms, and free chlorine residual concentrations in solution.

EXPERIMENTAL SYSTEM:

The City of Asheville and their Consulting Engineers (McGill & Associates) assisted the investigators in identifying a promising research site. The City of Asheville’s Water Resources Department was in the process of replacing many older residential distribution mains when this research project was proposed. City Water Officials were interested in the prospect of evaluating some of their older distribution pipes as they were taken out of service, and were interested in evaluating an innovative approach to flushing. Drinking water and wastewater regulators from the North Carolina Department of Environment and Natural Resources were appraised of the research proposal, and all necessary approvals were obtained. Resultantly, an approximately 400 foot long section of 40+ year old, two-inch distribution line located on Madison Avenue was selected as the primary research site.

The City’s distribution crew and pipe replacement contractor worked with the research team to isolate the distribution line evaluated in the study, collected an initial pipe coupon for biofilm scrape sample collection, and installed the required pipe fittings in order to allow for injection of the flushing water (mixed with carbon dioxide).

Gaseous carbon dioxide was injected from a storage cylinder into the distribution water used for flushing by means of an injection panel (Illustration 1). This apparatus was installed on the back of a truck and was piped to obtain City of Asheville distribution water from a nearby hydrant (connection made with the use of a backflow prevention device). The water was mixed with carbon dioxide in the truck-mounted injection panel, and introduced into the distribution line for the flushing experiment. The carbon dioxide feed rate was adjusted to control the pH of the water used for flushing in this experiment. The flushing feed rate was set at 10 gpm as a scale-up based on the experimental flushing conditions of 1 ft./sec velocity in the EPA laboratory study. Illustration 2 shows the experimental layout of the research project.
ILLUSTRATION 1 – Carbon Dioxide Storage Tank and Injection Panel
(courtesy Airgas Carbonic)

ILLUSTRATION 2
EXPERIMENTAL SYSTEM LAYOUT

Flushing Effluent
Discharge Point

pH Adjusted
Flushing Injection Point

CO₂ Injection Panel

Hydrant w/ backflow preventer
EXPERIMENTAL CONDITIONS:

This research project consisted of two phases. The first phase of the study (Initial Flushing Study) involved injection of carbon dioxide at a rate resulting in a pH range of 4.9 to 5.7 in the flushing water. During this first phase of the research, flow rates were adjusted to approximately 10 gallons per minute. The experiment was conducted for 39 consecutive hours under these conditions. Pipe coupons were removed both before and after this phase of the experiment in order to collect scrape samples from the inner walls of the pipe for determination of biofilm organism concentration in the laboratory. Influent and effluent samples were collected at the following time intervals: baseline, 0.5 hr., 1 hr., 3 hr., 5 hr., 9 hr., 14 hr., 19 hr., 24 hr., 29 hr., 34 hr., and 39 hr. Temperature, pH, Heterotrophic Plate Count (HPC) bacteria, calcium, iron, and suspended solids analyses were conducted on each influent and effluent sample taken during the first phase of the research. Lead and copper analyses were conducted on the influent and effluent of the first and last set of samples taken during the first phase of this research.

The second phase of the research project was conducted immediately following the first phase at the same site (Madison Avenue), on the same distribution line, and involved injecting a higher rate of carbon dioxide into the flushing water (later referred to as the ‘high dose study’). As a result, the pH of the flushing water dropped to a range of 4.7 to 4.9. Flow rates for this second phase of the study remained at 10 gallons per minute. This phase of the experiment was conducted for 2.75 hours. A pipe coupon was removed after this phase of the experiment in order to collect a scrape sample from the inner wall of the pipe for determination of biofilm organism concentration. Influent and effluent samples were collected at the following time intervals: baseline, 0.25 hr., 0.5 hr., 1 hr., 1.75 hr., and 2.75 hr. Temperature, pH, Heterotrophic Plate Count (HPC) bacteria, calcium, iron, and suspended solids analyses were conducted on each influent and effluent sample taken during the second phase of the research. Lead and copper analyses were conducted on the influent and effluent of the last set of samples taken during the high dose study. Free chlorine residual concentration was measured in the last set of samples taken during the high dose study.

With the exception of the biofilm scrape samples, all sampling was conducted and analyzed in accordance with Standard Methods for the Examination of Water and Wastewater \(^\text{10}\) and/or approved United States Environmental Protection Agency (USEPA) methods. The biofilm scrape samples were analyzed using a methodology developed by the USEPA’s National Risk Management Research Laboratory’s Water Supply & Water Resources Division \(^8\).

RESULTS:

1. Initial Flushing Study (Phase One of the Research)

1.1 Temperature
Raw water (influent) and flushing effluent temperatures were recorded at each sampling interval and are illustrated in Figure 1.1. Although slight differences were found in the influent and effluent temperatures, they were within sampling and thermometer range of error. Temperatures ranged between 6.6 and 9 degrees Celsius. Note that this temperature range is not normally considered conducive to biological regrowth in distribution systems.

**FIGURE 1.1**
Temperature Change
Madison Avenue Initial Flushing Study

1.2 pH

Raw water (influent) and flushing effluent pH were monitored and recorded at each sampling interval and are shown in Figure 1.2. As expected, flushing effluent pH dropped from 7.8 pH units down to approximately 5 pH units as the carbon dioxide gas was injected into the water used for flushing. Effluent pH rose from 5.1 to 5.7 pH units between hours 9 and 19 of operation as the carbon dioxide tank pressure decreased due to usage. This condition was corrected by hour 24 of operation. However, the effluent pH rose slightly from hour 24 to hour 34 as carbon dioxide tank pressure decreased again.
1.3 Heterotrophic Plate Count Organisms

HPC samples were taken from the raw water and flushing effluent water during every scheduled sampling event. The results are shown in Figure 1.3. Although raw water HPC concentrations were initially found to be 60 colony-forming units per milliliter (CFU/mL), they dropped to a range between 1 and 10 CFU/mL during the duration of the initial flushing study. It should be noted that the laboratory analyst questioned the validity of the initial raw water sample in light of its high level.

Flushing effluent HPC concentrations ranged between 1 and 13 CFU/mL. It is interesting to note that the flushing effluent HPC concentrations were slightly higher than the raw water HPC concentrations from study hours 1 to 9 and again at hour 34. Although these elevated effluent HPC concentrations are obviously related to the raw water HPC spikes that occurred concurrently, it is theorized that the elevated effluent HPC concentrations are also the result of the slow breakup of the biofilm on the pipe walls that could have allowed the release of HPC organisms into the bulk fluid. Since this experiment was conducted in the presence of a free chlorine residual (1.4 mg/L in the influent and 1.3 mg/L in the effluent), many of the HPC organisms broken loose or exposed within the biofilm would have probably been quickly inactivated and would not have been seen in these sampling results.

The lack of HPC organisms in the flushing effluent during hours 14 – 24 of the study may be related to the decrease in CO₂ dosing that occurred during that time period as the CO₂ pressure decreased (indicated by the rise in flushing effluent pH in Figure 1.2). On a related note, the reappearance of HPC organisms in the flushing effluent during hour 34 of the study may be related to the adjustment of CO₂ dosing which occurred at hour 24 of the study and lowered the pH (Figure 1.2).
1.4 Calcium

Raw water and flushing effluent samples from each sampling event were analyzed for calcium. The results are shown in Figure 1.4. Raw water calcium concentration remained at 1.1 mg/L for the first 14 hours of the study, dropped to 1.0 mg/L from hours 19 to 34, and dropped to 0.97 mg/L at hour 39. Effluent calcium concentration spiked to 1.8 mg/L immediately following the onset of flushing enhanced with carbon dioxide. Effluent calcium concentration then slowly tailed off to baseline levels by hour 9 of the study, and rose again to 1.1 mg/L at hour 29 of the study.

The initial spike in effluent calcium concentration was to be expected as the carbon dioxide in the flushing water formed carbonic acid and reacted with the lining of the distribution pipe wall, thus placing calcium back into solution. This data indicates that this reaction was fairly rapid, and that conditions had stabilized back to baseline calcium levels by hour 9 of the study. The slight rise in concentration of calcium in the flushing effluent that is seen at hour 29 may have been related to the adjustment of CO₂ dosing that occurred at hour 24 of the study and lowered the pH (Figure 1.2).
1.5 Iron

Raw water and flushing effluent samples from each sampling event were analyzed for iron. The results are shown in Figure 1.5. Raw water iron concentration ranged between 0.067 and 0.16 mg/L during the study period. With the exception of one spike (0.16 mg/L at hour 1 of the study), the raw water iron concentration remained below 0.1 mg/L. Effluent iron concentration ranged between 0.069 and 0.11 mg/L in the samples.

The observed trend in effluent iron samples involved a slight increase in concentration from 0.081 mg/L in the baseline sample to 0.11 mg/L in the sample taken 0.5 hours. This is to be expected, as the carbonic acid reacted with the tubercules inside the pipe and allowed the iron to reenter the solution. Iron concentration in the flushing effluent decreased to its minimum value by hour 19. Both effluent and influent iron concentrations slowly rose from hour 24 until the end of the study, with effluent concentrations at hours 24 and 29 rising slightly above raw water levels. This may have resulted from the adjustment of CO\(_2\) dosing that occurred at hour 24 of the study and lowered the pH (Figure 1.2).
1.6 Suspended Solids

Suspended solids analyses were conducted on every raw water and flushing effluent sample collected during the study. The results are shown in Figure 1.6. Although the data is difficult to interpret, general trends can be observed.

Raw water suspended solids concentration spiked at 0.8 mg/L during hour 1 of the study, and decreased steadily to hour 9. From hour 9 to hour 39 of the study, the raw water suspended solids concentration bounced between 0 and 0.2 mg/L. Effluent concentration of suspended solids trended higher (up to 0.6 mg/L) to hour 9 of the study and fell to 0 mg/L by hour 14. Flushing effluent suspended solids concentration then rose sharply at hour 24 (up to 0.8 mg/L) and then slowly dissipated.

Effluent suspended solids trending offers more evidence to support the theory that the carbon dioxide in the flushing water formed carbonic acid and reacted with the lining of the distribution pipe wall, thus placing particulate matter into solution. This data indicates that this effect occurred during the first 9 hours of the study, and that conditions stabilized by hour 14 of the study. The prominent increase of suspended solids in the flushing effluent that is seen at hour 24 seems to correspond to the adjustment of CO₂ dosing that occurred simultaneously and lowered the pH (Figure 1.2).
2. **High CO₂ Dose Flushing Study (Phase Two of the Research)**

2.1 Temperature

Raw water (influent) and flushing effluent temperatures were recorded at each sampling interval during the high dose study and are illustrated in Figure 2.1. Although slight differences were found in the influent and effluent temperatures, the data was within sampling and thermometer range of error. Temperatures ranged between 7.8 and 8.3 degrees Celsius.
2.2 pH

Raw water (influent) and flushing effluent pH were monitored and recorded at each sampling interval during the high dose study and are shown in Figure 2.2. High dose CO$_2$ flushing effluent pH dropped from 7.6 pH units down to 4.7 pH units as the carbon dioxide gas was injected into the water used for flushing. Effluent pH fluctuated between 4.7 and 4.9 pH units during the 2.75 hour high dose study.

![Flushing Effect on pH](image)

2.3 Heterotrophic Plate Count Organisms

HPC samples were taken during the high dose study from the raw water and flushing effluent water during every scheduled sampling event. The results are shown in Figure 2.3. All raw water and flushing effluent samples were found to contain <1 CFU/mL. This data seems to indicate that any HPC organisms that were exposed or released from the biofilm during the high dose study were inactivated.
2.4 Calcium

Raw water and flushing effluent samples from each sampling event during the high dose study were analyzed for calcium. The results are shown in Figure 2.4. Raw water calcium concentrations fluctuated between 1.0 and 1.1 mg/L during this study. High dose flushing effluent calcium concentrations immediately rose to 1.4 mg/L and then steadily decreased to raw water levels.

It is interesting to note the magnitude of the effluent calcium spike, especially when one considers that the distribution line utilized for the high dose study is the same one that was previously utilized for the initial study. The higher dose of carbon dioxide added to the flushing water in this study allowed for the formation of a stronger solution of carbonic acid than in the initial study. The carbonic acid then reacted with calcium that remained in the lining of the distribution pipe wall and enabled it to enter the bulk phase.
2.5 Iron

Raw water and flushing effluent samples from each sampling event during the high dose study were analyzed for iron. The results are shown in Figure 2.5. Raw water iron concentration ranged between 0.064 and 0.077 mg/L during the study period. Effluent iron concentration ranged between 0.08 and 0.098 mg/L in the samples.

Interestingly, the baseline effluent iron concentration slightly exceeded that of the baseline raw water iron concentration. This may be due to a residual leaching process from the initial flushing study (which had been completed at this site immediately prior to the start of the high dose study). Iron concentration in the flushing effluent dropped slightly as the high dose study began and then increased through the duration of the study as the carbonic acid reacted with the tubercules inside the pipe and allowed the iron to reenter the solution.
2.6  Suspended Solids

Suspended solids analyses were conducted on every raw water and flushing effluent sample collected during the high dose study. The results are shown in Figure 2.6. As with the initial study, the high dose suspended solids data is difficult to interpret, allowing only general trends to be observed.

Suspended solids concentrations in the raw water and flushing effluent trended downward during the high dose study. However, the flushing effluent yielded a slight increase in suspended solids at hour 1.75 of the study. This slight increase in flushing effluent suspended solids concentration may have been the result of particulate matter dissolving into the solution as the high dose study progressed.
3. **Biofilm Sampling**

3.1 **Biofilm Scrape Samples**

Pipe coupons were collected prior to the start of the initial flushing study, at the conclusion of the initial flushing study, and at the conclusion of the high dose flushing study. Scrape samples were obtained from these coupons and analyzed for microbiological organisms. The results of this sampling are found in Figure 3.1.

The baseline sampling resulted in a microorganism concentration of 1400000 CFU/cm². The scrape sample obtained at the conclusion of the initial flushing study yielded a microorganism concentration of 2232 CFU/cm². The scrape sample obtained at the conclusion of the high dose flushing study revealed a microorganism concentration of 248 CFU/cm². These results indicate that 99.84% (2.8 logs) of biofilm microorganisms were removed from the distribution line during the initial flushing experiment, and 99.98% (3.8 logs) of microorganisms were removed from the distribution line during the high dose experiment.
4. Other Sampling

4.1 Free Chlorine Residual

Free chlorine residual was determined on the last raw water and flushing effluent sample of the high dose study. The raw water sample was found to contain 1.4 mg/L of free chlorine, and the flushing effluent sample was found to contain 1.3 mg/L of free chlorine. This data indicates that carbon dioxide enhanced flushing has no significant impact on free chlorine residual in this experiment.

4.2 Lead Sampling

Lead analyses were conducted on the first and last raw water and flushing effluent samples taken during the initial study, along with the final raw water and flushing effluent sample of the high dose study. The results of all lead samples were non-detectable (detection limit of 0.005 mg/L). This data indicates that carbon dioxide enhanced flushing had no significant impact on lead concentration in this experiment. The effect of the carbon dioxide enhanced flushing process on lead leaching cannot be ascertained in this study.

4.3 Copper Sampling

Copper analyses were conducted on the first and last raw water and flushing effluent samples taken during the initial study, along with the final raw water and flushing effluent sample of the high dose study. The initial raw water copper sample was 0.0058 mg/L and the remaining two raw water samples were found to be non-detectable (0.005
Flushing effluent copper samples were found to range from 0.0055 mg/L in the baseline sample of the initial study, up to 0.043 mg/L in the final sample of the initial study and 0.038 mg/L in the final sample of the high dose study. These results are to be expected, as the carbonic acid in the flushing water reacted with the pipe material and allowed the copper to leach out and enter the bulk phase.

**FIGURE 4.3**
Flushing Effect on Copper Concentration

<table>
<thead>
<tr>
<th>Copper Concentration (mg/L)</th>
<th>Initial Study - Start (-0.5 hrs)</th>
<th>Initial Study - End (39 hrs)</th>
<th>High Dose Study - End (54.75 hrs)</th>
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</thead>
<tbody>
<tr>
<td>Raw</td>
<td>0.0055</td>
<td>0.043</td>
<td>0.038</td>
</tr>
<tr>
<td>Effluent</td>
<td>0.0055</td>
<td>&lt;=0.005</td>
<td>&lt;=0.005</td>
</tr>
</tbody>
</table>

**CONCLUSIONS**

This study shows that carbon dioxide enhanced flushing at lower pH levels can remove significant concentrations of biofilm organisms in a drinking water distribution system. Although leaching of calcium, iron, and copper was observed during this study, the leaching levels were relatively insignificant. The results of this study indicate that carbon dioxide enhanced flushing is a safe and effective means of treating drinking water distribution systems to control biofilm organisms.

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9. Report on Testing of Carbon Dioxide Treatment for Biofilm Control/Remediation in Drinking Water Systems; Project NO. 964047, Micor-Ambient Corporation, 4610 Central Avenue, St. Petersburg, Florida 33711, Telephone: (813) 328-0268.