

EVALUATION OF DRINKING WATER BIOSTABILITY USING BIOFILM METHODS

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ABSTRACT: A biofilm based, annular reactor method was developed and used to measure the biological regrowth potential of effluent water from various pilot treatment processes at the New York City Croton Lake Pilot Plant. A series of studies were carried out over the year-long study to collect bacterial growth and organic carbon biodegradation data for waters from six treatment options, including the raw source water. Quantitative and qualitative evaluations were made to determine the effects of filter media type, direct filtration, preozonation, and primary chlorination on the relative biostability of the produced waters compared to that of the original source water and water currently being distributed to consumers. In addition, results were compared to those obtained using traditional biodegradable organic material measuring methods such as assimilable organic carbon and biodegradable organic carbon. Quantitative biostability factors were developed that take into account both biological growth potential and biodegradability of the tested waters. Results from these studies were used to compare various piloted treatment processes and to assess pilot plant operation, design parameters, and seasonal source water quality.

INTRODUCTION

Microbiological growth in potable water distribution systems is a growing public health concern and quality of service issue. Regrowth can lead to numerous problems in a distribution system including microbial-induced corrosion, accumulation and proliferation of indicator or problem organisms, accumulation of nongrowing pathogens, and the general decline in water quality (i.e., color, odor, taste) (Geldreich et al. 1972; Ford 1999). These problems may be minimized/controlled by various treatment processes that are designed to remove biodegradable organic material from the water and optimize disinfectant stability. Processes that control bacteria-escaping treatment, as well as those present in the distribution system, are also important.

The biodegradability and regrowth potential of a finished water is a function of many parameters (Camper et al. 1996) including source water quality (Goel et al. 1995; Owen et al. 1995), temperature, type, and amount of biodegradable organic material (BOM) (Huck 1990), preozonation (Price et al. 1993; Goel et al. 1995; Joret et al. 1997), treatment (Huck et al. 1991; LeChevallier et al. 1996; Collins et al. 1996; Urfer et al. 1997; Carlson and Amy 1998), disinfection type and concentration (Koudjonov et al. 1997; LeChevallier et al. 1998; Haas 1999; LeChevallier 1999), and pipe material (Camper et al. 1996).

Methods for analyzing and monitoring the biostability or regrowth potential of water are useful in evaluating the effectiveness of different treatment processes at removing BOM from the water, gauging the suitability of treatment options during pilot study operations, and assessing water quality as it passes through a treatment or distribution system. The ma-

ajority of methods used today by the drinking water industry and researchers are either biomass-based methods or biodegradability methods (Huck 1990; Oxenford and Crozes 1998).

Development of a universal and reliable method for analyzing BOM has proven difficult because of the variety and variability of source waters. Surface waters are typically associated with having the potential to cause significant regrowth in distribution systems. Each surface water source has its own unique chemistry and bacterial ecology, with variable types and amounts of organic material (Goel et al. 1995; Owen et al. 1995). Established methods used to measure biodegradable organic carbon include numerous versions of the assimilable organic carbon (AOC) (van der Kooij 1992) and the biodegradable organic carbon (BDOC) (Servais et al. 1987) methods. However, these methods have significant limitations at measuring the regrowth potential of drinking water because they are batch methods that utilize either nonindigenous bacterial monocultures or highly acclimated bacteria, which do not necessarily represent the dynamic indigenous bacterial communities found in most waters and distribution systems. These bacterial communities can change with water quality, season, and treatment. Additionally, recent research has shown independent changes in AOC and BDOC concentrations as the water travels through a treatment train, restricting the reliability of these methods in evaluating treatment process performance (Huck 1990; Paode et al. 1997; Hu et al. 1999; LeChevallier 1999).

A relatively new method for measuring the regrowth potential or biostability of drinking water is the biofilm annular reactor (BAR) method. Because of the continuous flow and dynamic nature of the BAR system, it can simulate the environment of water flowing through a biofilm-laden pipe containing attached indigenous bacterial populations. The BAR method allows for the biostability assessment of a specific water by both biomass and organic carbon biodegradation-based methods. The biomass methods involve the measurement of the total amount of indigenous, multispecies biofilm growth supported by the water. The biodegradation methods involve the measurement of the amount and type of natural organic material (NOM) available to or utilized by the biofilm bacteria within the reactors.

The study presented here was modeled after those described by Camper (1997) and was developed to help evaluate the performance of piloted water treatment processes at New York City's Croton Reservoir. The findings of the first phase of this research are presented, revealing interesting relationships between various biomass and biodegradation results obtained using the BAR method. The results indicate that the BAR

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method can be a valuable tool for evaluating the biostability and biological regrowth potential of drinking water and may be used to provide insights into the design and operation of full-scale treatment processes.

EXPERIMENTAL METHODS

BAR

Biofilm annular reactors (BioSurfaces Technologies, Bozeman, Mont.) were used to monitor the regrowth potential of various treated waters. Fig. 1 shows a schematic of the BAR used in these studies. The BARs consist of a solid drum rotating on its vertical axis inside a stationary section of glass pipe (20 cm tall, 20 cm in diameter). The rotating drum is equipped with 20 removable polycarbonate slides from which biofilm samples can be taken. Polycarbonate slides were used as the biofilm support media to eliminate the confounding influence of reactor materials on biofilm growth. Using polycarbonate gives an indication of the growth potential of the water alone, avoiding the influence of corrosion and corrosion products. The completely mixed, continuous flow annular reactors were run at a rotating speed of 60 rpm (to simulate the hydraulic shear that is equivalent to a velocity of 0.61 m/s in a 30.5 cm drinking water main) and a hydraulic residence time of 2 h. The reactors have 1 L working volume and were equipped with influent and effluent sampling ports.

A diagram of the typical reactor system used in these studies, along with an example of the reactor locations in the direct filtration treatment train is shown in Fig. 2. The influent storage tanks used in the system had a 20-min residence time during pilot plant operation. The storage tanks were an important part of the system because they enabled continuous feed to the reactors during power outages and other pilot plant downtimes. Prior to the beginning of each run, the reactors were autoclave sterilized. The reactors were inoculated by the bacterial cells that entered each reactor, which included the cells that escaped previous treatment.

A total of six reactors were in operation during both the winter and summer runs. The nomenclature, location, and operating conditions of the treatment processes being evaluated by each reactor during the two runs are presented in Table 1. Each reactor run lasted 2 to 3 months with periodic influent, effluent, and biofilm sampling. The frequency of sampling increased toward the end of each run when it had been determined that a pseudo-steady state had been reached. Each system was considered to be at a pseudo steady state when changes in either dissolved organic carbon (DOC) concentrations or heterotrophic plate counts (HPCs) across the reactor

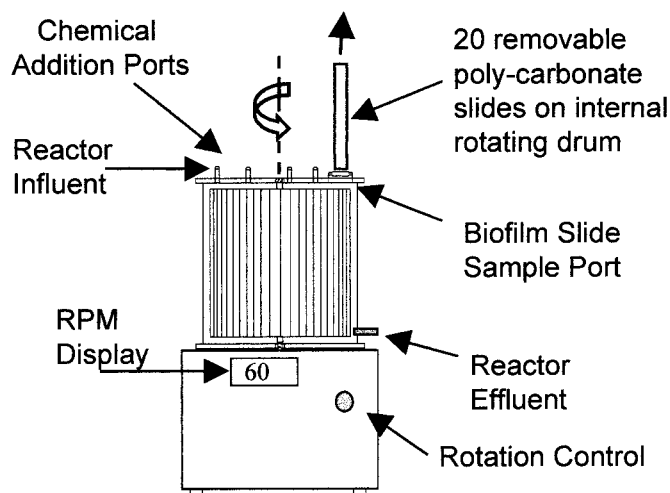


FIG. 1. Schematic of BAR

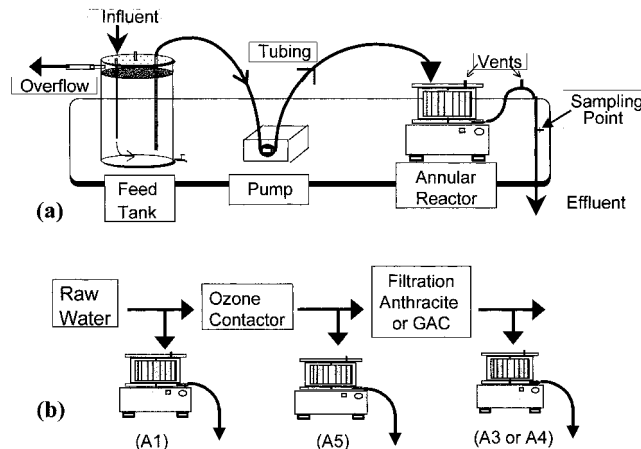


FIG. 2. (a) Schematic of BAR System; (b) Schematic Showing Locations of BAR Systems to Evaluate Individual Piloted Processes in Direct Filtration Treatment Train

TABLE 1. Nomenclature, Location, and Treatment Operation for Each BAR

Reactor	Treatment and operational parameters of each tested water
(a) Winter Runs (Run A) Reactor Numbers and Test Waters	
A1	Raw water from Croton Lake.
A2	Chlorinated raw water. Total chlorine residual = 0.35 ± 0.05 mg/L. 2-h contact time. Prior to distribution and booster chlorination.
A3	Direct anthracite filtered water. Ozone: HDT = 22 min at 1.4 mg/L. Alum coagulant. Filtration: 2.25 m of 1.4 mm anthracite, EBCT = 5 min, 366 L/min/m ² .
A4	Direct GAC filtered water. Ozone: HDT = 22 min at 1.4 mg/L. Alum coagulant. Filtration: 2.25 m of 1.4 mm GAC, EBCT = 5 min, 366 L/min/m ² .
A5	Preozonated raw water. Ozone: HDT = 22 min at 1.4 mg/L.
A6	Ozonated-GAC filtered water. Ozone: HDT = 22 min at 1.4 mg/L. No coagulation. Filtration: 2.25 m of 1.4 mm GAC, EBCT = 10 min, 183 L/min/m ² .
(b) Summer Run (Run B) Reactor Numbers and Source Waters	
B1	Raw water from Croton Lake.
B2	Chlorinated raw water. Total chlorine residual = 0.31 ± 0.05 mg/L. 2-h contact time. Prior to distribution and booster chlorination.
B3	Direct anthracite filtered water. Ozone: HDT = 22 min at 1.4 mg/L. Alum coagulant. Filtration: 2.44 m of 1.4 mm anthracite, EBCT = 5 min, 366 L/min/m ² .
B4	Direct GAC filtered water. Ozone: HDT = 22 min at 1.4 mg/L. Alum coagulant. Filtration: 2.44 m of 1.1 mm GAC, EBCT = 5 min, 366 L/min/m ² .
B5	Preozonated raw water. Ozone: HDT = 21 min at 1.4–2.8 mg/L.
B6	Ozonated-GAC filtered water. Ozone: HDT = 22 min at 1.4 mg/L. No coagulation. Filtration: 2.25 m of 1.4 mm GAC, EBCT = 10 min, 183 L/min/m ² .

Note: EBCT = empty-bed contact time; HDT = hydraulic detention time.

became relatively constant. An average of 15 influent, effluent, and biofilm samples were taken during each run, and all average data values consist of at least five samples taken toward the end of each run, when each reactor system was operating at a pseudo steady state. It is also important to note that the granular activated carbon (GAC) media in the biofilters had been exhausted prior to the beginning of the study.

Analyses

The biofilm annular reactors were analyzed for two primary parameters: biological growth within the reactors and BOM utilization or availability across the reactors. Biological growth was measured using three different methods: (1) biofilm bio-

mass using standard spread plate heterotrophic plate counts (biofilm HPCs); (2) biofilm protein content using a high sensitivity protein assay (Pierce Co., Rockford, Ill.); and (3) net biofilm growth determined by the difference between the BAR influent and BAR effluent HPC counts at steady state (Δ HPCs). All HPC spread plates used R2A agar and were incubated at 25°C for up to 10 days to optimize recovery of all microorganisms present in the oligotrophic samples [Maki et al. 1986; American Public Health Association (APHA) et al. 1989]. The biofilm samples were harvested from the annular reactors by aseptically scraping the contents of a reactor slide into a sterile test tube containing 10 mL of sterile phosphate buffer [(PBS) 8.7 g/L NaCl, 0.4 g/L KH_2PO_4 , 1.23 g/L K_2HPO_4]. The slides were repeatedly rinsed with PBS and returned to the reactor. All samples were homogenized for 1 min using a 1-cm bit (IKA Labortechnik, Charlotte, N.C.) to separate the biofilm cells.

The NOM of the various waters entering and exiting the biofilm reactors was analyzed for DOC and AOC concentrations. The DOC content of the reactor influent and effluent samples was determined according to Standard Method 5310B (APHA et al. 1989) using a Shimadzu TOC-5000 Analyzer equipped with a high sensitivity catalyst, Shimadzu, Columbia, Md. [± 0.025 mg total organic carbon (TOC)/L]. DOC samples were taken from each reactor in duplicate, and each of the duplicate samples was analyzed in triplicate. Averages and standard deviations from the mean were determined using the average of the two sets of triplicate data for each sample. The reactor influent and effluent samples were also analyzed for AOC using a modified van der Kooij method (van der Kooij et al. 1982). The modified AOC method used a *Pseudomonas* strain that had been acclimated and continuously cultured at the New York City Department of Environmental Protection Drinking Water Labs. Periodically, BDOC content of the reactor influent waters was determined at the University of New Hampshire using a 7-day, batch, acclimated sand method to allow for comparisons between various biodegradability measurements (Allgeier et al. 1996; Mercier and Collins 1998).

Influent, effluent, and biofilm samples were also analyzed for total coliform counts using both mT-7 (McFeters 1990) and m-Endo medium (Difco, Sparks, Mo.). The temperature and pH of the bulk fluid in each reactor were also monitored continuously throughout each run. All cell densities [colony-forming units/cm² and total coliform (TC)/cm²] were calculated by determining the total number of cells/cm² on the sample slide. Each HPC and coliform count was done in triplicate and included the appropriate controls and verifications.

RESULTS AND DISCUSSION

Average Results of BAR Method

Influent, effluent, and biofilm samples from each BAR were collected 15 to 18 times during each of the 2 to 3 month-long runs depending on both the season and the pseudo steady-state condition in the biofilm reactors. The final five separate pseudo steady-state samples from each reactor were averaged for the comparative analysis between the various treatment processes and seasonal changes in water quality.

Table 2 shows the summary of results from the winter run (Run A). Table 2 is divided into organic carbon biodegradability and biomass analyses, and includes the average influent, effluent, and biofilm results for each of the six reactor systems.

Indicators of Bacterial Growth Supported by Water

Indicators of biofilm growth measured by the BAR method include (1) density of HPC on the surface of the reactor (colony-forming units/cm²); (2) differences in the HPC concentrations (Δ HPC = HPC_{out} - HPC_{in}, colony-forming units/mL) in the water; (3) protein content in the biofilm ($\mu\text{g}/\text{cm}^2$); and (4) total coliforms [influent (colony-forming units/100 mL), effluent (colony-forming units/100 mL), and biofilm (colony-forming units/cm²)]. As can be seen in Table 2, all of the waters supported measurable biofilm growth under the ideal conditions provided by the biofilm reactors. The ozonated raw water supported the greatest amount of biofilm growth, further supporting previous results indicating the effects of ozonation on biodegradability and regrowth (Huck 1990; Price et al. 1993; Goel et al. 1995; LeChevallier 1999). The degree of biofilm growth varied with treatment and with the chlorinated (~ 0.35 mg/L total chlorine residual leaving the reactor) and biologically filtered waters supporting the least growth.

For all reactor runs, the TC counts found in the reactor effluents were greater than those found in the influent water, indicating that the biofilms were colonized with the organisms. As shown in Fig. 3, this was true for the ozonated source water (A5) and primary chlorinated water (A2) which had a constant effluent chlorine residual of 0.35 mg/L ± 0.05 . The ozonated raw water contained the least total number of coliforms of any of the reactor influents with an average of one coliform/100 mL; however, the few that escaped disinfection were able to attach and proliferate within the biofilm reactors resulting in high effluent (58 TC/100 mL) and biofilm coliform (117 TC/cm²) concentrations. These values were second only to those

TABLE 2. Average Results from BAR Studies—Winter Run (Run A)

Analysis	Units	A1	A2	A3	A4	A5	A6 ^a
(a) Biodegradability Measurements							
DOC _{in}	mg/L	2.69	2.80	2.17	2.21	2.80	2.15
Δ DOC	mg/L	0.16	0.00	0.07	0.06	0.19	0.06
ACO _{in}	$\mu\text{g}/\text{L}$	143	51	40	52	149	60
Δ AOC	$\mu\text{g}/\text{L}$	21	17	13	22	17	19
BDOC	mg/L	0.63	0.64	0.86	0.56	1.20	0.67
(b) Biomass/Growth Measurements							
Δ HPC $\times 10E-6$	Cells/mL	4.00	2.20	1.10	0.92	15.00	89.00
Total coliforms _{in}	Colony-forming units/100 mL	63	6	12	1	2	12
Total coliforms _{out}	Colony-forming units/100 mL	70	10	17	21	58	20
Δ Total coliforms	Colony-forming units/100 mL	7	4	7	20	56	8
Biofilm HPC $\times 10E-6$	Cells/cm ²	5.40	0.54	0.83	0.53	9.20	0.52
Biofilm total coliforms	Colony-forming units/cm ²	262	5	12	20	117	20
Biofilm protein	$\mu\text{g}/\text{cm}^2$	42	7	30	5	43	3

Note: Subscript "in" = influent water to biofilm reactor; subscript "out" = effluent water from biofilm reactor; Δ = absolute value of difference between influent and effluent concentrations.

^aA6 is BAC filtration without coagulation.

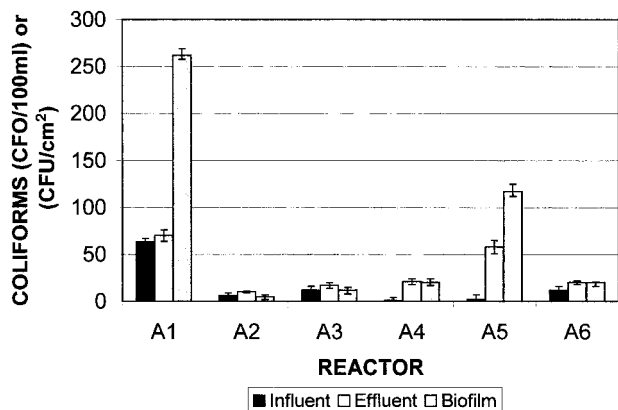


FIG. 3. Average TC Concentrations for Influent, Effluent, and Biofilm Samples from Each BAR System (Error Bars Indicate Standard Deviation from Mean)

found in the raw water. All of the filtered water effluents showed relatively low coliform numbers, even without the presence of chlorine. The biofilm coliform counts for the filtered waters were over a magnitude lower than those found in the raw water. The chlorinated raw water had the fewest influent, effluent, and biofilm coliforms. It is probable that these coliform numbers would be lower if a higher TC or secondary/final disinfection were applied as would be practiced in full-scale operation.

Biodegradability of Various Waters

Table 2 shows the removal of AOC (Δ AOC) and DOC (Δ DOC) within each of the reactor systems. Fig. 4 shows the influent AOC, BDOC, and DOC concentrations, and the Δ DOC for each of the reactors to compare the type and amount of biodegradable organic material present in each of the tested waters. In Fig. 4 it can be seen that the degradability measurements were highest for the raw (A1) and ozonated (A5) waters. However, AOC concentrations were not significantly impacted by ozonation (AOC of ozonated water compared to raw water for both runs; p -value < 0.001). It is apparent that ozonation enhanced the biodegradability of the NOM in the source water resulting in a greater utilization of DOC (Δ DOC) within the biofilm reactor (6.8% for ozonated waters and 5.9% for raw water; p -value < 0.05). Chlorination significantly decreased the AOC content in the water (143 μ g/L in raw water, 51 μ g/L in chlorinated water; p -value < 0.001), yet it showed only a slight effect on BDOC and DOC values as compared to the raw water. All of the filtration processes (A3, A4, and A6) significantly reduced the biodegradability of the organic carbon in the water as measured by reduction in influent AOC compared to the raw water influent AOC (A3 = 69%, A4 = 73%, A8 = 58%; p -values for all three compared to raw water < 0.001). Coagulant addition (A3 and A4) had only a slight influence on the biodegradability of the organic material biodegradability resulting in an average of 21.5% increase (12.5% total increase) in the removal of influent AOC compared to the noncoagulated filtered water entering reactor A6.

As seen in previous studies (Huck et al. 1991; Paode et al. 1997), AOC and BDOC values may vary unexpectedly with treatment and do not always correlate well with each other or DOC measurements. In this study, both influent AOC and Δ DOC measurements across the reactors appear to accurately describe the expected effects of the various treatment processes on the biodegradability of the test Croton Reservoir water.

Fig. 5 shows the percentages of influent DOC (%DOC) and influent AOC (%AOC) that are utilized within the BARs. Also shown is the portion of the influent DOC that is measured as

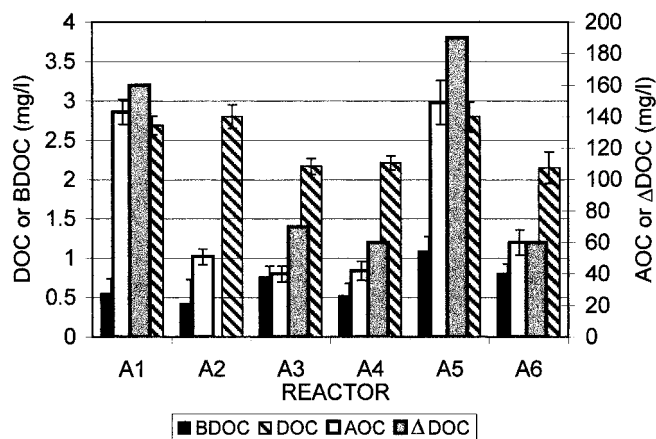


FIG. 4. Average DOC, AOC, BDOC, and Δ DOC Concentrations for Each of BAR Systems (Error Bars Indicate Standard Deviation from Mean)

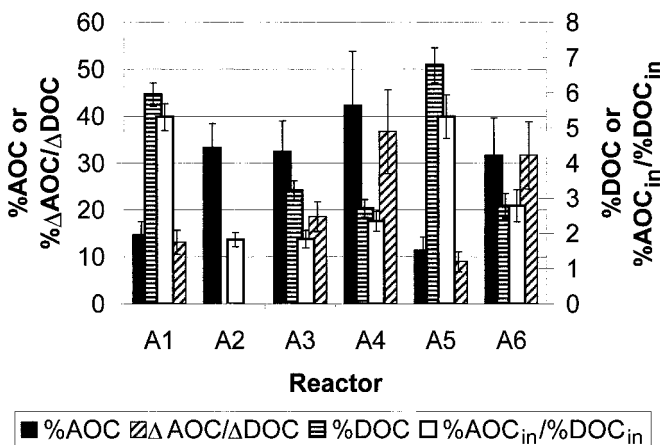


FIG. 5. Organic Carbon Biodegradability Measurements for Each BAR System: Percent Influent DOC Measured As AOC (%AOC_{in}/DOC_{in}), Percent AOC and DOC Removed within BARs, and Percent DOC Utilized Measured As AOC (Δ AOC/ Δ DOC) (Error Bars Indicate Variance of Each Quotient)

AOC (AOC/DOC) and the proportion of DOC utilized in the BARs that is in the form of AOC (Δ AOC/ Δ DOC; mg AOC/mg DOC). These values illustrate the types and amounts of NOM that leave the various treatment processes and become available for utilization within the biofilm reactors. It can be seen that ozonation (A5), which precedes all of the filtration processes, significantly increases the biodegradability of DOC (%DOC, ozonated water 6.8% reduction, raw water 5.9%; p -value < 0.05) in the raw water. Although ozonation did increase the amount of AOC in the water, it did not result in greater biological utilization within the annular reactors when compared to the raw water (A5 %AOC = 11%, A1 %AOC = 14.6%: A5 % Δ AOC/ Δ DOC = 8.5%, A1 % Δ AOC/ Δ DOC = 13%). The fraction of AOC/DOC decreases as a result of all filtration processes indicating that the biofiltration with either anthracite or exhausted GAC preferentially removes the AOC fraction of NOM. It appears that the biofiltration process removes the most readily biodegradable organics in the water, leaving the more recalcitrant fractions to be removed in the biofilm reactors. This suggests that a significant fraction of the AOC and DOC that escapes treatment is still available to promote regrowth. The anthracite (A3) and GAC (A4) direct filters produce waters that have very similar biodegradability, although GAC filters demonstrated a higher affinity for removing the AOC fraction of DOC (A4 % Δ AOC/ Δ DOC = 36.5%, A3 % Δ AOC/ Δ DOC = 18.5%). Fig. 5 also shows that coagulation and flocculation prior to filtration resulted in a

slight, but statistically insignificant increase in the biodegradability of the organic material in the water (%AOC and %DOC for A3 and A4 compared to A6; $0.1 > p\text{-values} > 0.05$).

Correlations between Biofilm Growth and Biodegradability

Correlations of pseudo steady-state biofilm growth characteristics and biodegradability were developed using the averaged data for the winter (Table 2) and the summer run (data not shown). The most significant correlations were used to develop biostability guidelines for Croton Lake water and aid in the design and operation of the full-scale biofiltration systems.

Correlations between biofilm protein and both influent AOC and Δ DOC are shown in Figs. 6(a and b). Influent AOC concentration did not demonstrate a particularly strong correlation with biofilm protein content ($R_A^2 = 0.63$, $R_B^2 = 0.61$), while the correlation between Δ DOC and biofilm protein content was quite good, especially for the winter run ($R_A^2 = 0.77$, $R_B^2 = 0.93$).

The correlations between both biofilm HPC and influent AOC and Δ DOC are shown in Figs. 7(a and b). The relationship between biofilm HPC and influent AOC was significant for all runs, showing the strongest relationship in the winter run ($R_A^2 = 0.89$, $R_B^2 = 0.64$). (Note that these relationships are linear; the trend lines appear curved because of the log scale x -axis which is needed to show the complete data on the figure.) The correlation between biofilm HPC and Δ DOC was excellent for both runs, showing a strong relationship between the biodegradability of the organic carbon in the water and the amount of biomass that can be supported ($R_A^2 = 0.96$, $R_B^2 = 0.92$).

The relationships between Δ HPC (net growth across the reactor at steady state) and both influent AOC and Δ DOC are shown in Figs. 8(a and b). The correlation between Δ HPC and influent AOC was significant and similar for both runs ($R_A^2 =$

0.61 , $R_B^2 = 0.61$). The correlation between Δ HPC and Δ DOC was good for the winter run, but insignificant for the summer run ($R_A^2 = 0.73$, $R_B^2 = 0.1$).

As can be seen in all of the correlations, the relationships are typically stronger for the winter run. This may indicate that the winter run had reached a more consistent steady state

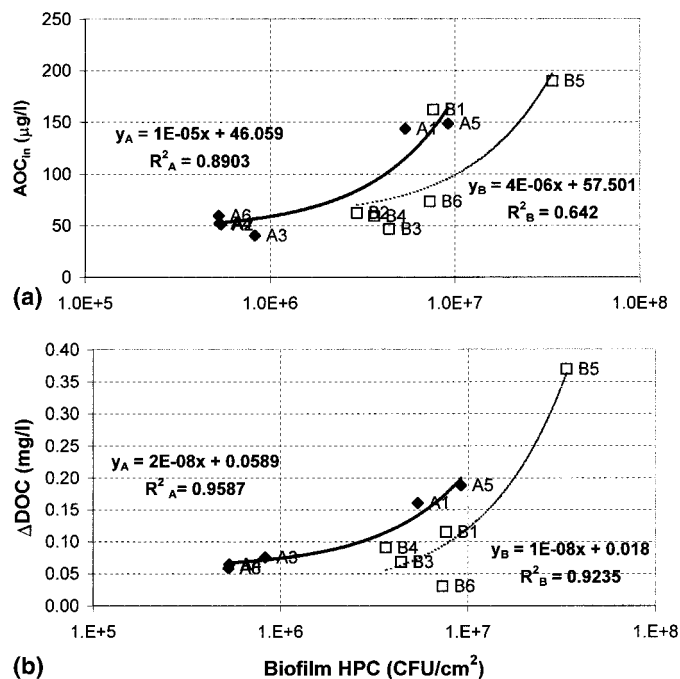


FIG. 7. Correlations between: (a) Influent AOC Concentrations and Biofilm HPCs; (b) Δ DOC Concentrations with Biofilm HPCs for Each of BAR Systems [Linear Regressions with R^2 Values for Both Winter (A) and Summer (B) Runs Shown]

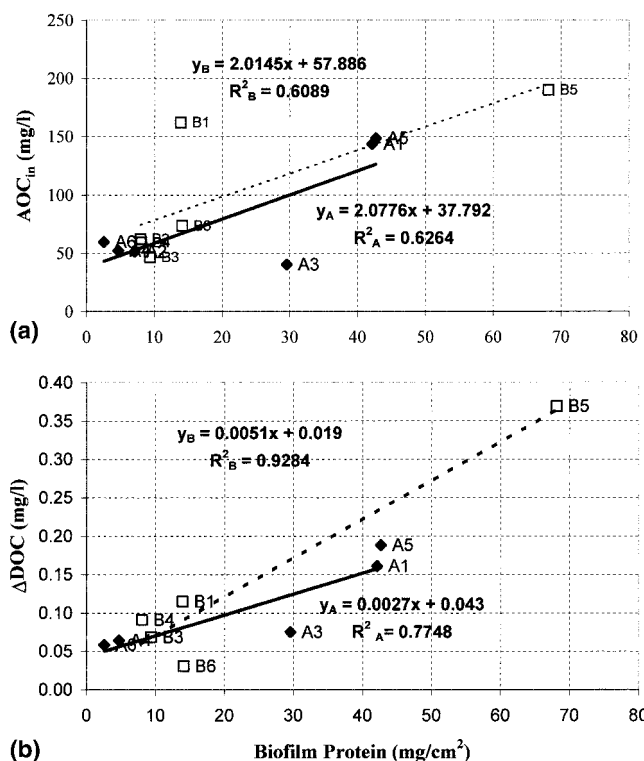


FIG. 6. Correlations between: (a) Influent AOC Concentrations and Biofilm Protein Production; (b) Δ DOC Concentrations and Biofilm Protein Production for Each of BAR Systems [Linear Regressions with R^2 Values for Both Winter (A) and Summer (B) Runs Shown]

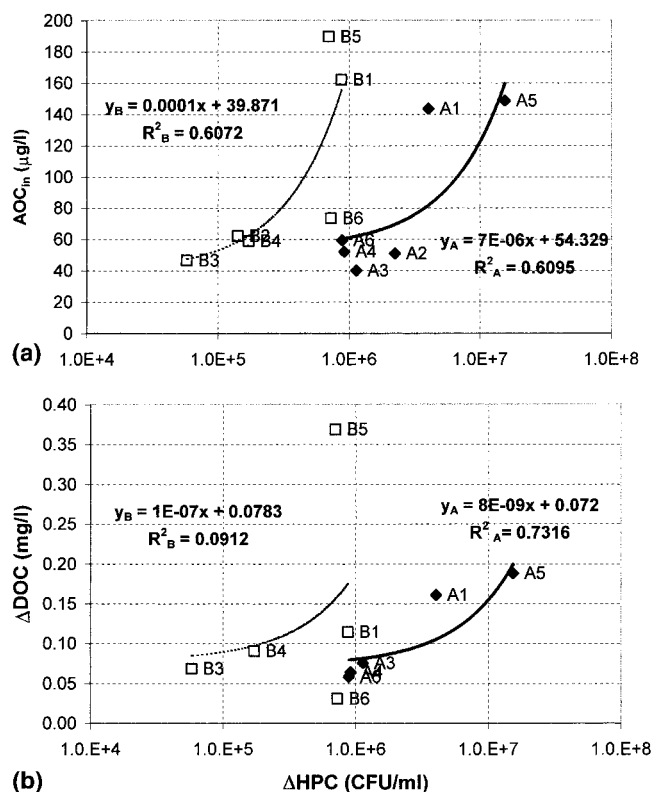


FIG. 8. Correlations between: (a) Influent AOC Concentrations and Δ HPC (Net Growth Rate); (b) Δ DOC Concentrations and Δ HPC (Net Growth Rate) for Each of BAR Systems [Linear Regressions with R^2 Values for Both Winter (A) and Summer (B) Runs Shown]

than the summer run. Additionally, the variability in the summer run data was greater than that of the winter run, which may be attributed to changes in both source water quality and pilot plant operation during the summer run. Changes in water quality and plant operation are one of the major concerns when using the biofilm annular reactor method to assess piloting operations, and can require longer biofilm reactor run times and increased data collection and analysis efforts.

In most of the correlations there were evident groupings among the filtered waters (reactors 3, 4, and 6) and the chlorinated waters (reactor 2) in both runs. The raw and ozonated waters typically grouped near each other but in regions of relatively high biological growth and/or high biodegradability. In Figs. 7(a) and 8(a), it can be seen that there is a gap between 140 $\mu\text{g/L}$ AOC and 60 $\mu\text{g/L}$ AOC, which separates the filtered and chlorinated waters from the raw and ozonated waters. In addition, it can be seen in Figs. 6(b), 7(b), and 8(b) that all of the filtered waters had a ΔDOC of 0.10 mg or less, whereas the raw and ozonated waters all exceeded 0.10 mg/L utilization of DOC.

Development of Biostability Factors

The apparent relationship between biodegradability and the biofilm growth characteristics, as demonstrated in Figs. 6–8, suggest possible expressions for the development of biostability factors (BFs). Biostability factors may provide a single parameter to gauge the relative biostability of a specific water based on both the biodegradability of the NOM in the water and the amount of bacterial biofilm growth the water can support. Over 24 different expressions were developed and tested using the data from these pilot plant studies. The different expressions were calculated by looking at the correlations and general trends obtained when applying the expressions to data collected from each individual treatment process. Then the expressions that most accurately described the data and showed the strongest correlations for all of the treatment processes were chosen as the expressions that best describe the biostability of the specific water tested in terms of organic carbon biodegradability and biofilm growth potential. Of all of the potential biostability factors examined, four expressions were found to best predict the relative biostability of each of the waters tested. The relative accuracy of each predictive expression was determined by quantitative and qualitative assessments of the complete biostability, biodegradability, and re-growth results obtained from the Croton Pilot Plant Study (Mercier 1988; Sharp et al. 1998, 1999). The factors that were found to be most significant for the Croton Lake water are shown in (1)–(4). Units are not considered in these equations

$$\text{BF}_1 = \log(\text{biofilm HPC}) \cdot \Delta\text{DOC} \quad (1)$$

$$\text{BF}_2 = \log(\text{biofilm HPC}) \cdot \text{AOC}_{\text{in}} \quad (2)$$

$$\text{BF}_3 = \log(\Delta\text{HPC}) \cdot \Delta\text{DOC} \quad (3)$$

$$\text{BF}_4 = \log(\Delta\text{HPC}) \cdot \text{AOC}_{\text{in}} \quad (4)$$

The BF values were compared to the established relative biostability of each water as determined in the complete biostability study, and BF values separating the relatively biologically stable and unstable waters were established. These results are shown in Table 3. For all the BF expressions, the higher the BF value, the less biologically stable the water; and conversely, the lower the BF value the more biologically stable the water. Despite the different biofilm growth characteristics (biofilm HPC and ΔHPC) there is a similarity between BF_1 and BF_3 ($\text{BF}_1 = 0.55$, $\text{BF}_3 = 0.50$), and BF_2 and BF_4 ($\text{BF}_2 = 425$, $\text{BF}_4 = 400$). This similarity lends support to the relevance of these biostability factors, but the dampening effect on the

TABLE 3. Relative Biostability of Each Tested Water As Determined by Value of Specific Biostability Factors

Reactor	Biostability Factors				Relative biostability
	BF1	BF2	BF3	BF4	
A1	1.08	963	1.06	944	US
A2	N/A	292	N/A	323	S
A3	0.41	237	0.42	242	S
A4	0.34	298	0.36	310	S
A5	1.32	1,038	1.36	1,069	US
A6	0.34	343	0.36	357	S
B1	0.76	1,116	0.65	963	US
B2	N/A	402	N/A	319	S
B3	0.47	312	0.33	224	S
B4	0.59	388	0.47	309	S
B5	2.79	1,431	2.16	1,111	US
B6	0.21	501	0.18	428	US

Note: Estimated separation between relatively stable and unstable water for each biostability factor: $\text{BF}_1 = 0.55$, $\text{BF}_2 = 425$, $\text{BF}_3 = 0.50$, and $\text{BF}_4 = 400$. S = relatively biological stable water; US = biological unstable water; N/A = not available due to chlorine interference.

values due to the log transformation should also be noted. It should also be noted, that these biostability factors do not take into account the type of pipe material or the effects of final disinfection; they are strictly a measure of the biofilm growth potential of each specific water and how various treatment processes effect that potential.

Comparison of Treatment Trains

The BF was used to rate the waters from most biologically stable to least biologically stable. These BF ratings were compared to the relative biostability rating given for the same waters by the Croton Reservoir Pilot Plant Biostability Study (Mercier and Collins 1998; Sharp et al. 1999). The relative biostability ratings for the waters tested were determined by qualitatively comparing the complete biofilm growth and organic carbon biodegradability results obtained during the 2-year pilot study. The quantitative and qualitative ratings for each of the waters tested using the BAR method are given in Table 4. The two rating systems provide very similar relative rankings for all of the waters tested in both the winter and summer runs. The only disagreement between the two rating systems occurred when considering the biostability of the two direct filtration waters (GAC filters A4 and B4; anthracite filters A3 and B3). The differences in biostability between the two direct filtered waters were difficult to distinguish in the overall study. However, because the BFs are numerical values calculated from several pertinent quantitative parameters, it was possible to distinguish between the relative biostability of the two direct filtration trains in both runs. BFs gave a slight advantage to the anthracite filtered waters (A3 and B3), when compared to the GAC direct filtered waters (A4 and B4). These results demonstrate how biostability factors may be more sensitive and useful for evaluating and rating water re-

TABLE 4. Relative Biostability Ratings for Each Water Tested

Order of relative stability	Winter Run (A)		Summer Run (B)	
	Biostability study rating	Biostability factor rating	Biostability study rating	Biostability factor rating
Most stable	A6	A6	B3 = B4	B3
↑	A4	A3	—	B4
	A3	A4	B2	B2
	A2	A2	B6	B6
	A1	A1	B1	B1
Least stable	A5	A5	B5	B5

growth potential than qualitative observations. Although more data are needed, and further study is required to validate the use of BFs for other waters, the quantitative biostability factors presented here and the annular reactor studies used to obtain them, proved to be an excellent method for evaluating the overall regrowth potential of the Croton Reservoir-piloted waters. In addition, the quantitative results provided by the biostability factors were used in treatment process selection and design of the full-scale Croton Reservoir Treatment Plant.

It should be noted that the BF values (Table 3) selected to differentiate between relatively stable and unstable waters are only arbitrary points suggested by the equations. In actuality, the area between unstable water and relatively stable water is a large region with only generally defined borders. This region will vary depending upon source water characteristics, seasonal water quality changes, and treatment. Due to the variability of source waters, further study and evaluation are required to establish the validity of this biostability factor method, and subsequent studies will provide the needed data. However, the results presented here illustrate the value of biofilm-based methods for measuring the biostability of treated waters, and suggest that a standard method for characterizing the biostability of drinking water is possible using a combination of biofilm-based methods and standardized NOM measurements. Furthermore, the results demonstrate that biofilm-based methods are effective in evaluating piloting operations, and can provide meaningful data for process selection and final design of drinking water treatment plants.

SUMMARY

The reduction of biodegradable organic matter (BOM) concentration is perhaps the most effective means of controlling (or reducing) biofilm growth in the distribution system. However, measuring and monitoring biodegradable organic matter for evaluating treatment performance or monitoring water biological stability in distribution systems remains a major challenge for the drinking water industry. Additional tools for measuring the biostability of water are needed and the biofilm annular reactor (BAR) method is a tool with greater sensitivity and flexibility than the batch bioassay methods currently being used. The BAR method is a continuous flow dynamic system utilizing indigenous bacteria in the water stream, with the ability to simulate the distribution system. This method can give an indication of the consequences of water quality, treatment process, and plant operation on a variety of biostability parameters, including biofilm formation potential. The BAR method can be used in conjunction with standard methods (AOC and BDOC) to fully assess the biostability of drinking water.

The studies presented demonstrate how the BAR method can be used to obtain both biofilm-based parameters and organic carbon biodegradability measurements to thoroughly and effectively assess the biological stability of water. The results obtained from these studies were used to both qualitatively and quantitatively compare each water tested with respect to its regrowth potential. Furthermore, the quantitative development and use of biostability factors derived from these results proved to be valuable in the selection and design of the full-scale treatment plant.

The results presented demonstrate that organic carbon biodegradability measurements alone do not adequately describe the biostability of a given water. Depending on treatment, the standard organic carbon biodegradability measurements (AOC, BDOC, and DOC) can give contradictory results as to the biostability of a given water. However, a combination of biofilm-based methods and standard NOM measurements can be used to directly compare treatment process and evaluate the effectiveness of numerous design parameters such as filter media type, coagulant addition, and disinfection type. In this

study, the value of the biostability factors for the various piloted waters were compared to establish which treatment processes and operating conditions produced the water with the greatest biological stability. The biostability factor results indicated that coagulation prior to filtration had little effect on biostability. The results also showed that GAC and anthracite filter media produced essentially the same quality of water with respect to biostability and regrowth potential. All of the piloted treatment processes produced waters that were equivalent or more biologically stable than that currently being distributed to New York City consumers, with the dissolved air flotation followed by either GAC or anthracite filtration producing the most biologically stable water. Finally, the presence of a chlorine residual was shown to greatly increase the apparent biostability of the water, but did not completely remove the biofilm growth potential of the water. The chlorine residual repressed the biofilm growth potential of the water, and if removed, that potential could be realized in the distribution system.

The results from this study were used to select the best treatment processes and design the full-scale drinking water filtration plant for Croton Reservoir source water. The combined use of organic carbon biodegradability and biofilm growth potential measurements allowed for the biological stability of each piloted water to be qualitatively and quantitatively characterized to assist in process selection and design. In addition, this study provided fundamental insights into the effects that various treatment processes have on the biological stability of drinking water. The results illustrate the need for better tools and methods to adequately characterize the regrowth potential of drinking water so that higher-quality water can be provided to the public to protect public health and to minimize the deterioration of distribution systems.

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