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# ATTACHMENT AND FATE OF CARBON FINES IN SIMULATED DRINKING WATER DISTRIBUTION SYSTEM BIOFILMS

PIERRE MORIN and ANNE K. CAMPER*

Center for Biofilm Engineering, Montana State University, 409 Cobleigh Hall, Bozeman, MT 59717, U.S.A.

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**Abstract**—Concern over the release of colonized carbon fines from filters has increased with the interest in operating granular activated carbon filters optimized for biological activity. These fines may transport bacteria to the distribution system and become entrained in biofilms. The capture and release of carbon fines of two size ranges (1.2–50 μm and 1.2–8 μm) in biofilms and their impact on the biomass was studied using a bench-scale simulated drinking water system. Size of the particles was important, with large carbon fines (1.2–50 μm) remaining in the biofilms while the small ones (1.2–8 μm) were entrained and then detached. The presence of carbon fines did not influence the number or activity of bacteria present in the biofilms. Disinfection assays on biofilms containing particles were performed with chlorine (0.5 mg/L) and monochloramine (1 mg/L). Even in the presence of substantial numbers of carbon fines, the effect of the disinfectants on the biofilms as determined by destructive analysis and direct microscopic observation was limited. Carbon fines were released from the biofilms during disinfection.© 1997 Elsevier Science Ltd. All rights reserved

*Key words*—carbon particle, biofilm, disinfection, water treatment

## INTRODUCTION

Granular activated carbon (GAC) has been used widely and successfully for decades in the area of water treatment (Schulhof, 1979). In drinking water treatment, GAC filters remove a wide range of organic and inorganic compounds, and improve taste and odor (Rittmann, 1990; DeWaters and DiGiano, 1990; Loper *et al.*, 1985). Research in the last few years has shown that carbon particles from GAC filters were released in the water effluent, similar to sand particles from classical filters (Camper *et al.*, 1987; Stringfellow *et al.*, 1993). However, unlike sand, carbon fines (CF) combine large surface area with high affinity for bacteria. Several authors have demonstrated that these particles can be colonized by heterotrophic and coliform bacteria (Stewart *et al.*, 1990; Camper *et al.*, 1986). This issue has received increased attention with the development of biological activated carbon (BAC) filtration. Biological activated carbon filtration units are composed of GAC on which a biofilm is cultivated (Servais *et al.*, 1992). The BAC process has been developed to decrease the concentration of specific organic compounds called biodegradable organic matter (BOM) via bacterial metabolism. The BOM level in the water is decreased, and therefore deprives downstream bacteria of nutrients (Rittmann and Huck, 1989; Bouwer and Crowe, 1988; Cauchi and Servais, 1993). This increases the biological stability of drinking water and limits bacterial regrowth (Rittmann and Snoeyink, 1984; LeChevallier *et al.*, 1992). Since colonized CF have been found in GAC filter effluents, and the bacteria attached to the fines were resistant to disinfection by chlorine (Stewart *et al.*, 1990), questions have arisen about the potential health risks of the BAC process.

Although the efficacy of the BAC process is recognized, concerns exist with respect to the possible seeding and subsequent regrowth of undesirable bacteria, such as coliform and pathogenic bacteria, in the network. It is known that heterotrophic bacteria carried by particles can penetrate the disinfection barrier, attach to the pipe surface, grow, and develop into a biofilm; this becomes critical if pathogens are involved (LeChevallier, 1990; McEnroe *et al.*, 1992). It is therefore important to understand and characterize the relationships that exist between the released GAC particles and the biofilm existing in the network. This is nearly impossible to accomplish in full-scale distribution systems due to their complexity. Therefore, laboratory biofilm reactors operated under conditions relevant to drinking water were used to study extreme conditions of CF loading. Two specific objectives were established. Since particle

*Author to whom all correspondence should be addressed [Fax: 406-994-6098].

size affects transport rate and the potential number of attached bacteria, we wished to compare the transport and incorporation of two size ranges of CF. Second, the gross behavior of imbedded particles and the associated biofilm as well as the bacteria in the immediate vicinity of the fines were evaluated to determine if the fines afforded protection from disinfection.

## MATERIALS AND METHODS

### *Experimental system*

Biofilms were grown in polycarbonate annular reactors, previously described by Siebel and Characklis (1991). The reactors have 12 removable slides that allow periodic biofilm sampling. The rotational speed of the inner cylinder created a shear stress simulating a pipe velocity of 0.3 m/s. The wetted surface for each reactor was 0.19 $m^2$, with a liquid volume of 0.6 L. The reactors were fed with biologically treated water from a two column system operated in an upflow mode. This system consisted of a GAC filter followed by a biological activated carbon filter. The columns were used to remove natural biodegradable organic matter (BOM) from tap water, and also to act as a source of consistent indigenous microbial inoculum for the annular reactors. The utilizable carbon in the column effluent, as measured by the AOC test of van der Kooij (van der Kooij *et al.*, 1982) was always less than 20 $\mu$g C/L. The treated water was supplemented with an assimilable organic carbon (AOC) solution (final concentration 0.5 mg C/L) and a nitrate buffer solution (0.1 mg/L each phosphate and nitrate). The AOC solution consisted of equimolar concentrations based on carbon content of acetic acid, benzoic acid, proprionaldehyde parahydroxybenzoic acid, and ethanol. These carbon sources do not exert a measurable chlorine demand. Effluent AOC concentrations were typically 100–150 $\mu$g C/L. To avoid the presence of external CF in the reactor, a 0.2 $\mu$m filter was located between the water dilution reservoir and the reactors. A residence time of 2 h was used to minimize the contribution of planktonic growth of the HPC so a bacterial mass balance across the reactor reflected net attachment or detachment of cells from the surface. The reactors were partially immersed in a temperature-controlled water bath at 20 $\pm$ 1°C.

### *Microbial population*

A mixed population biofilm was grown in the reactors under conditions relevant to drinking water distribution systems. The source of the bacteria was water from the laboratory columns which contained approximately $1 \times 10^4$ HPC/ml, as enumerated on R2A medium. To minimize the introduction of extraneous carbon particles from the columns but allow for sufficient inoculation, the reactors were initially run for 5 h with settled column effluent. After this time the systems were switched to filtered column effluent.

### *Carbon fines characteristics and colonization*

Two different CF size ranges (1.2–50 $\mu$m and 1.2–8 $\mu$m) were to study the transport of fines to established biofilms. A suspension of the smaller CF was obtained by washing and filtering powdered activated carbon through a 50 $\mu$m screen, followed by filtering the screened suspension through a double 8 $\mu$m cellulose acetate filter. The average size of the carbon fines in the filtrate was 2.09 $\mu$m as determined by the observation of at least 150 CF with the image analysis system. A suspension of larger fines was obtained by washing and filtering powdered activated carbon through an 8 $\mu$m cellulose acetate filter. The particles on the surface on the filter were removed by washing with a known volume of water. The average size of the fines for this suspension was 7.88 $\mu$m (count of at least 60 CF). For the second part of the study, improvements were made to produce CF of an average size of 11.5 $\mu$m (count of at least 50 CF). Colonization was accomplished by mixing the fines suspension with a HPC solution containing $3.4 \times 10^6$ bacteria/ml for 2 days. The inoculum was grown in a glass bead column reactor receiving the same dilution water used for the annular reactors.

### *Disinfectants*

Chlorine and monochloramine solutions were prepared using commercial bleach (5.25%) diluted with nanopure water. Monochloramine was prepared by reacting bleach with ammonium chloride at a ratio $N - NH_3/Cl_2 = 1:3$ (pH 8.3). Solution concentrations were measured using a DPD digital chlorine colorimetric kit (Lamotte).

### *Staining solutions*

The DNA-binding fluorochrome 4′6-diamino-2-phenylindole (DAPI) was purchased from Sigma Chemical Co. (St Louis, MO, U.S.A.), the redox dye 5-cyano-2,3-ditolyl tetrazolium (CTC) from Polyscience Inc. (Warrington, PA, U.S.A.) and the fluorescent dye rhodamine 123 from Eastman Kodak Co. (Rochester, NY, U.S.A.).

### *Sampling and analytical methods*

Reactors were run at least 10 days before sampling to allow for adequate colonization. Previous results obtained under identical conditions showed that steady-state numbers of biofilm HPC were achieved within 7 days operation (data not shown). Biofilm was sampled 1 h before the addition of the CF suspension, and HPC and CF numbers determined. The reactors were then emptied and refilled with a suspension of CF containing a concentration of approximately $2.5 \times 10^6$ CF/ml for the smaller size range and $2 \times 10^5$ CF/ml for the larger particles. These concentrations greatly exceed the measured average CF concentration in backwash water from a full-scale biological filter of $6 \times 10^3$ CF/ml. Two different sampling procedures were used for the two parts of the study. For the first part of the study (attachment of CF to the biofilm), an additional set of samples was removed immediately after refilling the two reactors. A total of 11 biofilm samples were collected over a 24-h period. Biofilm with the associated CF was scraped from the slides (surface area between 22.5 and 27.5 $cm^2$) and suspended in 10 ml of water. Each sample was homogenized for 30 s at 100 rpm with a tissue homogenizer (Tekmar). The homogenized sample was split and used for HPC and CF size analysis. The size and the mass of the CF were determined by exposing the suspension for 5 min to a 8.5% phosphoric acid solution to remove biological material, followed by filtration through a 1.2 $\mu$m cellulose acetate membrane. The filter was examined under a microscope at 1000 × magnification. Counting and sizing of the CF was carried out with the aid of a computerized image analysis system (American Innovision). The fines were assumed to have a rectangular shape, thus allowing their averaged length and surface area to be used to calculate the particle mean volume. The mass of fines for comparison between initial and final samples was obtained by using the particle mean volume and the carbon density value as provided by the manufacturer. HPC were evaluated by the spread plate method on R2A medium; CFUs were counted after 7 days incubation at 20°C.

In the second half of the study, which used pre-colonized fines at a final concentration of *ca.* $3 \times 10^5$ CF/ml, the first sample was taken 24 h after the addition of the CF suspension. Immediately after the first sampling, one reactor was continuously fed a disinfectant solution (influent concentration of 0.5 mg/L chlorine or 1 mg/L monochloramine), while the other received an equal amount of nanopure water. This produced a residual of 0.1 $\pm$ 0.03 mg/L free chlorine and the monochloramine

residual was never less than 0.6 mg/L. A third reactor was set up for the trials with chlorine, to act as double control (no CF, no oxidant). Two slides were removed weekly from each reactor for a total of 6 weeks. The first slide was analyzed as described above. The first half of the second slide was scraped into water and prepared for the enumeration of active and total bacteria. Total bacteria were estimated by DAPI staining (0.1 g/L) and direct microscopic count, while active bacteria were enumerated using two dyes, CTC (final concentration 5 mM) and rhodamine 123 (5 g/L) (Yu and McFeters, 1994; Schaule *et al.*, 1993; Rodriguez *et al.*, 1992). Biofilms on the second half of the slide were stained intact for direct microscopic observation of the organisms relative to CF. During the first replicate of the disinfection experiment with chlorine, we observed the total disappearance of CTC activity in the treated reactor after 1 week. Therefore, rhodamine 123 was used to stain active bacteria in subsequent experiments.

*Control reactors*

Two control reactors were run following the sampling and analytical method described above. In one of these reactors, a thick biofilm was grown by replacing the AOC solution with a glucose solution at 20 mg/L. The other control consisted of a sterile reactor.

## RESULTS

*Attachment of carbon fines*

These experiments were designed to simulate conditions where filter fines enter a distribution system and contact biofilms growing on pipe walls. It was of interest to know if the size of the particles was important, since larger fines would be capable of transporting more bacteria. It was found that there was a difference in the behavior of the two size ranges of carbon particles (Fig. 1). The small fine concentration (Fig. 1B) increased on the slides for the first 4 h to a concentration representing approximately 20% of added fines, and then decreased for a few hours to a concentration at 24 h of at most 5%. In contrast, the percentage of larger particles incorporated on the surface initially reached 70% of those added, declined after 1 h, and remained at a level representing either 30 or 52% of the added particles. Forty-five percent of the particles in the large fine suspension added to the reactors were longer than the average 7.88 $\mu$m, and over 75% of the particles were larger than the average size of the fines used in the first experiments. Based on these observations, calculations to assess the relative importance of the particle size relative to retention in the biofilm were made. Table 1 presents ratios of the mean mass of fines in the biofilms between last and first samples for the different experiments. These results indicate that the mean particle mass did not change significantly when the small fines were added, but there was about a 10-fold increase in mean particle mass when the large fines were added. Another consideration is the potential ability for a particle to transport bacteria to a biofilm. By assuming the % retention in Fig. 1 and using the measured length and width of remaining carbon particles, the amount of CF surface area in the biofilm after 24 h was approximately seven times higher when larger particles were added. When it is considered that a 1.2 $\mu$m fine is approximately the same size as a bacterium and is therefore unlikely to transport undesirable organisms to the biofilm, the potential importance of large fines becomes even more significant.

Since the biofilms in the above experiments were thin (*ca.* 60% surface coverage), an additional control experiment with a thick biofilm was conducted to determine whether there was a lack of binding sites for CF attachment to the thin biofilms. The small and large particles again showed different responses (Fig. 1). There was no statistical difference ($p = 0.05$) between the three regular trials with thin biofilms and the thick biofilm control for the attachment of small fines (Fig. 1B). However, the attachment of large CF was enhanced in the thicker biofilm (Fig. 1A). It therefore appears that there was no lack of available sites in the biofilm for the attachment of the small CF, and that larger fines have a higher affinity for the fixed biomass.

Another control experiment focused on the interaction between CF and the polycarbonate in a sterile reactor. The concentration of particles on the uncolonized slides (average $23.2 \times 10^3$ CF/cm$^2$) remained constant over the course of the experiment (24 h) (Fig. 1B). The recovered CF were believed to be present in the water drops adhering to the slides when they were removed from the reactors.

*Impact and fate of CF embedded in biofilm*

Once it had been established that larger CF were retained in biofilms, the long-term impact of these particles on biofilm behavior was investigated. One reactor received a slug dose of colonized large fines (average size 11.5 $\mu$m) while the other was operated as a control. There was no difference in the numbers of biofilm heterotrophs and total bacteria (Fig. 2) between the reactors over the 40-day time period following CF addition. In the dosed reactor, the ratio of CF to culturable cell numbers was in the range of 1:2 to 1:10, while the control reactor contained a negligible number of fines. Therefore, the total number of bacteria in the reactor was controlled by factors other than the amount of CF.

Carbon fine addition did not result in elevated numbers of bacteria in biofilms, but it was unknown if the presence of the particles would provide a protected local environment from disinfection. This was of particular interest since the disinfectant residuals in the reactors with carbon fines were essentially the same as those obtained in similar experiments where no fines were added (data not shown). Therefore parallel reactors received a suspension of large (average size 11.5 $\mu$m) carbon particles with one subsequently being disinfected. Biofilms were destructively sampled for total cell counts (DAPI), culturable bacteria (plate counts), respiring cells (CTC or rhodamine 123), and CF

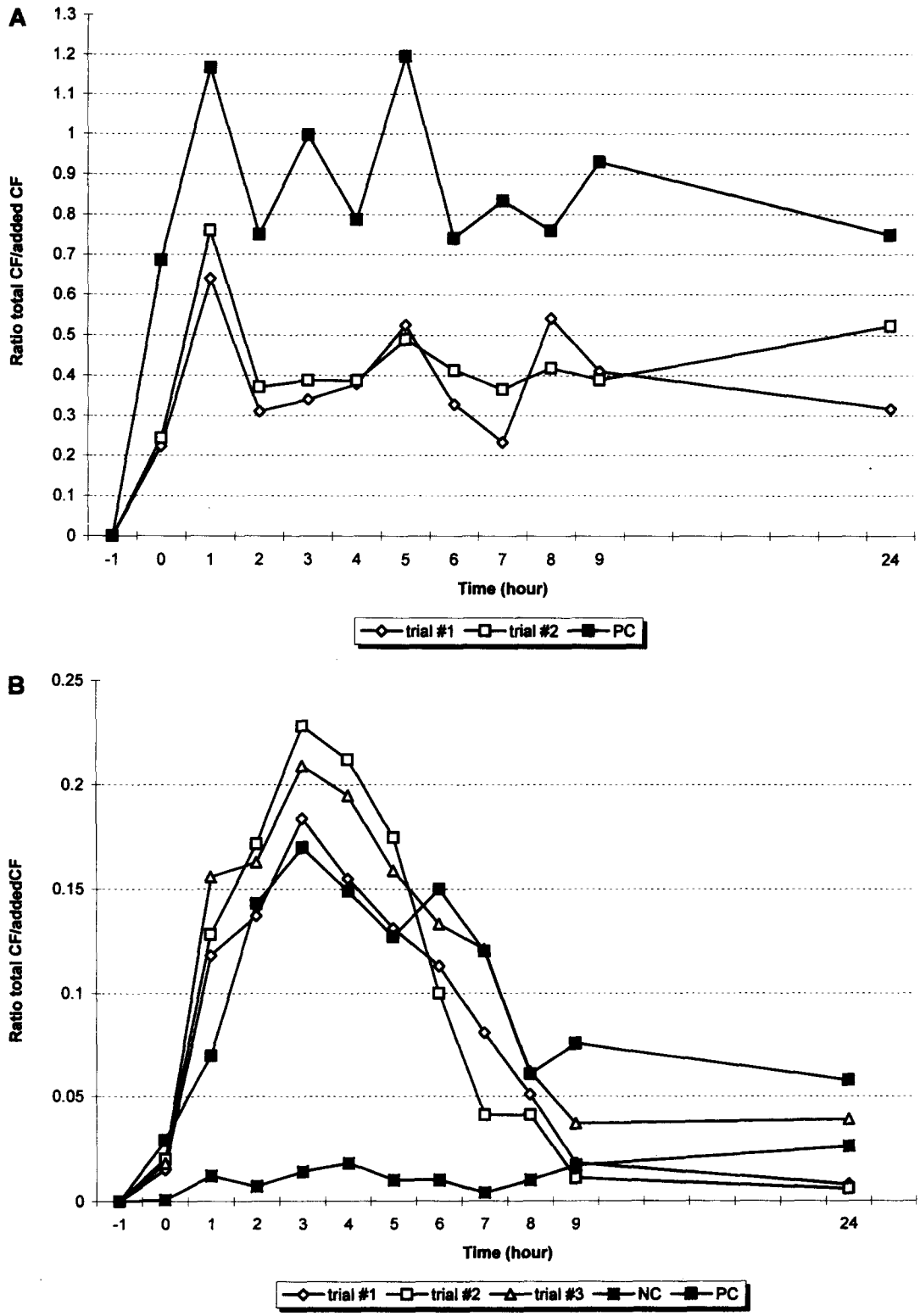



Fig. 1. Attachment of carbon fines to biofilm. (A) Large carbon fine suspension (size range 1.2–50 $\mu$m). (B) Small CF solution (size range 1.2–8 $\mu$m). Positive control (PC) is the control experiment run with a thick biofilm and negative control (NC) was run with no biofilm. The data have been transformed by subtracting the average number of background carbon fines ($n = 4$) from each experiment and expressing the values as a ratio of remaining fines to those added.

Table 1. Mass ratios of carbon fines in biofilm between last and first samples ($t = 24/t = 1$) for experiments with the small (1.2–8 $\mu$m) and the large (1.2–50 $\mu$m) size ranges. The length and surface area of particles for each sampling time was used to obtain a volume ($V$ = surface area$^2$/length). The apparent density provided by the manufacture (503 kg/m$^3$) was used to determine the total average mass

| | Replicate 1 | Replicate 2 | Replicate 3 | Thick control |
|---|---|---|---|---|
| Small CF | 1.28 | 0.90 | 1.56 | 2.48 |
| Large CF | 10.47 | 11.23 | — | 22.94 |

concentration. In addition, CF site-specific information (microscopic observations of intact biofilms) was compiled. Figure 3 shows the bacteriological results for the chlorine and the monochloramine disinfection assays for scraped biofilms containing added colonized CF. Chlorination caused a decrease of 99.9% in the number of CFUs in the biofilm over a 7 day period. The effect of chloramination on the viable bacterial population was slower, resulting in a 99.6% decrease in viability by day 46.

Figure 4 presents the results of active cell counts for the disinfection assays. A dramatic decrease in bacterial activity was observed for chlorinated reactor 2 (Fig. 4A) when measured by CTC. Since growth was monitored by the plate count technique for the same sample, the disparity between the two techniques indicated that CTC was not working properly in the conditions of our experiment. CTC was then replaced by rhodamine 123, which measures transmembrane potential. The decrease in bacterial activity was limited for both agents and never greater than 0.7 log over the time period of the experiments (Fig. 4A and B, reactor 2′). However, it does appear that chlorine resulted in a slightly greater decrease in bacterial activity.

The effect of both disinfectants on the detachment/release of biofilm organisms and carbon fines was determined by comparison of control and disinfected biofilms. Biofilm removal was assessed by scraped total cell counts. Chlorine appeared to result in greater biofilm removal than monochloramine (Fig. 5), with log decreases of 0.85 and 0.61 log or 0.07 and 0.36 log, respectively. The number of fines greater than 5 $\mu$m in the biofilm from the same slides was also determined. A decrease of CF concentrations in disinfected biofilms was apparent with both treatments (Fig. 6) with a steady-state concentration reached in approximately 3 weeks. This reduction was substantial, with a loss of nearly 80% of the large carbon particles.

Knowing that there was a reduction in both the number of large carbon fines and bacteria, further comparisons were made between the culturable cell counts and CF numbers. At each sampling time, the ratio of large (>5 $\mu$m) particles to colony forming units was determined. This ratio increased by at least 100 fold in the disinfected reactors with time. However, the final ratios were still 1:1 or above at steady state, demonstrating that carbon fines persisted in significant numbers relative to the culturable bacterial population. The ratio of CF to bacteria in the control reactors remained unchanged for the duration of the experiment.

The average biofilm responses over the entire sampling surface were compared to observations taken microscopically of total and respiring cells in the vicinity of carbon particles in directly stained biofilms. Over 1200 observations were made with approximately 500 from disinfected reactors. In control reactors, there was no evidence of increased numbers or enhanced activity of bacteria next to carbon particles. When either disinfectant was applied, there was no preferential retention of total or active bacteria associated with carbon particles.

## DISCUSSION

Although studies on the release of colonized fines from filters were conducted nearly a decade ago (Camper *et al.*, 1986), the actual impact of carbon fines on a drinking water biofilm has never been assessed. As early as 1980, Amirtharajah and Wetstein (1980) recognized particle presence in the effluent of GAC filters as a cause for the quality degradation of drinking water. Wilcox *et al.* (1983) showed that microbial growth was associated with GAC. The efficiency of biological activated carbon filtration, mostly used in Europe, has been gathering interest from the American drinking water utility community. The concerns voiced earlier about GAC filtration have been reinforced for BAC since bacterial growth is encouraged. From the few studies published specifically about CF, it is known that these particles can be colonized by many kinds of bacteria including coliforms (Camper *et al.*, 1986), and that bacteria attached to GAC are very resistant to disinfection (Stewart *et al.*, 1990; LeChevallier *et al.*, 1984). What is unknown, however, is whether CF may have an impact on distribution system biofilms by providing sites for additional growth or protection from disinfection. Particle size is important as it (1) has been shown to influence the susceptibility of attached bacteria to disinfection (Stringfellow *et al.*, 1993), (2) would potentially affect the number of bacteria per particle transported to the biofilm surface, and (3) may subsequently impact a higher number of surrounding bacteria once in the biofilm.

To investigate the importance of particle size, two size ranges of carbon particles (1.2–8 $\mu$m and 1.2–50 $\mu$m) were used. The size distribution analysis indicated that 75% of the particles in the suspension

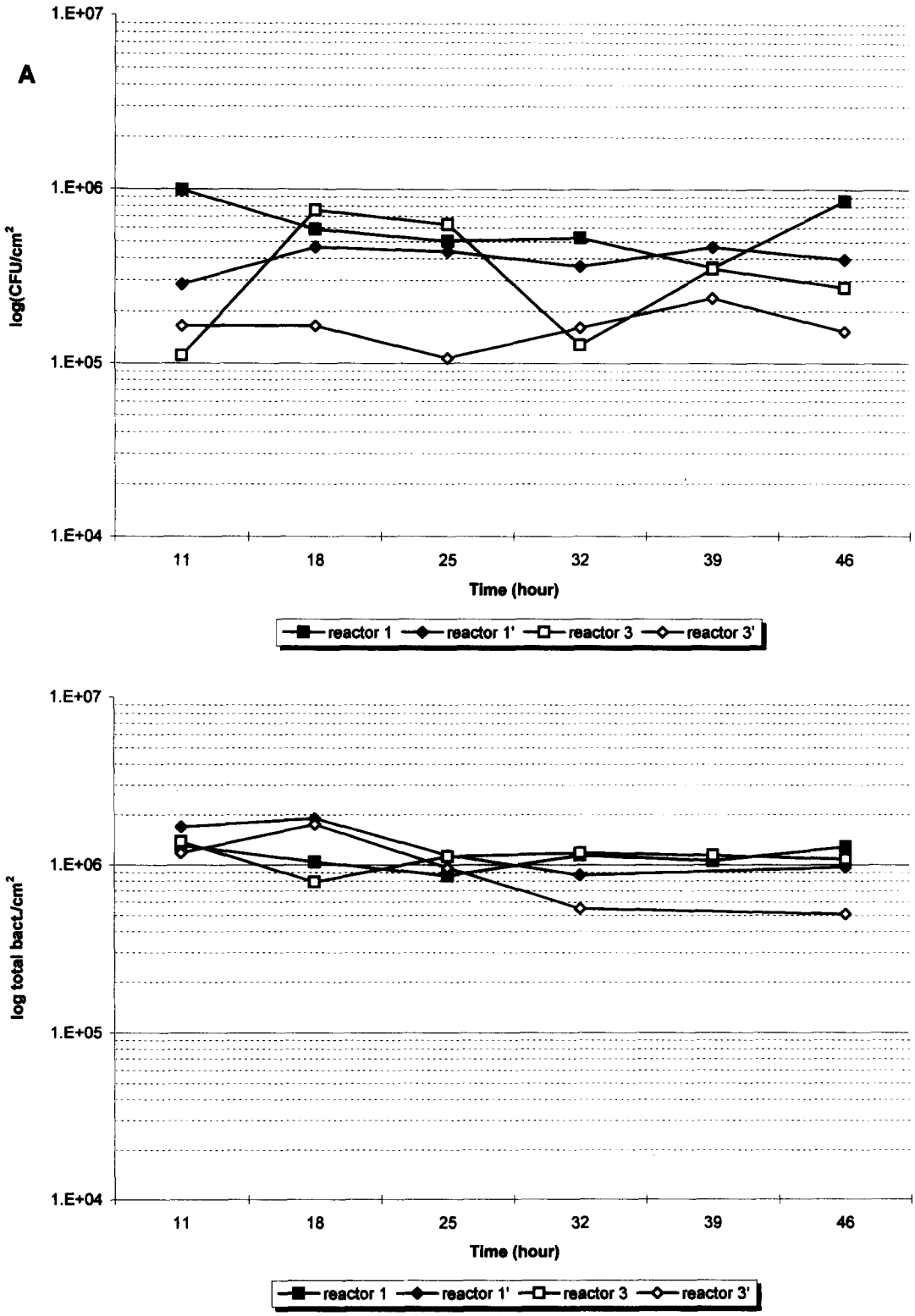



Fig. 2. Impact of the presence of CF on biofilms. Reactors 1 and 1′ received a slug dose of carbon fines ($3.5 \times 10^5$ CF/ml, average size 11.5 $\mu$m). Reactors 3 and 3′ did not receive carbon fines. (A) Culturable heterotrophs. (B) Total bacterial counts.

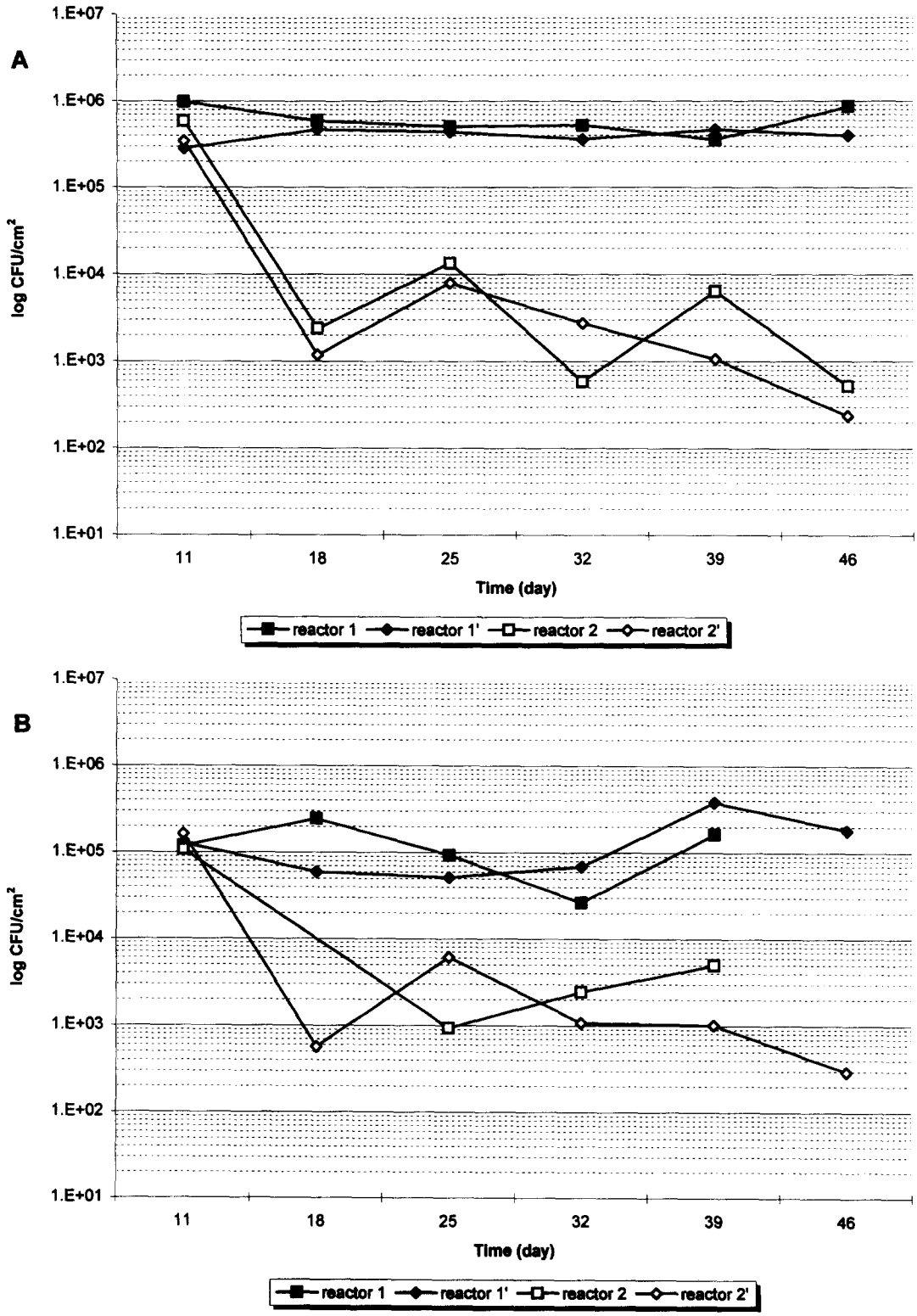



Fig. 3. Effect of disinfection on the culturable heterotrophic population of biofilms containing carbon fines. Reactors 1 and 1′ were fed nanopure water (controls). Reactors 2 and 2′ received disinfectant solutions: (A) chlorine (0.5 mg/L); (B) monochloramine (1 mg/L).

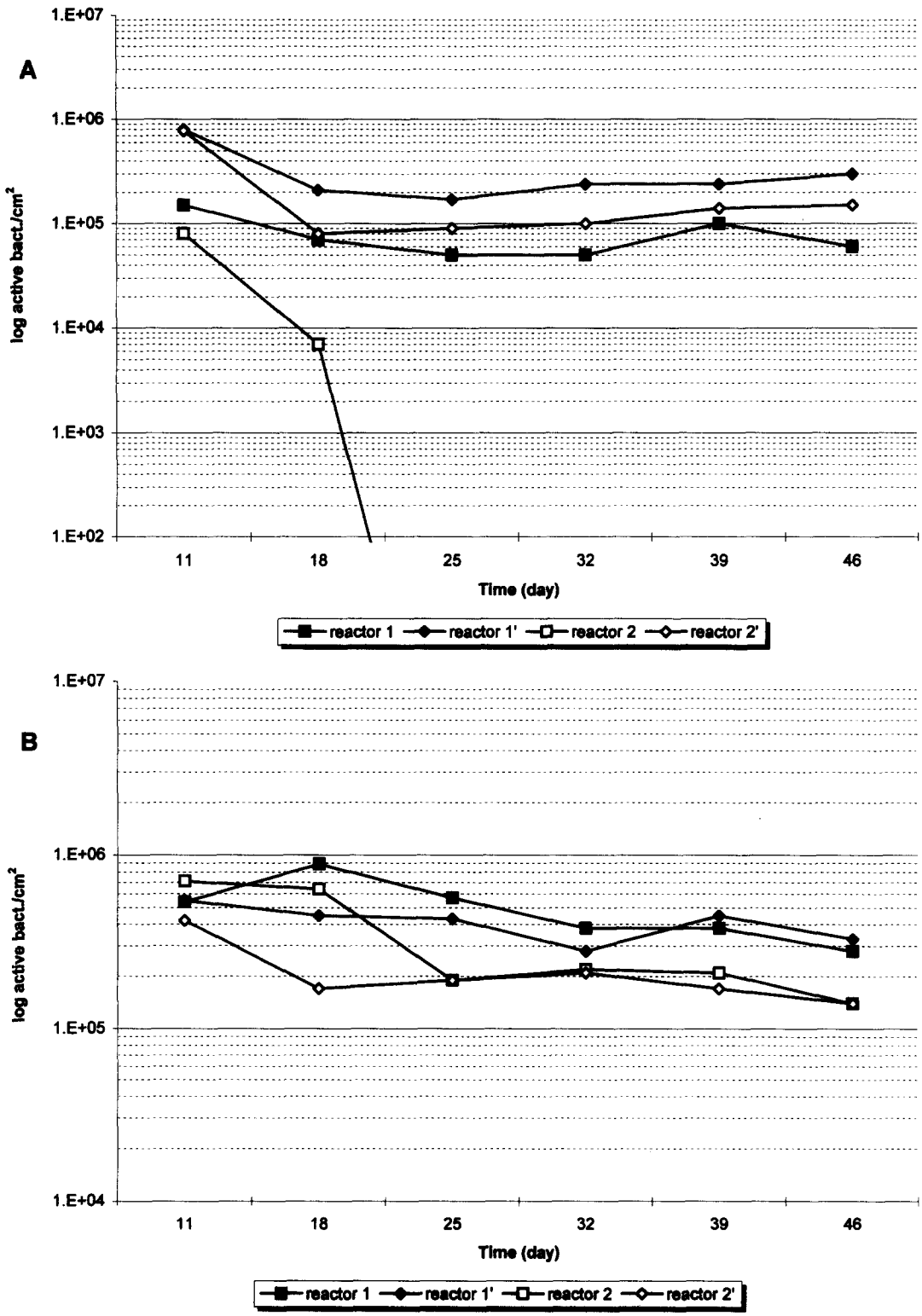



Fig. 4. Effect of disinfection on the active bacterial population of biofilms containing carbon fines. Results using CTC are shown in Fig. 4A, reactor 2. All others are results using rhodamine 123. Reactors 1 and 1′ were fed nanopure water (control). Reactors 2 and 2′ received disinfectant solutions: (A) chlorine (0.5 mg/L); (B) monochloramine (1 mg/L).

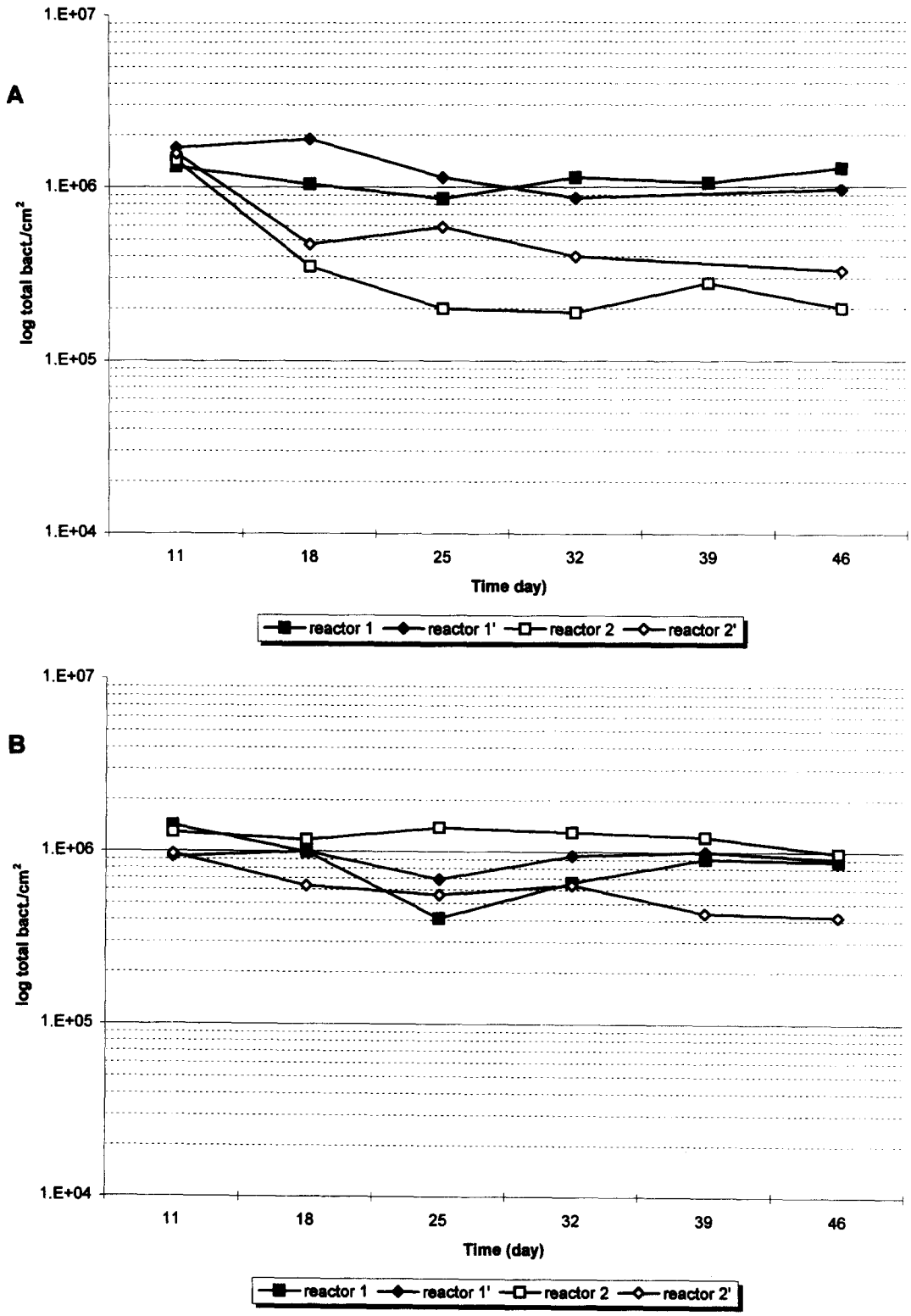



Fig. 5. Effect of disinfection on the total bacterial population of biofilms containing carbon fines as determined by epifluorescence microscopy. Reactors 1 and 1′ were fed nanopure water (control). Reactors 2 and 2′ received disinfectant solutions: (A) chlorine (0.5 mg/L); (B) monochloramine (1 mg/L).

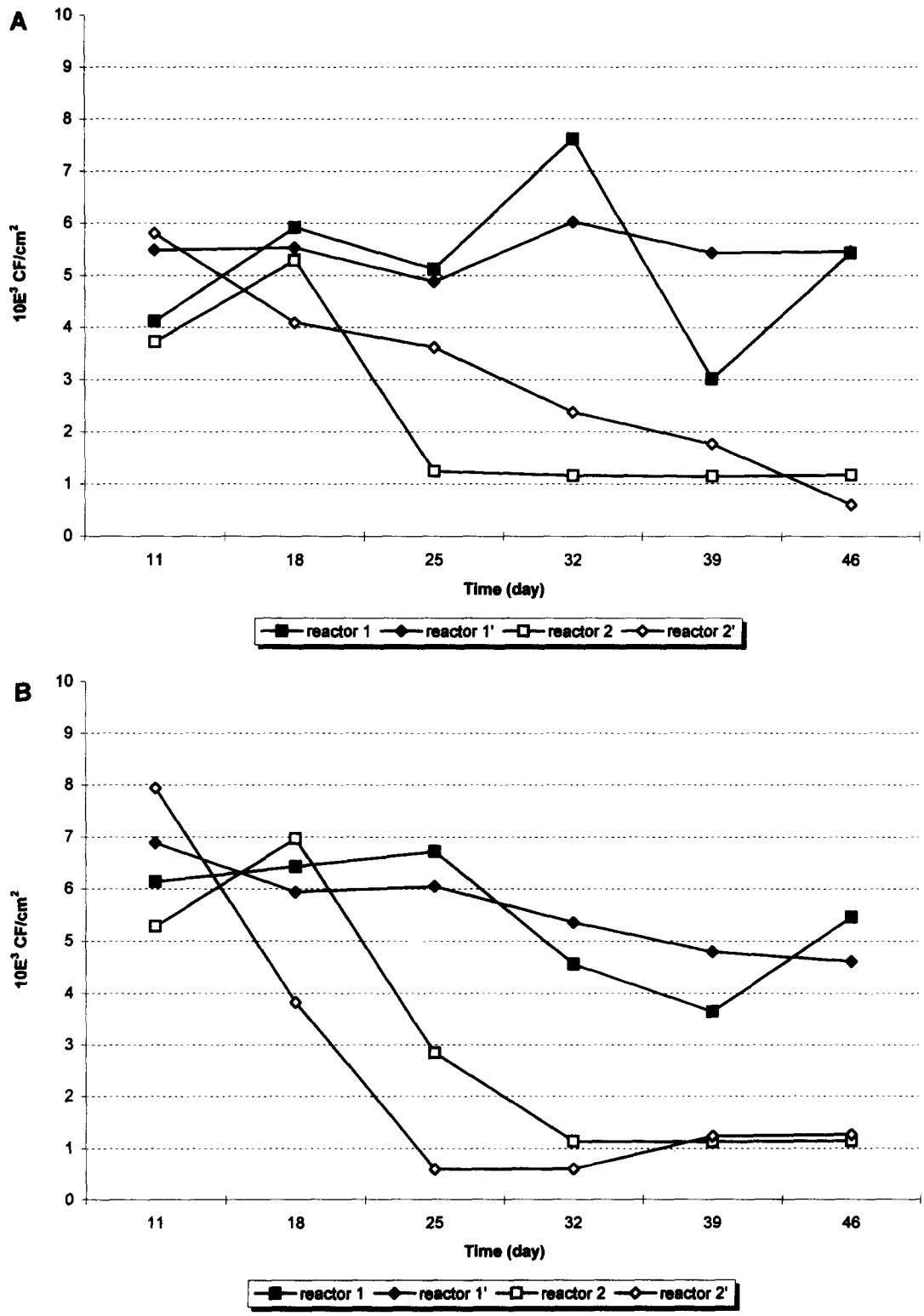



Fig. 6. Fate of large carbon fines in disinfected biofilms. Reactors 1 and 1′ were fed nanopure water (control). Reactors 2 and 2′ received disinfectant solutions: (A) chlorine (0.5 mg/L); (B) monochloramine (1 mg/L).

of large fines were larger than the average size of the small fine suspension. This size difference resulted in distinct particle behavior both in terms of attachment and persistence in the biofilm. A higher fraction of the total number of large fines added to reactors became associated with the biofilm when compared to results with the smaller size range of CF. Additionally, the larger fines persisted longer in the biofilm. As shown with the control experiment with a thick biofilm, the difference in response is not due to a lack of available binding sites for the small CF. Conversely, there was an enhancement of attachment of larger CF in the thicker biofilms. There may also be a volume-related kinetic phenomenon where the larger particles are transpored to the biofilm with a higher momentum, allowing them to be embedded more strongly, and to be less disturbed by the growth and detachment process in the biofilms.

Since granular activated carbon is a known chemisorbant, and that a GAC surface loaded with certain organic substrates can support enhanced colonization (Davies and McFeters, 1988), it was of interest to known if the addition of carbon particles would increase biofilm cell numbers under the low nutrient conditions of these experiments. Even though the quantity of retained CF was of the same order of magnitude as the culturable cell numbers, there was no noticeable overall impact on biofilm cell numbers. These results were substantiated by direct microscopic observation of the biofilms, which showed no discernable correlation between carbon particle location and microcolony formation. It therefore appears that the carbon fines did not act as a site for substrate enhancement under these conditions.

Chlorine and monochloramine are known to have different effects on biofilms (LeChevallier *et al.*, 1988a, b). Comparisons were made on particle retention and the biofilm cell behavior in response to these two common disinfectants. While chlorine seemed slightly more efficient than monochloramine in controlling the culturable bacterial population, the overall detachment monitored by total cell count and active cell count determined by rhodamine 123 was small for both agents. This observation is consistent with other work showing the great resistance of attached communities to disinfection (LeChevallier *et al.*, 1988a, b; Pedersen, 1990). However, it has also been shown that bacteria associated with large carbon particles are less susceptible to disinfection. Even though the carbon particle concentrations in these experiments were initially 100 fold greater than the culturable cell populations, there was no apparent protection of the biofilm from disinfection. The inability of the carbon particles to protect the bacteria was further confirmed by microscopy on intact stained biofilms. Although there was an elimination of individual, isolated cells from the biofilm surface after disinfection, there was no selective retention of total or active bacteria associated with carbon particles.

The inability of carbon particles to protect the biofilm from disinfection may be the result of two phenomena. First, the particle may not act as a "sink" for the disinfectant as previously thought. This is somewhat confirmed by the observation that there was no substantial difference between the residuals in reactors with carbon particles and those observed in other experiments where no fines were added. Second, the particles may be removed selectively from the biofilm during disinfection and therefore not be able to act as protective zones. As shown in the control experiments, the CF need to interact with the bacteria and/or excreted bacterial polymers to attach. When the biofilm was subjected to the disinfectants, there was a preferential loss of large carbon particles. This implies that the interaction of the fines with the biofilm is not very strong, since total biomass was not greatly reduced. The loss of 80% of the carbon particles in excess of 5 $\mu$m may partially explain why the results from the disinfection experiments were comparable to those found in other experiments with reactors operated under identical conditions receiving chlorine where no carbon particles were added (Camper *et al.*, 1994).

## CONCLUSIONS

This study presents information on the interactions of CF in a simulated drinking water biofilm, and the consequences of their presence in such a biofilm, particularly under disinfection conditions. Our findings can be summarized as follows:

(1) The attachment of CF into a biofilm is size-dependent. The very small particles (1.2–8 $\mu$m) are not retained while the larger ones (1.2–50 $\mu$m) do persist in biofilms. This is of interest since with more surface available, large CF are more likely to be colonized if released from a biological activated filter. They are, therefore, potentially more likely to impact distribution system biofilms.
(2) The fines accumulating in the biofilm do not appear to enhance the numbers of bacteria in the biofilm or act as a focal point for microcolony development.
(3) Carbon fines do not protect the biofilm against disinfection.
(4) The disinfectants induced the detachment of the CF.

These findings address some of the concerns over the release of colonized carbon particles from filters entering distribution systems. Even in these experiments with excessively large slug doses of CF, there was no discernable impact on the biofilms. Moreover, disinfection appeared to release the particles persisting in the biofilm.

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