Environ. Sci. Technol. **1998**, *32,* 3612−3617

Nanofiltration Foulants from a Treated Surface Water

THOMAS F. SPETH,*,† R. SCOTT SUMMERS,‡ AND ALISON M. GUSSES‡

U.S. Environmental Protection Agency, 26 West Martin Luther King Drive, Cincinnati, Ohio 45268, Civil and Environmental Engineering Department, Box 210071, University of Cincinnati, Cincinnati, Ohio 45221-0071

The loss of membrane flux due to fouling is one of the main impediments in the development of membrane processes for use in drinking water treatment. The objective of this work was to determine the nature of nanofiltration membrane foulants for a pilot system fed conventionally treated Ohio River water for 15 months. The foulant responsible for flux loss was shown to be a film layer 20−80 μm thick with the greatest depth in the first of three elements in series. Heterotrophic plate count, phospholipid, and pyrolysis-GC/MS analyses showed the film layer had a strong biological signature. The inorganic contribution to the dried film layer was low (less than 15%). Both acid and alkaline/detergent cleaning yielded only short-term flux recovery, although they were independent of each other. Neither acid cleaning, alkaline/detergent cleaning, nor hydraulic flushing removed the thickness of the film layer, nor changed its organic characteristics. Only alkaline/detergent cleaning inactivated a large percentage of the film-layer microbes. The short-term flux recovery without loss of biofilm thickness suggests a morphological change upon chemical cleaning. The results suggest that feedwater pretreatment to prevent cell deposition and subsequent biogrowth would be more successful than chemical cleaning the membrane after biofouling.

Introduction

Membranes can be very effective in removing a wide variety of contaminants from drinking waters. However, membrane fouling can reduce the applicability of membrane processes. Membrane fouling is defined as the accumulation of material on the surface, or in the pores, of a membrane that decreases the membrane's permeate flux. There are five broad foulant categories: sparingly soluble inorganic compounds, colloidal or particulate matter, dissolved organics, chemical reactants, and microorganisms (*1*−*4*). Often, the type of foulant is operationally defined by the type of membrane cleaning agent that is effective in recovering water flux. Inorganic foulants are typically removed with acid solutions, whereas organic and biological foulants are typically removed with alkaline/detergent solutions.

Fouling can reduce the water flux through a membrane up to 90% (*5*). Typically, however, nanofiltration membranes are cleaned whenever the permeate flux decreases to 85% of the initial permeate flux, or when the feed pressure increases to 115% of the initial feed pressure for elements operated at a constant flux (*6*). The degree of membrane fouling, which dictates the frequency of cleaning and the membrane area needed, will have a significant impact on the cost, design, and operation of full-scale facilities.

Membranes can be fouled either by adsorption or cake-layer formation. Adsorption involves materials attaching within the pores, or matrix, of the membrane. This type of fouling is generally considered irreversible although chemical cleaning can be effective in some cases (*1*). Cake-layer fouling involves the accumulation of material on the membrane surface. This type of fouling is often considered to be reversible. Cake-layer fouling is thought to be controlled by adjusting hydraulic operating parameters, such as cross-flow velocity or recovery of a system. Cross-flow velocity is the velocity of the water in the feed channel, whereas recovery is the percentage of permeate flow to the total feed flow.

Recovery and concentration polarization will dictate the concentration of dissolved species at the membrane surface. The increased concentration of dissolved species at the membrane surface increases the likelihood of precipitation of inorganic foulants, such as calcium carbonate, and the destabilization of organic contaminants. This increase in concentration is responsible for much of the foulant deposition and flux decline seen in reverse osmosis and nanofiltration membranes.

Mallevialle et al. (*7*) used pyrolysis-GC/MS and scanning electron microscopy to analyze ultrafiltration membrane foulants. Pyrolysis-GC/MS is an analytical method that can be used to characterize complex organic matter. Pyrolysis thermally cleaves nonvolatile organic molecules into volatile fragments that are then separated by gas chromatography and identified by mass spectroscopy. The resultant fragment profiles can then be associated with different types of organic compounds (*8*). The technique has been used extensively in the petrochemical industry and in the fields of biology, soil science, and geochemistry (*9*). Although pyrolysis-GC/MS can give valuable results, the technique is presently only semiquantitative.

Mallevialle et al. (*7*) analyzed the feed stream, permeate stream, and cleaning rinse of the ultrafiltration membranes studied. Four waters of varying concentrations of organic and inorganic materials were used in the study. Foulants related to concentration polarization were not a part of the study because the large pores of the ultrafiltration membranes allowed the dissolved organic and inorganic material to pass through the membranes. In all cases, clays associated with organics were found to be most responsible for ultrafiltration fouling. Carbohydrates, proteins, and polyhydroxyaromatics were also linked to membrane fouling.

The objective of this work was to determine the nature of nanofiltration membrane foulants for a pilot system fed conventionally treated Ohio River water for 15 months. The foulant layer was analyzed by pyrolysis-GC/MS to classify the types of organic materials present, confocal microscopy to determine biofilm depth, heterotrophic plate counts (HPCs) and phospholipid analyses to enumerate microorganisms, and inorganic analyses. Foulant removal and flux recovery were assessed after application of acid, alkaline/detergent, hydraulic, and physical cleaning approaches.

Experimental Section

The pilot run utilized three 4 in. × 40 in. TFCS-4921 (Fluid Systems, San Deigo, CA) elements in series with the reject stream of one element becoming the feed stream to the next.

* Corresponding author e-mail: speth.Thomas@EPAMAIL.EPA.GOV.; phone: (513) 569-7208; fax: (513) 569-7892.
† U.S. Environmental Protection Agency.
‡ University of Cincinnati.

10.1021/es9800434 CCC: $15.00
Published on Web 10/02/1998

TABLE 1. Mean Water Quality Parameters for the Membrane Feed Water

	method[a]	no. of samples	mean feed concentration[b]	mean bulk rejections (%)
		Organics		
total organic carbon, mg/L	USEPA 415.1	43	1.99 (0.41)	94.8 (3.6)
UVA at 254 nm, 1/m	Hitachi U-2000	46	4.54 (1.39)	97.9 (1.2)
		Particulates		
turbidity, NTU	S.M. 2130	64	0.23 (0.08)	72.7 (11.8)
HPCs, CFU/μL	S.M. 9215A6cC	25	33.5 (27.4)	96.9 (4.7)
		Inorganics		
calcium, mg/L	USEPA 200.7	50	37.0 (4.7)	96.6 (0.5)
chloride, mg/L	S.M. 4500-Cl^-D	44	23.4 (7.9)	87.0 (1.1)
magnesium, mg/L	USEPA 200.7	50	10.0 (2.0)	96.4 (0.5)
potassium, mg/L	USEPA 200.7	42	2.8 (0.8)	85.9 (2.5)
silicon, mg/L	USEPA 200.7	50	2.2 (0.6)	87.2 (1.5)
sodium, mg/L	USEPA 200.7	50	18.1 (7.5)	84.1 (1.6)
sulfur, mg/L	USEPA 200.7	50	26.0 (5.4)	97.5 (0.2)
conductivity, μS/cm	Oakton 35607	68	388 (46)	92.6 (1.7)

[a] S.M. = Standard Methods for the Examination of Water and Wastewater, (18th ed.). [b] Standard deviations indicated in parentheses.

The TFCS membrane is of aromatic polyamide thin-film construction with a nominal molecular weight cutoff below 200 D, and a design sodium chloride rejection of 85% (*6*). The membrane feedwater was alum coagulated, settled, and filtered (conventionally treated) Ohio River water (ORW) from the Cincinnati Water Works (Cincinnati, OH). The sand filters at the plant were operated without a disinfectant, thus no chlorine residual came into contact with the membranes. Table 1 shows the mean water quality parameters for the membrane feedwater and the mean bulk rejections for the three elements combined over the 15-month study period.

After approximately three weeks of run time, it was determined that the pilot unit required weekly acid cleaning to maintain a steady water flux in the summer months. The cleanings were conducted at low pressures in normal-flow direction at two times the cross-flow velocity with a sulfuric acid solution at a pH of 2.0 for 45 min. An alkaline/detergent cleaner consisting of sodium silicate, sodium sulfate, sodium carbonate, trisodium phosphate, and sodium dodecylbenzene sulfonate at a pH of 11.3 was found to offer no advantage over acid cleaning. During the winter months, the membranes only needed to be cleaned every two to three weeks based on the 15% flux loss criteria. However, weekly cleanings were maintained for comparison purposes.

The membrane elements were operated as a constant pressure system. The initial feed pressure entering the first element of 6.3 bar yielded a pressure of 4.8 bar at the end of the third element. However, over the 15 months of operation, the build up of foulants increased the resistance to flow through the feed spacer, and the initial feed pressure had to be raised 0.9 bar to maintain 4.8 bar at the end of the third element. This restriction of the feed channel due to foulant material build up occurred mainly in the first element. The cross-flow velocity for the entire system ranged from 4.4 to 6.2 m/min. The individual elements had mean cross-flow velocities of 6.0, 5.3, and 4.6 m/min for elements 1, 2, and 3, respectively. The recovery of the pilot system varied from 20 to 47% over the 15 months of run time because of changes in the average pressure and temperature, and the reduction of flux due to fouling.

Upon completion of the pilot run, the membrane material was unrolled and small sections were cut out and placed in a 4 in. × 6 in. flat-plate membrane vessel (*10*). The flux of each flat-sheet sample was measured at a feed pressure of 5.5 bar with conventionally treated ORW before chemical cleaning, after chemical cleaning, and after physically removing the foulant layer with a PTFE spatula. This entire cleaning process is referred to as the bench-scale autopsy flux experiments. The integrity of the membrane after physical cleaning was confirmed with conductivity and ultraviolet absorbance (UVA) measurements. The recovery was maintained between 5 and 12%. This low recovery allowed for quick determination of permeate fluxes because the concentrate's steady-state concentration was close to the feed concentration. As with the pilot data, all bench-scale data were normalized by temperature (*6*), operating pressure, and osmotic pressure (*11*) by the following equations:

$$\text{flux (at 25 °C)} = \text{flux (at } T \text{ °C)} \exp[2600[[1/(T + 273)] - 1/298]] \quad (1)$$

$$\text{specific flux} = \text{flux}/(\Delta P - \Delta\pi) \quad (2)$$

$$\Delta\pi = 0.00069[(\text{TDS}_f + \text{TDS}_c)/2 - \text{TDS}_p] \quad (3)$$

where T is the membrane operating temperature in Celsius, ΔP is the net pressure difference in bars, $\Delta\pi$ is the osmotic pressure gradient in bars, and TDS_f, TDS_c, and TDS_p are the feed, concentrate, and permeate total-dissolved-solid concentrations in milligrams per liter, respectively.

All bench-scale autopsy flux cleanings were conducted for 45 min at a cross-flow velocity twice that used during pilot operation, which corresponded to pilot-scale cleaning conditions. Some bench-scale autopsy flux cleanings used treated ORW to determine the amount of flux that was recovered due to hydraulic shear. Other cleanings utilized sulfuric acid at a pH of 2.0, or the commercial alkaline/detergent cleaner designed for biogrowth removal. Some of these chemical cleanings utilized both acid and alkaline/detergent cleaners in succession.

The foulant layer before and after cleaning was quantified by weight, depth, inorganic species, HPCs, phospholipids, and pyrolysis-GC/MS biopolymer groupings. The inorganic species analyzed include aluminum, barium, calcium, chromium, copper, iron, lead, magnesium, manganese, nickel, phosphorus, potassium, sodium, sulfur, tin, and zinc. Digestions were conducted according to procedure SW846 method 3050 (*12*), and the analytics were conducted according to USEPA Method 200.7. Hydrofluoric acid was used to obtain the silicon content. Unfortunately, due to the small size of the bench-scale unit, insufficient material was collected for inorganic analyses after acid, alkaline/detergent, and hydraulic cleaning at the bench-scale.

The HPCs were determined using R2A agar with a spread-plate technique incubated at 25 °C for 7 days. The foulant layer was physically removed from a known area of the membrane with a PTFE spatula. It was then fluidized with

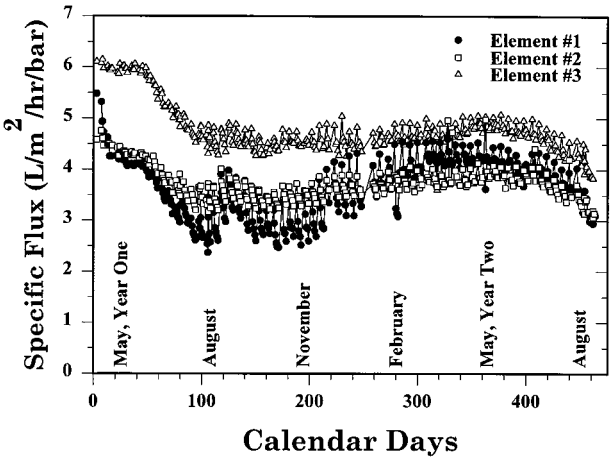

FIGURE 1. Specific flux for individual pilot membranes.

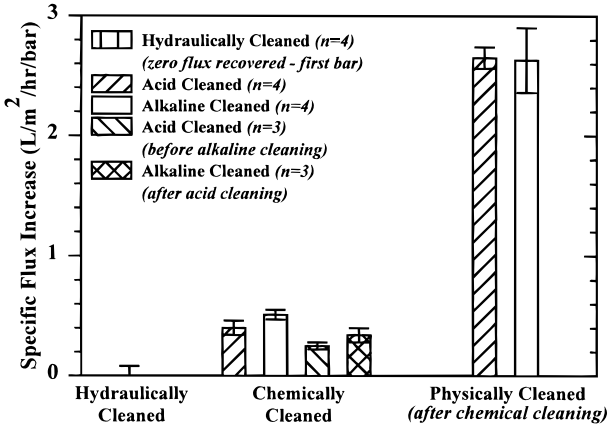

FIGURE 2. Increase in specific fluxes for various bench-scale cleaning scenarios for the first element in series.

sterilized water, and an aliquot was placed on the agar. Each sample was enumerated in triplicate and averaged. Samples for foulant dry-weight analyses were prepared in a manner similar to that for the HPC tests except the fluidized foulant was placed into tarred weighing pans that were previously dried to a constant weight. The sample was then dried in a forced-air oven at 105 °C to a constant weight. Blanks of the sterilized fluidizing water were also measured and subtracted from the sample weights.

The phospholipid analyses were based on the method developed by Findlay et al. (*13*). This involved extracting the lipids from a known area of fouled membrane with a chloroform/methanol/water solution. The lipid-containing chloroform phase was separated from the aqueous phase and evaporated. Five percent potassium persulfate was added before digestion at 100 °C for at least 8 h. After cooling, the ampule was opened and ammonium molybdate and malachite green indicator were added. The sample was then analyzed with a spectrophotometer at 610 nm. This absorbance was then related to phospholipid quantities and live cell counts as discussed by Findlay et al. (*13*).

The confocal microscopy analyses consisted of staining the membrane with Baclight Bacterial Viability Kit (Molecular Probes, Inc, Eugene, OR). The sample was then analyzed with a Zeiss Axiophot fluorescent microscope (Germany) and a Biorad MRC-1000 confocal imaging system (Hercules, CA). A krypton/argon laser was used with an excitation filter of 568 nm. The depths of the viable and nonviable microbes were visually determined by scanning the foulant layer from the top to the bottom and back.

The pyrolysis-GC/MS method used herein was developed by Bruchet et al. (*8*). This method categorizes natural organic matter fractions in terms of biopolymers: polysaccharides, proteins, *N*-acetyl amino sugars, and polyhydroxy aromatics. They are calculated relative to a known standard mixture of dextran (MW 70 000), bovine serum albumin, cellulose acetate, and humic acid. A simple empirical algorithm was developed for computing relative peak areas among the various classes of biopolymers (*8*). The pyrolysis-GC/MS procedure entailed ramping the pyrolysis sample from 250 to 625 °C at 20 °C per millisecond. The final temperature was held constant for 20 s. Splitless injection to a 60-meter Supelco WAX-10 GC column was used in the procedure. The GC temperature was ramped from 35 to 260 °C at a rate of 2 °C/min. A Hewlett-Packard 5892 GC was used with a HP 5971 mass-selective detector.

Results and Discussion

Field Performance. Figure 1 shows the specific flux over the length of the pilot-scale study for the three elements in series. The specific fluxes show four distinct trends: seasonal variation, element variation, nonrecoverable flux loss, and recoverable flux loss. The greatest overall specific flux decline occurred in the summer and fall seasons, and the third element in series maintained the highest specific flux. After more than one year of operation, the nonrecoverable flux was at least 25 to 35% of the initial specific flux for all three membranes. The flux lost each week after cleaning ranged from 5 to 18%. Weekly acid cleanings with sulfuric acid recovered between 50 and 163% of the specific flux lost during the previous week.

The flux loss did not correlate to the temperature or the magnitude of permeate flux as others have seen (*14*−*16*). This indicates that the seasonal flux loss was related to chemical, particulate, or biological changes in the water as opposed to seasonal temperature changes. The greater fouling in the early elements suggest particulate or biological fouling. This is consistent with the greater module pressure drops experienced in the first element. Chemical destabilization would be expected to be greatest in the final element in series where the concentrations were the highest.

The flux loss also did not correlate to cross-flow velocity. The first element had the most fouling of the three elements, but had the highest cross-flow velocity. Likewise, the most fouling occurred early in the membrane run where the cross-flow velocity was the highest due to the lack of feed channel blockage by microbes/particulates.

The membrane exhibited high rejections of most constituents measured. Table 1 shows the mean feedwater concentrations and the mean bulk rejections over the course of the 15-month study. There were no discernible trends in the rejection with fouling over the study period. The rejections were consistent with the manufacturer's expected rejection of sodium chloride (85%) and hardness (95%). The turbidity rejection was artificially low due to low concentrations and high detection limits.

Autopsy Flow Studies. The bench-scale autopsy baseline fluxes, before cleaning, were expected to be similar to the final pilot-scale membrane fluxes. However, there was a consistent 30% increase in flux as compared to the final pilot-scale membrane flux. This was due to foulant layer disturbances that occurred while unrolling the fouled membrane from the element and placing the fouled membrane sheet into the bench-scale system. Similar rejections of both conductivity and UVA at the bench and pilot scale were used to confirm that the integrity of the membrane was not compromised during unrolling.

The specific flux increases for the membrane in the first element in series, after bench-scale autopsy cleanings, are shown in Figure 2. The amount of flux recovered by physical cleaning represents most of the total flux decline seen by

TABLE 2. Dried Weights of Material Removed from Membrane Elements

	weight of material removed,[a,b] μg/cm^2		
	first element	second element	third element
fouled	161 (22, 2)	191 (58, 3)	98 (18, 3)
cleaning agent			
hydraulic	695 (575, 2)	NT	NT
acid	227 (27, 2)	144 (44, 2)	76 (44, 2)
alkaline/detergent	166 (40, 2)	141 (15, 2)	98 (6, 2)
acid followed by alkaline/detergent	225 (117, 2)	NT	NT

[a] Standard deviations and number of samples indicated in parentheses. [b] NT = Not taken.

element one in the field (Figure 1). This indicates that the viscous film layer that was removed by physical cleaning was responsible for most of the flux decline. Viscous foulant material has been related to biological fouling (*17*). Hydraulically cleaning the membrane at twice the cross-flow velocity with conventionally treated ORW did not increase the flux. Acid cleaning at a pH of 2.0 and alkaline/detergent cleaning recovered only 20% of what physical cleaning recovered. The flux recovered with alkaline/detergent cleaning after acid cleaning was similar to the flux recovered with alkaline/detergent cleaning alone, suggesting that these cleaners removed different foulants because the fluxes recovered appear to be independent of each other. The amount of flux recovered by acid cleaning on the bench-scale was in the range of the short-term flux recoveries produced by weekly acid cleanings in the field.

Film Layer Mass and Inorganic Composition. The dried weights of material physically removed from the membranes, before and after cleaning, are shown in Table 2. The standard deviations are large for some of the samples, and the film-layer weight for the fouled first element seems suspicious because it was lower than that after chemical cleaning. These limitations withstanding, the data show that the third element had less material deposited than the first two elements. This suggests that the foulant was organic in nature because the greatest amount of inorganic deposition was expected in the third element in series where the inorganic constituents were most concentrated. The weights of the dried deposits also suggest that none of the chemical or hydraulic cleaning approaches were effective in removing the film layer. Therefore, any flux recovered by cleaning was due to a small amount of foulant layer that was removed, a morphological change, and/or a chemical change within the foulant layer.

Table 3 shows the quantification of inorganic species in the material physically removed from the membrane. The major constituents were iron, aluminum, and calcium. The percentages of the total dried weight identified as inorganic species were 13.1, 9.7, and 10.0% for elements one, two, and three, respectively. These low values, and the similarity of the values for the first and third elements, suggest that inorganic precipitation was not a significant mechanism for flux reduction, if it is assumed that the weight percent of the inorganic material relates to flux reduction. Unfortunately, the validity of this assumption cannot be addressed here. Silicates are not listed in Table 3 because of inconsistent recoveries. However, the results obtained indicated that silicates were not a significant foulant as determined by weight percent.

The low amounts of inorganics do not rule out their role in organic and colloidal fouling. Hong and Elimelech (*14*) found that divalent cations interact with humic carboxyl functional groups to reduce the charge of, and electrostatic repulsions between, humic macromolecules. This reduction in electrostatic repulsion led to increased NOM deposition on the membrane surface (*14*). Zhu and Elimelech (*15*) found that the membrane fouling rate increased with increasing ionic strength due to a reduction in the repulsive electrostatic forces between the colloids and the membrane surface.

TABLE 3. Inorganic and Confocal-Microscopy Results of Membrane Deposits before Cleaning

	element 1	element 2	element 3
inorganic, mg/g			
aluminum	23.7	18.0	12.4
barium	1.63	0.940	7.67
calcium	21.4	10.6	33.0
chromium	0.100	0.100	0.037
copper	0.236	0.224	0.198
iron	65.5	51.1	30.5
potassium	2.57	2.12	1.50
magnesium	2.33	1.67	1.42
manganese	1.26	1.60	1.57
sodium	0.537	0.348	0.353
nickel	0.071	0.048	0.035
phosphorus	4.67	4.74	4.06
lead	0.165	0.123	0.206
sulfur	5.53	4.97	7.05
tin	0.042	0.050	0.023
zinc	1.10	0.290	0.230
total summation	130.8	96.9	100.2
$\%_{wt}$ identified above	13.1	9.7	10.0
confocal microscopy			
viable depth, μm	76	35	22
nonviable depth, μm	80	38	30

Biofilm Analyses. The viable and nonviable biofilm layer thicknesses determined from confocal microscope analyses of the membranes with intact foulant layers are also shown in Table 3. The greatest thickness, 76−80 μm, occurred in the first element, and the thinnest, 22−30 μm, in the third element. This thickness gradient is similar to that of the dried-weight analyses. The nonviable microbe thickness was slightly greater than the viable thickness; however, the viable and nonviable microbes were interspersed with each other with no apparent layering.

The feed spacer material was 762 μm thick. Since the spacer material separates two sheets of membrane material in a spiral-wound element, the thickness of the biofilm was 21, 10, and 8% of the distance between membrane sheets for the first, second, and third element, respectively. This biofilm occupation of such a substantial portion of the flow channel height not only manifested itself in the membrane's reduced flux, but also in an increase in the module pressure drop across the feed channel stream over the length of the pilot run. The module pressure drops across elements one, two, and three were 0.76, 0.28, and 0.07 bar, respectively. Therefore, at the end of 15 months of operation, the greatest module pressure drop occurred in the first element where the biofilm thickness and the foulant mass were the greatest.

The confocal microscopy data only gives an indication of the biofilm thickness. Method limitations include destabilization from unrolling the membrane, inadequate penetration of stain through the biofilm, compression of the biofilm

TABLE 4. HPC Autopsy-Flow Results before and after Cleaning

	HPC counts per membrane surface area,[b] CFU/cm^2 $\times$ 10^{-6}		
	first element	second element	third element
fouled	4.5 (2.0, 3)	1.1 (0.9, 3)	2.2 (1.8, 3)
cleaning agent			
hydraulic	3.7 (1.6, 4)	NT	NT
acid	4.4 (2.6, 4)	1.2 (0.5, 4)	2.0 (2.1, 4)
alkaline/detergent	0.5 (0.2, 4)	0.1 (0.0, 4)	0.1 (0.0, 4)
acid followed by alkaline/detergent	0.2 (0.1, 3)	NT	NT

[a] Standard deviations and number of samples indicated in parentheses. [b] NT = Not taken.

TABLE 5. Phospholipids Autopsy-Flow Results before and after Cleaning

	phospholipid enumerated cells per membrane surface area,[a,b] cells/cm^2 $\times$ 10^{-6}		
	first element	second element	third element
fouled	20 (7, 3)	38 (4, 2)	36 (7, 2)
cleaning agent			
hydraulic	28 (15, 5)	NT	NT
acid	18 (4, 4)	55 (29, 4)	22(1, 4)
alkaline/detergent	13 (2, 4)	36 (16, 4)	15 (3, 4)
acid followed by alkaline/detergent	20 (4, 3)	NT	NT
physical	4 (3, 4)	19 (13, 5)	4 (1, 4)

[a] Standard deviations and number of samples indicated in parentheses. [b] NT = Not taken.

TABLE 6. Pyrolysis-GC/MS Results of the Feedwater and Deposits before and after Cleaning

	relative percentages of biopolymers[a]			
	polysaccharides	proteins	polyhydroxy aromatics	amino sugars
ORW feed water	26 (13, 6)	38 (7, 6)	28 (12, 6)	9 (2, 6)
fouled[b]				
first membrane	42	22	19	17
second membrane	52	16	9	23
third membrane	48	16	12	24
cleaning agent[b]				
hydraulic				
first membrane	48	21	13	18
acid				
first membrane	50	19	12	20
second membrane	51	16	9	24
third membrane	55	14	7	25
alkaline/detergent				
first membrane	42	20	21	17
second membrane	44	15	19	22
third membrane	57	12	11	20
acid followed by alkaline/detergent				
first membrane	47	18	20	15

[a] Standard deviations and number of samples indicated in parentheses. [b] $n = 1$.

from the microscope lense, and operator judgment in determining the biofilm boundaries. However, assuming a minimum film density of 1.0 g/cm^3, the confocal-microscopy and dried-weight results both suggest the percentage of dried solids is very low. The first, second, and third elements had maximum percent solids in the wet film layer of 2.0, 5.0, and 3.3%, respectively. These low percent solids in the wet film layer lend additional support to the notion that the film was largely biological in nature (i.e., mostly water).

The HPC values of the physically removed material per square centimeter of membrane are shown in Table 4. Due to the inexactness in extracting these samples, the standard deviations were very high. The counts of the first element were about twice that of the others. Overall, the results showed a high degree of biological fouling which was not surprising based on the high HPC counts in the feedwater (Table 1). After acid cleaning and after hydraulic cleaning with ORW, the HPC levels remained high. Therefore, the weekly acid cleaning over 15 months appears to have created a biological community structure that acclimated to cleaning. Other data from our lab show that a membrane that was cleaned infrequently with acid had a significant HPC drop upon acid cleaning. After alkaline/detergent cleaning, the HPCs were an order of magnitude lower than the fouled membrane results. This was also the case for acid cleaning followed by alkaline/detergent cleaning, presumably due to the alkaline/detergent cleaning. Therefore, the alkaline/detergent cleaner was able to inactivate, and/or remove, the bacterial cells.

The phospholipid enumerated cell counts, shown in Table 5, were an order of magnitude higher than the HPC results. This was presumably due to the method's ability to enumerate more microbes than what would plate out on the HPC agar (*13*). Assuming the microbes were 1 $\times$ 1 $\times$ 2 μm in size and

were arranged in a monolayer, a level of 50×10^6 cells/cm^2 would constitute complete surface coverage. The phospholipid results were in that range, although confocal microscopy demonstrated that the microbes were present throughout the film depth (up to 80 μm). Some of the phospholipid results are inconsistent with those of the HPC analyses. For example, the HPC results showed more biomass on the first element relative to the other elements while the phospholipid results did not.

Consistent with the HPC results, the phospholipid results show that hydraulic flushing and acid cleaning did not significantly remove the microbes. However, the phospholipid results did not show the order of magnitude drop in cells after alkaline/detergent cleaning and after acid cleaning followed by alkaline/detergent cleaning that the HPC results showed. These results suggest that the alkaline/detergent cleaner inactivated the microbes as measured by HPC, but did not remove them as measured by phospholipids. This would require that the alkaline/detergent cleaner preserved the phospholipids from the recently inactivated cells during cleaning and before analysis (1 h). Findlay et al. (*13*) report that the phospholipid method does not quantitate inactive organisms or extracellular material because the phospholipids released from inactivated microbes are rapidly degraded in the environment. Apparently, the alkaline/detergent cleaning solution prevented the natural degradation of the phospholipids. As discussed earlier, the alkaline/detergent cleaning did not impact the film layer as measured by dried weight. This is consistent with the phospholipid results that indicated the microbes were not being removed with cleaning. It is interesting to note that the number of viable microbes (Table 4) did not correlate to the amount of flux recovered by the different cleaners (Figure 2). This indicates that the viability of the microbes was not important with regard to flux reduction.

Table 5 shows that physically cleaning the fouled membranes reduced the biomass to levels of a new unused membrane ($2-5$ cells/cm$^2 \times 10^{-6}$). The results of the second membrane in series were an aberration, but these data were skewed by one high reading as reflected in the large standard deviation. These data confirm that microbes were present in the cake-layer foulants that were responsible for flux decline.

Pyrolysis-GC/MS Analyses. Pyrolysis-GC/MS was used to classify the organic material that had deposited on the membranes into four biopolymer groups (*8*). Table 6 shows the distribution of organic material for the conventionally treated ORW and the deposits that were physically removed from the membranes. The feedwater results, the average of six analyses sampled while the membrane was in operation, showed that the protein and amino sugar compositions were very consistent with time. The film before cleaning had a higher percent composition of polysaccharide and amino sugar groups as compared to the feedwater. These two groups have historically been associated with biogrowth with the polysaccharides arising from extracellular materials (*17*–*20*), and the aminosugars arising from the cell walls of bacterial or fungal biomasses (*9*, *21*). Therefore, it appears that the characteristics of the fouling matter were dominated by biological growth (cell mass itself and the extracellular materials), and not from the NOM from the water. These results were consistent with the high microbial levels found with the HPC and phospholipid analyses. The elevated extracellular portion suggests that the microbes had developed a colony relationship with a high degree of extracellular materials. This is consistent with the confocal microscopy results that showed a biofilm with significant depth.

As shown in Table 6, the chemical cleaners used in this study did not significantly change the organic-matter characteristics of the film as determined by pyrolysis-GC/MS. This coincides with the previous results that showed that the various cleaners did not significantly change the film weight and depth, or the phospholipid biogrowth enumeration. Thus, a morphological change of the foulant with cleaning, as opposed to foulant removal, was likely to have occurred. This suggests that feedwater pretreatment to prevent cell deposition and subsequent biogrowth would be more successful than chemical cleaning the membrane after biofouling.

Acknowledgments

The authors appreciate the efforts of Donald Mitchell (USEPA) for his help in building and monitoring of the pilot membrane units, Vanessa Hatcher (University of Cincinnati) for her help in monitoring the bench-scale membrane vessel, Keith Kelty (USEPA) for conducting the inorganic analyses of the membrane foulant, and Alan Lindquist (USEPA) for conducting the confocal microscope analyses. The authors would also like to thank the staff of the Cincinnati Water Works for their help in this work, especially Jeffrey Swertfeger, Melissa Fredrickson, and Jack DeMarco. Alison Gusses was supported by USEPA-OGWDW cooperative agreement (CX-820695-01). Mention of trade names or commercial products in this paper does not constitute endorsement or recommendation for use by the U.S. Government.

Literature Cited

(1) Belfort, G. *Desalination* **1977**, *21*, 285.
(2) Matthiasson, E.; Sivik, B. *Desalination* **1980**, *35*, 59.
(3) Potts, D. E.; Ahlert, R. C.; Wang, S. S. *Desalination* **1981**, *36*, 235.
(4) Barger, M.; Carnahan, R. P. *Desalination* **1991**, *83*, 3.
(5) van den Berg, G. B.; Smolders, C. A. *Filtr. Sep.* **1988**, *2*, 115.
(6) Fluid Systems, Inc. *TFCS Nanofiltration Membrane Product Manual*, 1994.
(7) Mallevialle, J.; Anselme, C.; Marsigny, O. In *Aquatic Humic Substances: Influence on Fate and Treatment of Pollutant;* Suffet, I. H., MacCarthy P., Eds.; American Chemical Society: Washington, DC, 1989; pp 749–767.
(8) Bruchet, A.; Rousseau, C.; Mallevialle, J. *J. Am. Water Works Assoc.* **1990**, *82* (9), 66.
(9) Irwin, W. J. *Analytical Pyrolysis;* Marcel Dekker: New York, 1982.
(10) Allgeier, S. C.; Summers, R. S. *J. Am. Water Works Assoc.* **1995**, *87* (3), 87.
(11) Talyor, J. S.; Jacobs, E. P. In *Water Treatment Membrane Processes*; Mallevialle, J., Odendall, P. E., Wiesner, M. R., Eds.; McGraw-Hill: New York, 1996; Chapter 9.
(12) *Test Methods for Evaluating Solid Waste*, 3rd ed.; U.S. Environmental Protection Agency: Washington, DC, 1986; Vol. 1A (SW-846), Chapter 3050A.
(13) Findlay, R. H.; King, G. M.; Watling, L. *Appl. Environ. Microbiol.* **1989**, *55* (11), 2888.
(14) Hong, S.; Elimelech, M. *J. Membr. Sci.* **1997**, *132*, 159.
(15) Zhu, X.; Elimelech, M. *Environ. Sci. Technol.* **1997**, *31*, 3654.
(16) Chellam, S.; Jacangelo, J. G.; Bonacquisti, T. P. *Environ. Sci. Technol.* **1998**, *32*, 75.
(17) Ridgway, H. R.; Flemming, H. C. In *Water Treatment Membrane Processes;* Mallevialle, J., Odendall, P. E., Wiesner, M. R., Eds.; McGraw-Hill: New York, 1996; Chapter 6.
(18) Christensen, B. E.; Characklis, W. G. In *Biofilms*; Characklis, W. G., Marshall, K. C., Eds.; John Wiley & Sons: New York, 1990; pp 93–130.
(19) Geesey, G. G.; Stupy, M. W.; Bremer, P. J. *Int. Biodeterior. Biodegrad.* **1992**, *30*, 135.
(20) Neu, T. R.; Marshall, K. C. *J. Biomater. Appl.* **1990**, *5*, 107.
(21) Gadel, F.; Bruchet, A. *Water Res.* **1987**, *21* (10), 1195.

Received for review January 20, 1998. Revised manuscript received July 6, 1998. Accepted August 31, 1998.

ES9800434