

MICROBIAL FILM DEVELOPMENT AND ASSOCIATED ENERGY LOSSES

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Abstract

Microbial fouling in power plant condensers increases heat transfer and fluid frictional resistance resulting in energy losses. Biofouling control is generally by chlorine addition creating potential toxicity problems in receiving waters. A better understanding of biofouling film development and destruction (i.e., stoichiometry and kinetics) is necessary to maintain effluent water quality while minimizing biofouling effects.

This paper reviews research progress in the following areas:

1. Development of various sensitive biofilm detection methods for monitoring the extent of biofouling.
2. Determination of effects of certain variables on the kinetics and stoichiometry of biofilm accumulation.
3. Correlation of biofilm development to increases in both heat transfer and fluid frictional resistance.
4. Determination of the effects of chlorine applications on established biofilms.

Introduction

The term fouling refers to the formation of inorganic and/or organic deposits on surfaces. In cooling systems, these deposits form on condenser tube walls increasing fluid frictional resistance, accelerating corrosion and impairing heat transfer. Four types of fouling, alone or in combinations, may occur:

1. crystalline fouling caused by precipitation of $CaCO_3$, $CaSO_4$ or silicates
2. corrosion fouling resulting from formation of insulating layers of metal oxides on the tubes
3. fouling due to adherence of particulate matter on tube surfaces
4. biological fouling resulting from attachment and growth of microbial organisms

This investigation was restricted to the study of biological fouling.

The Problem

The most common method for controlling the fouling biofilm development and maintaining condenser performance is periodic chlorination. However, concern over residual toxicity from hypochlorous acid or its reaction products has resulted in federal regulations which limit the allowable concentrations of free available chlorine in cooling water discharges. At the present time, there is no sound basis for assessing the impact of the regulations.

This investigation stems from the apparent need for a more basic understanding of fouling biofilm development and fouling biofilm destruction.

Project objectives included the following:

1. Develop a better understanding of fouling biofilm development, with particular emphasis on the effects of fluid flow rate, bulk water temperature, wall surface temperature and limiting nutrient concentration.
2. Determine the effectiveness of fouling biofilm destruction by chemical oxidants, primarily chlorine.
3. Develop a practical, reliable, sufficiently sensitive device for monitoring biofouling and for effectively operating and controlling biofouling destruction processes at operating power plants.

Laboratory experiments and a limited number of field tests were conducted with two reactor configurations:

1. a tubular reactor
2. an annular reactor consisting of a stationary outer cylinder and a rotating inner cylinder.

The tubular reactor geometry and its turbulent flow regime are identical to those existing in cooling water condensers. The annular reactor was tested as a biofouling monitor because it is very sensitive to fouling and is easy to operate and maintain. The annular reactor has the potential of being used in a sidestream from the cooling water supply to continuously monitor biofouling for control of the addition of oxidant. Biofouling in the experimental reactors was measured by observing changes in the following parameters:

1. biofilm thickness
2. attached biomass
3. fluid frictional resistance
4. heat transfer resistance

Processes in Fouling Biofilm Development

Microbial fouling is the combined result of phys-

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ical transport and biological growth rate processes as shown in Fig. 1. The following processes contribute to overall biofouling accumulation:

Biofouling Processes

1. Organic Adsorption
2. Transport of Microbial Particles to the Surface
3. Microorganism Attachment to the Surface
4. Growth of Attached Microorganisms
5. Reentrainment of Biofilm by Fluid Shear Stress

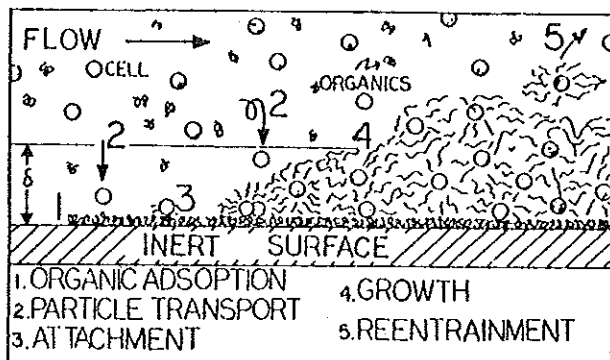


Fig. 1 Biofouling Development Processes

Organic adsorption. Figure 1 illustrates an initially "clean" surface exposed to a turbulent flow of fluid containing dispersed microorganisms, nutrients, and organic macromolecules. Adsorption of an organic monolayer occurs within minutes of exposure. Investigations have shown that, once exposed to natural waters with low organic concentrations, materials of originally diverse surface properties (i.e., wettability, surface tension, electrophoretic mobility) are rapidly conditioned by adsorbing organics.^{2,3,4,5,11,12,18,19}

Transport of microbial particles to the surface. Figure 1 indicates the physical transport of bacterial particles from the bulk fluid to the organically-coated surface. Within a turbulent flow regime, particles suspended within the fluid are transported to the solid surface by only two mechanisms: molecular diffusion and turbulent eddy transport. Theory indicates that the flux of particles to the surface increases with increasing fluid velocity and particle concentration in the bulk fluid. However, particle flux is also strongly dependent on the physical properties of the particles (e.g., size, shape, density).⁶

Microorganism attachment to the surface. Bacteria do not attach to solids very tenaciously unless left in contact with the water source for several hours. Then, some cells are attached so strongly that they are not dislodged by washing or staining. This suggests that bacteria grow on, and/or attach themselves to solid surfaces rather than merely being passively stuck.

Research confirms the existence of a two-stage attachment process: reversible adhesion followed by an irreversible attachment. Reversible adhesion refers to an initially weak attachment of a bacterium

to a surface. Organisms still exhibit Brownian motion and are readily removed by mild rinsing. Conversely, irreversible attachment is a permanent cohabitation with the surface, usually aided by production of cellular exopolymers. Photomicrographs show that biofilms do not consist of successive layers of microorganisms, but rather microorganisms entrapped in a gelatinous matrix.^{10,15} Irreversibly attached bacteria can only be removed by severe methods.

Most attachment studies were concerned with the biological sequences of early fouling and were conducted at very low shear rates. Kinetics determined from these studies may be mass transfer limited and not necessarily applicable to condenser biofouling.

Growth of attached microorganisms. Attached and dispersed microorganisms assimilate nutrients, produce extracellular polymers, maintain biosynthesis, and replicate. Biofilm "growth" is considered the combined effect of cellular reproduction and exopolysaccharide production, as shown in Fig. 1.

Biofilm growth can be described by a wide variety of rate expressions whose rate constants are functions of pH, temperature, limiting nutrient concentration, nutrient type, terminal electron acceptor, and organism concentration.

Postulated rate expressions for nutrient depletion by a fixed biofilm are numerous, but all agree that nutrient depletion rates are first order in biofilm mass and, generally, diffusion-limited in the biofilm.^{1,14,16,17,24}

Reentrainment of biofilm by fluid shear. At any point in the development of a biofilm, portions of biofilm peel away from the inert surface and are reentrained in the fluid flow (Fig. 1). Reentrainment is a continuous process of biofilm removal and is highly dependent on hydrodynamic conditions. Sloughing, on the other hand, appears to be a random, massive removal of biofilm attributed to oxygen/nutrient depletion deep within biofilms. Sloughing is more frequently witnessed with thicker, dense films. More work is needed to quantify either effect.

In summary, biofouling is the net result of all these rate processes occurring simultaneously. Naturally, at specific times in the overall development certain processes may contribute more significantly than others.

Experimental

Protocol

The experimental methods used in this laboratory study were chosen to simulate the fundamental processes of biofilm development and destruction occurring in a small portion of a power plant condenser.

Nutrients, glucose and, in some cases, a synthetic growth media were added to provide the necessary mineral, energy and carbon requirements for microbial growth. The feed water to the various experimental reactors was tap water which had been treated to remove residual suspended solids and chlorine.

Apparatus

Two reactor geometries were used in this research

to study biofilm development and destruction:

1. The circular tube was used because it is the prevalent geometry in power plant condensers.
2. The sensitivity of the rotating annular reactor was tested because it could provide a more practical, reliable means for monitoring biofouling in the field since it requires little maintenance or support equipment. This paper will not discuss results from the annular reactor.

Tubular fouling reactors. The tubular fouling reactors are chemostats (continuous stirred tank reactors) with internal recycle as indicated in Fig. 2. Advantages of this configuration for laboratory experimentation include the following:

1. At high recycle rates employed ($F_R \gg F$), the reactor contents are completely mixed and no concentration gradients exist. This simplifies mathematical descriptions and sampling. It also provides a relatively uniform biofilm in the recycle section while allowing simple control of pH and temperature. From a practical standpoint, this system minimized the consumption of water and microbial nutrient medium.
2. A short mean residence time can be maintained which minimizes biomass activity in the bulk fluid and restricts microbial activity to the reactor surfaces.
3. Fluid shear stress at the wall in the recycle loop is independent of mean residence time in the reactor system.

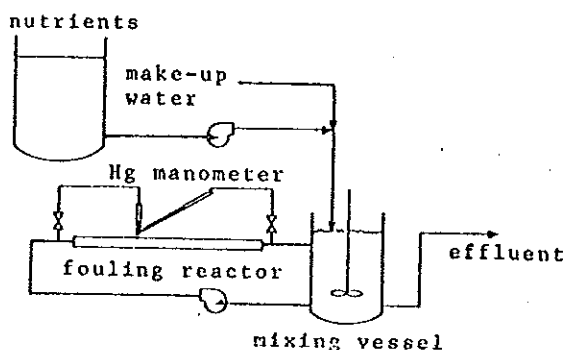


Fig. 2 Biofouling Reactor System

Results from three tubular reactor systems are discussed in this paper. Each system is similar to the design depicted in Figure 2 but differ from each other according to their specific intent as follows:

Tubular Fouling Reactor 1 (TFR1) was used for determining the effect of fluid shear rate at the wall, bulk water temperature, and limiting substrate concentration on biofilm development as determined by attached biomass, biofilm thickness and fluid frictional resistance. Tubular Fouling Reactor 3 (TFR3) was an improved version of TFR1.

Tubular Fouling Reactor 4 (TFR4). Biofilm thickness, fluid frictional resistance, and heat transfer resistance were measured by TFR4 experiments. Heat transfer resistance was measured in a Test Heat Exchanger (THE) which was an integral part of the recycle reactor.

Tubular Fouling Reactor 5 (TFR5) was used for studying biofouling development during the initial fouling phase. It consists of a continuous culture reactor (i.e., chemostat C.S.T.R.) with internal recycle loops. In this way one can maintain a constant bulk microbial culture at a desired activity while independently varying fluid flow rate.

Further details concerning methods and experimental protocol can be found elsewhere.^{7,13,21,25}

Analytical Methods

Biofouling was measured by observing changes in the following parameters:

1. biofilm thickness
2. attached biomass
3. fluid frictional resistance
4. heat transfer resistance
5. biofilm constituent chemical analysis

Details of the analytical methods are presented elsewhere.^{7,13,21,25}

Results and Discussion

Biofilm Development

The process of biofilm development on a surface exposed to turbulent flow is adequately described by a sigmoidal-shaped curve. The progression of biofilm development can be conveniently divided into three phases:

1. Induction Phase - the initial development period during which the biofilm accumulation increases but fluid frictional resistance remains constant.
2. Logarithmic Growth Phase - the period of time during which the accumulation increases logarithmically (may include a portion of the induction period).
3. Plateau Phase - the period during which deceleration of the accumulation rate is observed and pseudo-steady state in biofilm accumulation occurs.

The three phases of biofilm development are indicated in Fig. 3.

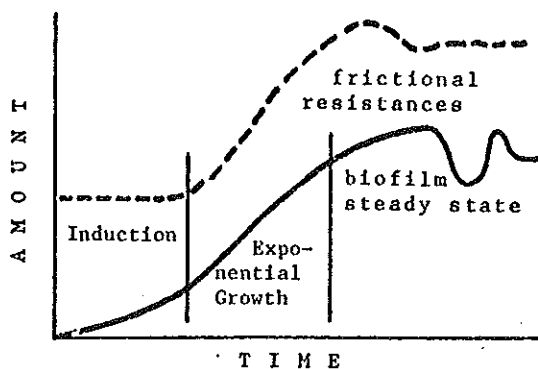


Fig. 3 Phases of Biofilm Development

Induction Phase. Primary biofilm formation occurs during that period of time from "clean" surface conditions to the onset of frictional resistance increase.

Experiments were conducted to ascertain the rate of primary biofilm formation as a function of dispersed biomass concentration (X), Reynolds number (Re), and dispersed organism growth rate (μ). The relationship can be described by Eq. 1:

$$R_i = dX_A/dt = k \cdot f(x, Re, \mu) \quad (1)$$

where X_A = attached biofilm amount/area; R_i = rate of initial biofilm formation.

All primary biofilm experiments were conducted in TFR5. Details of the experimental protocol for this research are provided by Bryers.⁷

Results of the primary biofilm formation experiments can be summarized as follows:

1. Biofilm develops faster at higher Reynolds number (i.e., increased velocity) at constant dispersed biomass concentration and dispersed growth rate.
2. Increasing dispersed biomass concentration, at constant Re and dispersed growth rate, increases the rate of initial biofouling
3. An increased dispersed organism growth rate increases the rate of primary biofilm formation, at constant Re and dispersed biomass concentration.

Growth and plateau phase. Biofilm development progression for a typical experiment is illustrated in Fig. 3. The logistic or sigmoidal curve for biofilm development can be described analytically by the following function:

$$Th = \frac{Th_0 e^{kt}}{1 - (Th_0/Th_{MAX})(1 - e^{kt})} \quad (2)$$

where Th = biofilm thickness (L); Th_0 = Th at $t=0$ (L); Th_{MAX} = maximum value to Th attained (L); k = rate constant (t^{-1}). The rate of change of Th can be described by the following:

$$\frac{d(Th)}{dt} = k(Th) - \frac{k(Th)^2}{Th_{MAX}} \quad (3)$$

The rate constant, k , describes the logarithmic rate of development which occurs at $Th \ll Th_{MAX}$. The second term accounts for a process opposing development. Regardless of mechanism, $k(Th)$ represents the logarithmic development rate or fouling rate characteristic of the growth phase while Th_{MAX} characterizes the plateau phase.

The logarithmic fouling rate (based on either biofilm thickness or biofilm mass) varies with nutrient loading rate as indicated in Fig. 4. The effect of wall shear stress is also indicated. As wall shear stress increases, the second term in Eq. 3 becomes more important in controlling biofilm development rate. This suggests that Th_{MAX} decreases with increasing wall shear stress. Figure 5 indicates that, in fact, Th_{MAX} decreased with increasing wall shear stress, especially at high nutrient loading rates.

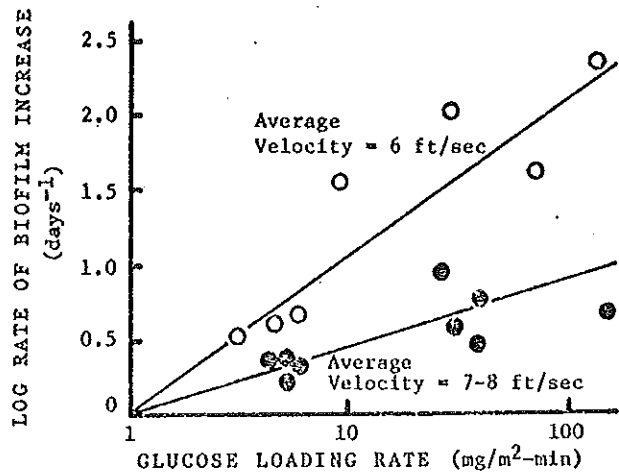


Fig. 4 Glucose loading and rate of biofilm Increase

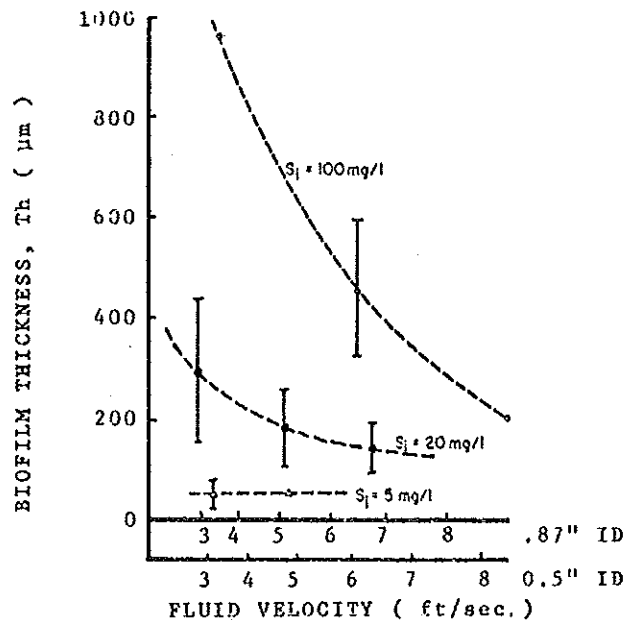


Fig. 5 Velocity effect on maximum biofilm Th Biofilm Detriments

Effects on frictional resistance. Increase in fluid frictional resistance due to biofilm accumulation during experiments in which flow rate is maintained constant causes an increase in pressure drop and power requirements for pumping as shown in Fig. 6 for a typical experiment.

Conversely, if pressure drop is held constant, flow capacity is reduced. Fig. 7 shows a typical experimental curve where flow capacity was reduced to 42% of the original capacity in a 100-hour laboratory experiment.

Frictional resistance can be represented by a dimensionless friction factor defined as follows:

$$f = 2.0 \frac{d}{L} \frac{\Delta P}{\rho_w \bar{v}^2} \quad (4)$$

where f = friction factor; d = tube diameter (L); ρ_w = fluid density (M/L^3); \bar{v} = average fluid velocity; (L/t); ΔP = pressure drop along length L ($M/L-t^2$).

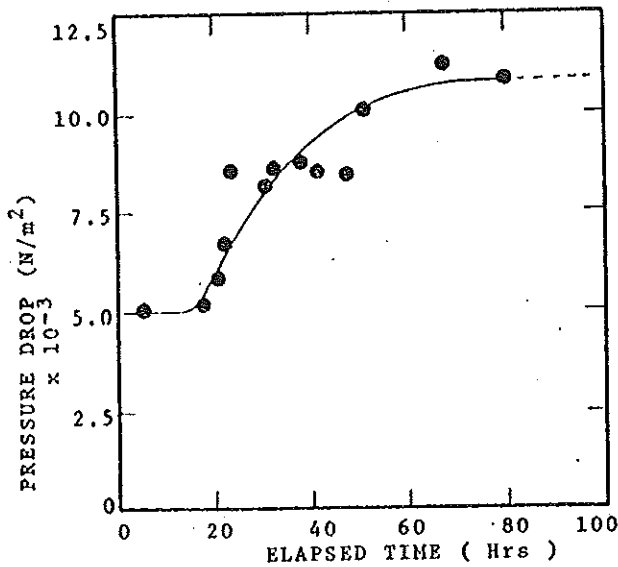


Fig. 6 Change in ΔP with time; constant flow

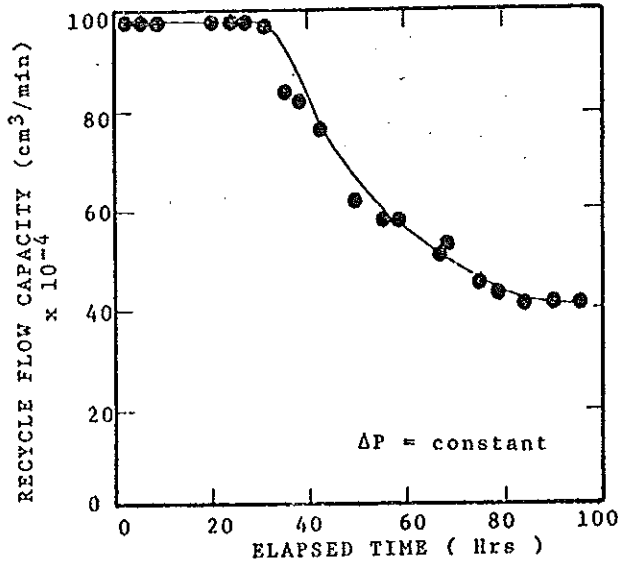


Fig. 7 Change in flow capacity with time

The change in friction factor with time for a typical experiment is shown in Fig. 8. The measured biofilm thickness for the same experiment is also indicated.

The friction factor is related to the Reynolds number and the equivalent sand roughness k_s through the empirical Colebrook-White equation:

$$k_s = \frac{d}{2} 10^{(0.87 - 0.50f^{-0.5})} - \frac{18.70}{Re f^{0.5}} \quad (5)$$

where d = tube diameter (L); $Re = vd/\nu$ = Reynolds number; \bar{v} = mean fluid velocity (L/t); ν = kinematic viscosity (L^2/t). This expression can be used to compute an equivalent sand roughness for the biofilm from a measurement of the flow rate and pressure drop. In all TFR experiments except TFR5, k_s increases with time. Figure 9 indicates the dependence of k_s on biofilm thickness for the range of shear stress investigated (6.5 - 7.9 N/m^2).

The Blasius-Stanton or Moody diagram can be used to compare frictional resistance due to biofilm development with frictional resistance of rigid rough

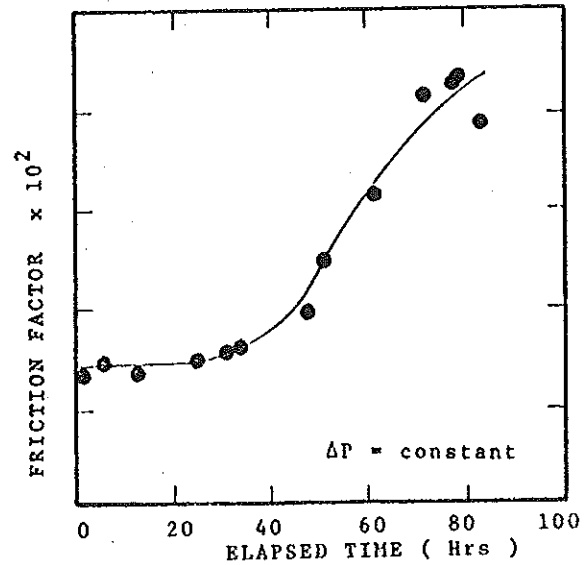


Fig. 8 Change in friction factor with time

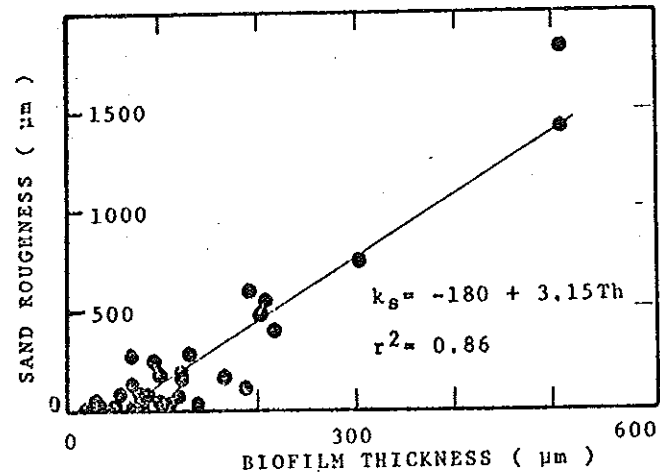


Fig. 9 Change in equivalent sand roughness with biofilm thickness for all constant ΔP experiments

surfaces.²⁰ The Blasius-Stanton diagram is a plot of friction factor vs. Reynolds number for a series of pipes with different equivalent sand roughness; the friction factor in a pipe with a rigid rough surface depends on both the relative roughness and the Reynolds number.

The relationship between friction factor and Reynolds number for the fouled TFR3 system is presented in Fig. 10. This figure shows the dependency of friction factor on Reynolds number is similar to that for a tube with a rigid rough surface between the range of Reynolds numbers investigated (5,000 to 48,000).

The relationship between biofilm thickness and friction factor for all experiments at a wall shear stress from 6.5 - 7.9 N/m^2 is shown in Fig. 11. Friction factor is dependent on film thickness after a critical thickness (Th_{crit}) approximately equal to the thickness of the viscous sublayer is attained. Thickness of the viscous sublayer is defined by Eq. 6:

$$\delta = \frac{10d}{Re} \left(\frac{f}{2} \right)^{-0.5} \quad (6)$$

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References

1. Atkinson, B. and M.E. Abdel Rahman Ali. 1976. Wetted area, slime thickness, and liquid phase mass transfer in packed bed biological film reactors. Trans. Instn. Chem. Engr., 54, pp. 239-250.
2. Baier, R.E., E.G. Shafin, and W.A. Zisman. 1968. Adhesion: mechanisms that assist or impede it. Science, 162.2, p. 1360
3. Baier, R.E. 1972. Influence of the initial surface condition of materials on bioadhesion. Proc. 3rd Int'l. Cong. on Mar. Corrosion and Biofouling, Gaithersburg, Md.
4. Baier, R.E. 1973. Applied chemistry at protein interfaces. Adv. Chem. Series, No. 145, p. 1.
5. Baier, R.E. and V.A. Depalma. 1977. Microfouling of metallic and coated metallic flow surfaces in model heat exchanger cells. Calspan Corp. Report, P.O. Box 235, Buffalo, N.Y.
6. Beal, S.K. 1970. Deposition of particles in turbulent flow on channel or pipe walls. Nucl. Sci. and Engr., 40, pp. 1-11.
7. Bryers, J.D. and W.G. Characklis. 1979. Measurement of primary biofilm formation. Proc. Biofouling of Power Condenser Symposium - EPRI, Atlanta, Ga.
8. Characklis, W.G. and T. Dydek. 1975. Influence of carbon to nitrogen ratio on the chlorination of microbial aggregates. Water Res., 10, pp. 515-522.
9. Colburn, A.P. 1933. Correlation of momentum and energy transfer. Trans. A.I.Ch.E., 29, pp. 174-210.
10. Costerton, J.W., G.G. Geesey and K.J. Cheng. 1979. How bacteria stick. Scientific American, Jan., p. 86.
11. Dexter, S.C., J.D. Sullivan, J. Williams, and S.W. Watson. 1975. Influence of substrate (surface) wettability on the attachment of marine bacteria. Appl. Microb., 30.2, pp. 298-308.
12. Dexter, S.C. 1976. Influence of surface wettability on the formation of bacterial slime films on solid surfaces immersed in natural sea water. Proc. 4th Int'l. Cong. on Mar. Corr. and Fouling, Antibes, France.
13. EPRI. 1979. Biofilm development and destruction. Final Report. EPRI No. RP902-1. Proj. Director: Roger M. Jordan. EPRI - Palo Alto, Ca. Res. Dir.: W.G. Characklis.
14. Harremoes, P. 1977. Half order reactions in biofilm kinetics. Vatten, 2, pp. 122-143.
15. Jones, H.C., I.L. Roth, and W.M. Sanders. 1969. Electron microscopic study of a slime layer. J. Bacter., 99.1, pp. 316-325.
16. Kornegay, B.H. 1967. Characteristics and kinetics of biological fixed film reactors. Ph.D. dissertation, Clemson University, Clemson, S.C.
17. LaMotta, E.J. 1974. Evaluation of diffusional resistances in substrate utilization by biological films. Ph.D. dissertation, Univ. of No. Carolina at Chapel Hill.
18. Loeb G. and R. Neihoff. 1972. Molecular fouling of surfaces in seawater. Proc. 3rd Int'l. Cong. on Mar. Corrosion and Biofouling, Gaithersburg, Md.
19. Loeb, G. and R. Neihoff. 1973. Marine conditioning films. Adv. Chem. Ser., 145, p. 319.
20. Moody, L.F. 1944. Friction factor dependency on Reynolds number and pipe roughness. Trans. ASME, 66, p. 671.
21. Nimmons, M.J. 1979. Biofilm effects on heat transfer resistances within a turbulent flow system. M.S. Thesis, Rice University, Houston, Tx.
22. Norrman, G. 1976. Control of biofilms in circular tubes with chlorine. M.S. Thesis, Rice University, Houston, Tx.
23. Picoglou, B., N. Zilver, and W.G. Characklis. 1979. Influence of biofilms on hydraulic deterioration in circular tubes. Submitted for publication, J. Hydraulics Div., ASCE.
24. Trulear, M. 1979. Dynamics of biofilm processes. Proc. 34th Annual Industrial Wastewater Trtmt. Conf., Purdue University, Purdue, In.
25. Zilver, N. 1979. Biofilm development and associated energy losses in water conduits. M.S. Thesis, Rice University, Houston, Tx.

DISCUSSION

Question: ...OTEC...what is the relative (inaudible) between warm water and cold-water concentration.

W. Characklis: I don't know what the concentrations are of warm water and cold water. This work wasn't done for OTEC.

J. Edwards, inaudible question:

W. Characklis: The organic content is relatively low, 5 to 10% by weight.

Question: It's not done in sea water though?

W. Characklis: No, it is not.

Question: I'm a little confused between the results of the two papers where, as I understood, Brenda Little's samples ended up without a slime. And I gather that you do end up with slime, mostly water. Is there a disparity, or am I confused by the whole situation?

W. Characklis: We ended up with a slime; Brenda will have to tell you what she had. Under normal circumstances, we have maintained flow and have found a film that is predominantly water. Several of the micrographs that Brenda showed had quite a bit of polymer material on them, but it does not look the same after it has been dried; it looks very different under those conditions.

Question: Your curves show no lag phase and the

R_f data show a lag phase, particularly in relation to the filling-in of the film with polymers rather than the laying down of micro-organisms in a thick mat. There is a change in the chemistry.

W. Characklis: R_f is, of course, a measure of heat transfer resistance, and there are data in my paper regarding the heat transfer resistance that we also measured with film. There is a lag in heat transfer resistance even though the biofilm thickness is increasing. There are a couple of possible reasons for this: in many of our experiments there was a decrease in heat-transfer resistance for a short period. And we hypothesize that it's due to the nature of these films. The film comprises filaments that grow quickly through the thermal boundary layer so that there is some increase in convective heat-transfer resistance when, in fact, the conductive heat-transfer resistance has gone up, and they counterbalance. And our sensitivity was not enough to do anything but give us a lag period. Now, in addition, metallurgy and chemistry are involved in your systems, which we did not have, because we used glass tubes to eliminate the corrosion processes from the biofouling processes. We did use aluminum alloy for heat-transfer experiments, but only for those experiments. I would also expect a lag period for your OTEC experiments because of the food availability. Probably the number and concentration of particles are much less than what we had in our system. But we have data in a report that is being prepared now that are compared with some of the data from OTEC and from HTRI in seawater, and the data compare very well.

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