95-047

Pergamon

Wat. Sci. Tech. Vol. 32, No. 8, pp. 19–26, 1995.

0273–1223/95 $9·50 + 0·00

PII:S0273-1223(96)00003-0

FLOW INDUCED VIBRATIONS, DRAG FORCE, AND PRESSURE DROP IN CONDUITS COVERED WITH BIOFILM

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ABSTRACT

Biofilm was grown in closed conduit reactors under turbulent flow conditions. Structural development of the biofilm suggests that individual microcolonies behave like blunt bodies shedding vortices. The microcolonies assumed elongated forms, termed "streamers", possibly because of an exerted pressure drag force. The streamers when entrained in the water flow vibrated rapidly dissipating kinetic energy from the bulk liquid. The energy was transferred through the biofilm causing the underlying microcolonies to oscillate. The measured pressure drop was partially attributed to the loss of energy due to these flow induced vibrations and oscillations.

KEYWORDS

Biofilm; drag; flow induced vibrations; hydrodynamics; pressure drag; pressure drop.

INTRODUCTION

Little is known about the nature of water flow near biofilms. It has been shown by many researchers that biofilm accumulation affects hydrodynamics and that, in turn, hydrodynamics affects biofilm accumulation. During the initial events of biofilm formation, hydrodynamics regulates the transport of microorganisms from the solution to the surface. Bouwer (1987) points out that the increased surface roughness due to biofilm formation can influence the particle transport rate and biofilm attachment rate by: (1) increasing convective mass transport near the surface, (2) providing shelter from shearing forces, and (3) increasing surface area for attachment. Once biofilm has accumulated, the modified near surface hydrodynamics controls the transport of substrates and metabolites to, from, and within the biofilm. Siegrist and Gujer (1985) postulate that biofilm roughness can increase eddy diffusion and the external mass transfer rate into the biofilm. Analytical description of the flow near biofilm-covered surfaces is difficult because of the viscoelastic nature of biofilms. Theoretical treatment is practically nonexistent and, therefore, most of the knowledge about such flow is empirical. However, even empirical data are sparse because of difficulties monitoring the near surface environment. The importance of biofilm accumulation to industrial, medical, and environmental systems motivates research into the nature of modifications of the hydrodynamics near the biofilm surface. Although the complexity of the process frequently prevents us from arriving at exact solutions, some parameters have to be approximated so we can design systems in which biofilm accumulation is expected or required. Examples of such industrially important parameters are flow friction resistance, heat transfer resistance, and mass transport resistance; the first being the subject of this paper. We present, herein, the results of our experimental and conceptual work contributing to the understanding of

chanisms of biofilm formation and the subsequent consequences for hydrodynamics in conduits colonized with biofilms.

Accumulation of biofilm in industrial installations increases the liquid frictional resistance leading to decreased flow capacity and increased pumping costs. This measurable effect of biofouling stimulated early researchers to study this field (Characklis, 1973). An influential work, shaping our ideas of hydrodynamics near biofilms was published by Picologlou *et al.* (1980). The authors operated a closed conduit biofilm reactor and measured the pressure drop as a function of biofilm development. They consistently monitored a sudden and significant increase in frictional resistance after the biofilm thickness reached a certain critical value, approximately equal to the thickness of the viscous sublayer evaluated for a smooth surface (without the biofilm). They postulated that this effect was caused when the biofilm elements protruded through the hydrodynamic boundary layer. Such an explanation clearly parallels the classical study by Nikuradse (1933) who studied the effects of rigid roughness elements in pipes. The analogy was familiar to the engineering community and was, therefore, readily accepted. Consequently, the hydrodynamics near surfaces covered with biofilms were described with the assumption that the biofilm formed rigid, rough surfaces. Despite this seductive simplicity the conceptual model offered by Picologlou *et al.* is based on two weak assumptions that were never verified: (1) that the biofilm surface roughness elements behave the same way as they were rigid, and (2) that the hydrodynamic boundary layer remains at the same position after the biofilm is formed. The first assumption is disputable. The second seems unlikely. Perhaps these inherent inconsistencies contribute to the fact that despite the clarity of this concept the relation between the biofilm surface roughness and the pressure drop was never quantified in general terms, as it had been for rigid surface elements.

Recently revised ideas of biofilm structure and the hydrodynamics in biofilm systems may shed new light on the relation between biofilm accumulation and frictional resistance. Studies by Lawrence *et al.*(1991) on the ucture of biofilms using Scanning Confocal Laser Microscopy (CSLM) followed by the studies of flow near microbially colonized surfaces using Nuclear Magnetic Resonance Imaging (NMRI) (Lewandowski *et al.*, 1992, 1993, 1994) and CSLM (De Beer *et al.*, 1994a,b; Stoodley *et al.*, 1994) delivered detailed information on the structure of biofilms and the nature of the water flow in biofilm systems. The conceptual image of biofilms became much more complex than the uniform layer with imbedded microorganisms that dominated the early studies. Microorganisms in biofilms are aggregated in cell clusters or microcolonies separated by interstitial voids. The new conceptual model assumes an inherent biofilm heterogeneity and constitutes the foundation for studying biofilm structure and the consequences of this structure to the physical and chemical microenvironments.

These revised ideas of biofilm structure and the demonstration of intrabiofilm liquid flow justifies reevaluating the relations between biofilm accumulation, friction resistance, and pressure drop. Our previous experiments were conducted in a small closed conduit reactor using the Nuclear Magnetic Resonance Imaging to evaluate the effect of biofilm accumulation on near surface hydrodynamics (Lewandowski and Altobelli, 1994) and Lewandowski *et al.* (1995). The entry length required for development of viscous flow was compared for reactors with and without the biofilm operated at the same flow velocity. It was found that biofilm accumulation decreased the entry length required for fully developed flow implying that biofilm formation was actually smoothing the walls and, consequently, decreasing the friction factor. Although the results were obtained under laminar flow conditions, and such flow is unusual in industrial settings, they showed that the relations between the surface roughness and biofilm formation can be quite complex, and that the declaration that biofilm accumulation always increases surface roughness may not be true.

To further explore the relations between biofilm accumulation and pressure drop we designed a series of experiments employing turbulent flow. For practical reasons a closed conduit reactor was selected and the ssure drop was used to monitor the effects of biofilm accumulation. The biofilm was accumulated at different flow velocities and the pressure drop across the reactor was monitored. Biofilm development was also routinely monitored microscopically through a specially designed observation port.

MATERIALS AND METHODS

The flow cell was a closed channel (1 cm wide, 1 cm deep, and 45 cm long) with an observation window placed at 31 cm from the entrance. The flow cell was placed in a recycle loop with a mixing chamber which had nutrient and dilution water influent streams delivered by peristaltic pumps. The flow cell could be placed on the stage of an inverted microscope (Olympus IMT-2) attached to a Bio-Rad MRC600 confocal scanning laser microscope (CSLM) without interrupting the flow conditions. The mixing chamber also had aeration and overflow effluent lines. The nutrients were mixed with the dilution water in a ratio of 1:100 (0.75 ml/min : 74.25 ml/min) for a final concentration of: glucose 40 ppm, potassium phosphate monobasic (KH_2PO_4) 70 ppm, potassium phosphate dibasic (K_2HPO_4) 30 ppm, ammonium sulfate ($(NH_4)_2SO_4$) 10 ppm, and magnesium sulfate ($MgSO_4.7H_2O$) 1 ppm. The volume of the system was 1500 ml, an influent flow rate (Q_I) of 75 ml/min was chosen to give a resulting residence time ($\theta = V/Q_I$) of 20 min so that suspended cells would be washed out of the reactor and biofilm growth favored.

The recycle flow rate (Q_R) was controlled using a vane head pump situated in a bypass loop giving flows in the approximate range of 50–275 cm^3/s (with corresponding average flow velocities ($u_{(ave)}$) of 0.5–2.75 m/s). Q_R was measured by volumetric displacement with an in-line water meter.

A differential pressure transducer (Foxboro Co., Model 843DP-H1V1NK, Foxboro MA) was used to monitor the pressure drop across the flow cell. The distance between the ports was 67 cm. The bypass valves had to be adjusted on a daily basis to maintain a constant Q_R as the reactor system became fouled. Biofilm was allowed to accumulate at 3 different Q_R; 57, 95, and 172 cm^3/s (with corresponding Reynolds (Re) numbers of 6800, 11,000, and 20,050). The biofilm was allowed to accumulate for between 2 and 3 weeks.

The flow cell, the mixing chamber and recycle lines were sterilized by exposure to a 1% bleach solution for 24 hours. The bleach was removed by rinsing with about 70 volumes of sparged filter sterilized tap water, then 3 volumes of sterile sodium thiosulfate (100 ppm), and finally about 70 volumes of sparged filter sterilized tap water again. The dilution water was tap water sterilized with in line capsule filters (1.0 μm prefilter and 0.1 μm filter). The nutrient feed and associated tubing was sterilized by autoclaving at 121°C for an appropriate exposure time.

The reactor was inoculated with stock cultures (1ml) of *Pseudomonas aeruginosa* $7.7x10^9$(CFU/ml), *Pseudomonas fluorescens* ($4.8x10^{10}$ CFU/ml), and *Klebsiella pneumoniae* ($7.2x10^{10}$ CFU/ml) and was initially run as a batch culture for 12 hours to ensure attachment before switching to continuous culture.

The biofilm was imaged daily in the same place using transmitted CSLM at 50, 100, and 200 magnification. The image location was found by lining up a mark made on the coverslip with a permanent marker with an outline of the same mark which was drawn on the monitor. The focus was then readjusted to view the biofilm. The pressure drop was also recorded daily.

In addition to the routine monitoring, the movement of the biofilm streamers was recorded in a flow cell colonized with a 21 day old biofilm grown at an $u_{(ave)}$ of 95 cm/s. Because the streamers moved very rapidly in the flow stream two techniques were used for visualization: (1) the flow rate was slowed down by opening the bypass valves completely so that the vibration of the streamers was reduced to a point that unblurred images could be taken; and, (2) neutral density fluorescent latex spheres (Molecular Probes, Eugene Oregon, density 20°C = 1055 kgm^{-3}, ex 580 nm / em 605 nm, diameter = 0.282 μm, $1.7x10^{12}$ spheres /ml) were used to mark the biofilm. When viewed in fluorescent mode with the CSLM (ex = 568 nm) the spheres stuck to the biofilm streamers appeared as bright dots against the dark background. By superimposing between 50 and 70 images on top of each other the complete range of motion of the spheres, and therefore, the streamers and underlying biofilm clusters could be seen.

The same biofilm was also used to measure the effects of biofilm accumulation on pressure drop at different recycle flow rates. The experiment was then repeated in a clean flow cell.

RESULTS

The results of pressure drop measurements are presented in Fig. 1. Three curves reflect the pressure drop changes for three different flow velocities, 0.57 m/s; 0.95 m/s, and 1.72 m/s. Results in Fig. 1 indicate that the increase in pressure drop from the baseline value for each of these three experiments was different. The reactor operated at highest flow velocity showed an immediate increase in pressure drop, while the two others had initial lag phase periods. Generally, the lower the flow velocity the longer the lag phase. After the lag phase period the pressure drop increased exponentially until reaching steady state, except for the reactor operated at flow velocity 0.95 m/s. However, the sudden decrease in pressure drop monitored after 300 hours of operation in this reactor was due to biofilm sloughing.

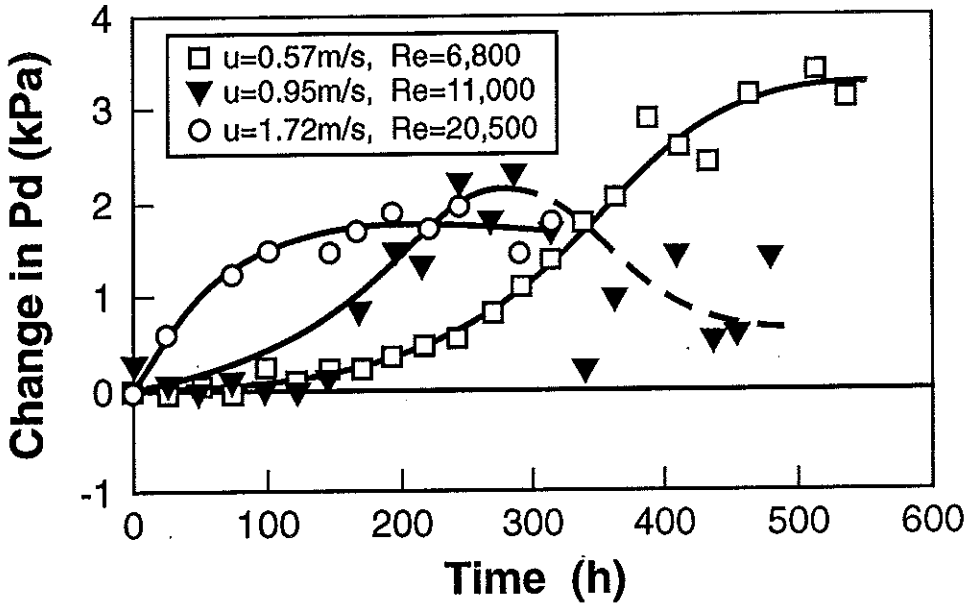

Figure 1. Pressure drop (Pd) increase across the flow channel for biofilms accumulating at various average flow velocities (u), the corresponding Reynolds numbers (Re) are indicated on the figure. To more easily compare the increase in Pd attributed to the biofilm accumulation the baseline Pd value (in a clean reactor, see Fig. 3) was subtracted from each of the measurements.

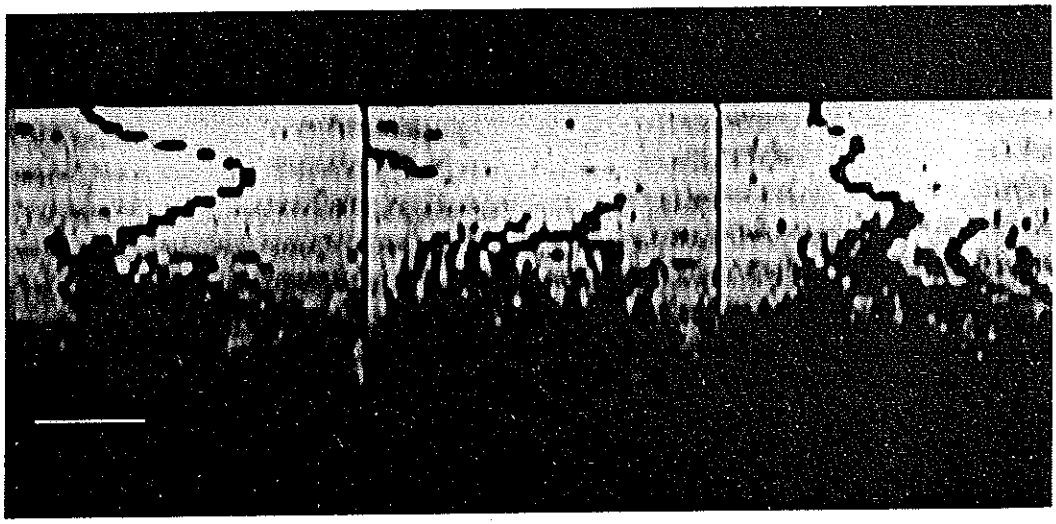

Figure 2. Biofilm streamers attached to the underlying biofilm clusters moving in the flow stream. The time sequence of images gives a plan view looking at the biofilm attached to the side wall of the channel (bottom of image). Scale bar = 1 mm.

We noticed that, as time progressed, the biofilm clusters elongated down stream to form what we termed "streamers". Streamers were subjected to rapid movements documented in (Fig. 2). This movement was transferred to the underlying clusters as evidenced by the circular oscillating paths described by attached fluorescent beads near the base of the biofilm (data not shown). As the flow velocity was increased the diameter of the bead path also increased.

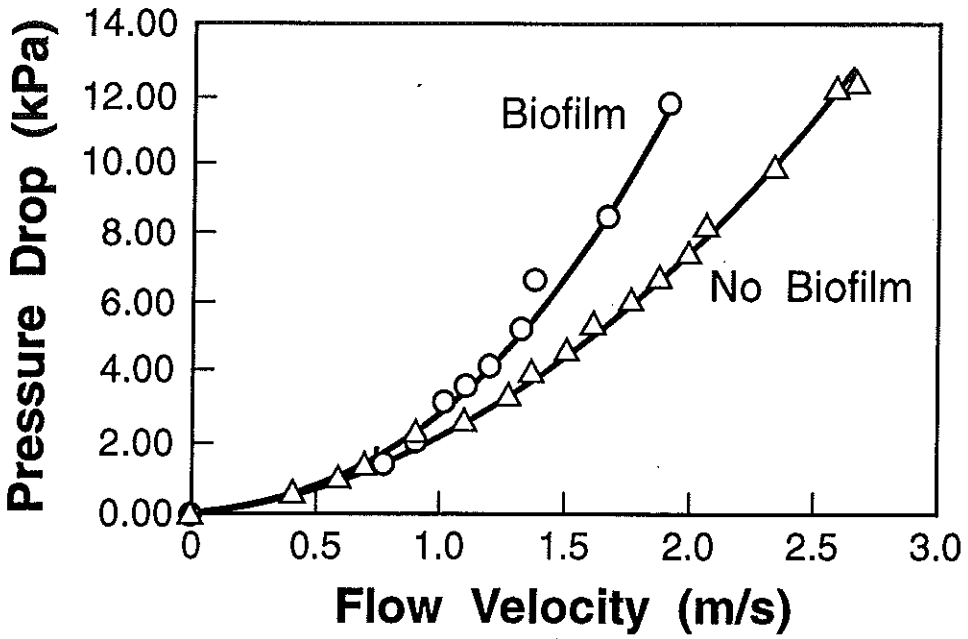

Figure 3. Pressure drop measured across a clean reactor and a biofilm colonized reactor at various average flow velocities. The biofilm was grown with a liquid flow velocity of 0.95 m/s.

The influence of flow velocity on the pressure drop was investigated in the presence and absence of biofilm (Fig. 3). The results show that the noticeable influence of biofilm on the pressure drop occurred above a flow velocity of approximately 0.6 m/s. At higher flow velocities there was a greater pressure drop across the reactor colonized with biofilm than across the clean reactor. The biofilm sloughed off when the flow velocity reached 1.7 m/s, with a corresponding pressure drop of about 12 kPa.

DISCUSSION

The interpretation of these results focuses on the nature of the friction resistance and the build-up of pressure drop in conduits colonized with biofilm as related to biofilm structure and elasticity, and in particular to the movement of the steamers. These streamers can reach considerable length (up to 2–3 mm) greatly exceeding the biofilm thickness (Fig. 2). The vigorous movement of the streamers is evidence that the kinetic energy of flowing water is being dissipated, resulting in an increase in pressure drop across the reactor.

The mechanism of streamer formation can be related to hydrodynamics. Initial stages of biofilm accumulation lead to the formation of discrete microcolonies firmly attached to the surface. Each of these microcolonies behaves as a bluff body attached to the reactor's wall. As water particles flow toward the leading edge of such a body the pressure in the water would increase above that of the free stream. The high pressure near the leading edge could induce the development of boundary layers on both sides of the microcolony. At higher flow velocities the viscosity forces may not be sufficient to force the boundary layers around the back (downstream) side of the microcolony. Near the widest section of the microcolony the boundary layers could separate from each side and form two shear layers. These two shear layers would in turn form a wake. Since the innermost part of the shear layers move slower than the outermost portions which are in contact with the free stream the free shear layers would tend to roll up into discrete vortices. A regular pattern of vortices is formed in a wake that interacts with body motion and is the source of the effect

ed vortex induced vibration (Griffin and Ramberg, 1974). In biofilms it may cause the development of downstream pressure drag. The biopolymers holding the microcolony together are viscoelastic and subjected to external force would be expected to assume an elongated shape. The dynamics of such action is directly related to the flow velocity, to the shear stress, and the magnitude of the pressure drag. This mechanism would explain the existence of lag phases for the development of pressure drop for lower flow velocities. It takes longer for the streamers to develop when the forces are smaller. For high flow velocities the length of the streamers is reduced because of the shearing action and the lag phase in pressure drop development disappears.

Biofilm accumulation is frequently associated with the increase of surface roughness. Our previous results using Nuclear Magnetic Resonance Imaging of flow (Lewandowski *et al.*, 1995) indicated that for low flow velocities biofilm formation actually smoothed the colonized surfaces. The result of the biofilm presence on pressure drop is obvious only after the flow velocity reaches a certain critical value, which under our experimental conditions was approximately 0.6 m/s (Fig. 3). The presence of biofilm, and the action of streamers did not influence the pressure drop below this flow velocity. Our NMRI measurements were conducted at the flow velocities of a few cm/s.

The Reynolds number at which the formation of a vortex street can be expected is reported to be much in the range of a few hundred (Blevins, 1977). This is much lower that the Reynolds numbers calculated for the reactors used in this experiment (a few thousand). However, the Reynolds number for biofilm reactors is traditionally calculated based on the geometry of the reactor. This may not be the best indication of flow stability near viscoelastic surfaces, such as that of the biofilm. Clearly, the characteristic length based on the geometry of the reactor may reflect the stability of flow in the reactor but not necessarily reflect the flow stability near the wall, particularly when the geometry of roughness elements (streamers) is changing in time. It is customary in such a situation to base the Reynolds number on the dimension of the object subjected to external forces rather than the geometry of the reactor. Blevins (1977) presents the regimes of fluid flow across circular cylinder. His numerical estimations may serve to analyze the effects of single microcolonies on the water flow. At very low Reynolds numbers (Re <5, based on cylinder diameter) the flow does not separate. Above Re=50 vortices are formed and shed periodically. Up to Re of 150 the vortex street is laminar. At Re 300, the vortex street is turbulent and it degenerates into fully turbulent flow beyond approximately 50 diameters downstream of the cylinder. Between the Re 300 and 3×10^5 the shedding occurs at a well defined frequency (Blevins, 1977). Assuming (from Fig. 2) that the average size of the streamer's base is 200 microns (2×10^{-6} m) the Reynolds number calculated for an average flow velocity applied in our experiments of 1 m/s is 200 which agrees well with the previous data and corroborates our assumptions of vortex street formation.

In biofilms the vortices cause the streamer to move. This movement is transferred back to the underlying biofilm causing the cluster to oscillate. When a bluff cylinder is excited into resonant oscillations by an incident flow, the cylinder and its shed vortices have the same frequency near one of the characteristic frequencies of the body. This coincidence of resonance of the shedding and vibration frequencies, termed lock-on (Hall and Griffin, 1993), cause the cylinder to vibrate. Such vibration, particularly near the shedding frequency may increase the vortex strength and result in the increase of the drag force (Bishop and Hassan, 1964). The oscillating forces cause vibrations in elastically mounted cylinders and the system behaves as a self-excited oscillator. Such vibrations induced in elastic structures by vortex shedding can have destructive effects for the entire structure. It is interesting to speculate that similar a effect in biofilms may induce localized sloughing events.

In summary we conclude that the pressure drop in conduits covered with biofilm to a large extent should be attributed to the oscillation of microcolonies and to the development of streamers. The formation of biofilm streamers and the effects of the biofilm on pressure drop can be explained using theories describing the force of liquid flow on a viscoelatic material. The streamers develop because of pressure drag exerted on a microcolony. The time needed to develop streamers depends directly on the magnitude of the exerted force, which originates from the flow velocity. The longer the streamers the more energy is dissipated by this way.

The final length of the streamers depends inversely on the flow velocity. For smaller flow velocities the streamers are longer and dissipate more energy but they take more time to develop.

CONCLUSIONS

1. The presence of biofilm influences the pressure drop only above a certain critical flow velocity. Below this flow velocity the presence of biofilm does not significantly influence the pressure drop.

2. The pressure drop in biofilm reactors is, at least partially, attributed to the oscillation of the biofilm and the formation of streamers. The streamers dissipate kinetic energy from the bulk liquid and vibrations of the streamers transfer energy to the underlying biofilm clusters which are induced to oscillate.

3. The pressure drop reaches steady (or pseudo steady) state in the reactor. This steady state is likely to correspond with the thickness of biofilms and the length of streamers, which also reach steady state.

4. It is, perhaps, timely to also recall our conclusions obtained by studying biofilm reactors using Nuclear Magnetic Resonance Imaging of flow. These results indicated that for low flow velocities biofilm formation effectively smoothed the channel walls. This apparently contradictory result fits into the hypothesis presented here and may contribute to our understanding of the nature of drag in biofilm reactors.

5. The interpretation of classical hydrodynamic parameters such as Reynolds number, friction factor, and surface roughness as related to biofilms should be reexamined in context to biofilm viscoelasticity and heterogeneity. It is also important to agree on what should be used as the characteristic length to calculate the Reynolds number to evaluate flow stability near biofilms. Although the Reynolds number calculated using the reactor geometry may be useful for predicting the overall flow stability, it may be more appropriate to use dimensions on a scale relevant to the biofilm structure to assess local flow conditions.

ACKNOWLEDGMENTS

This research was supported by the cooperative agreement EEC-8907039 between the National Science Foundation and Montana State University-Bozeman.

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