Comparison of models for bacterial regrowth in water distribution systems
by Naomi Ruth Wright Nichols

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in
Environmental Engineering
Montana State University
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Abstract:
This thesis evaluates dynamic water quality modeling programs for predicting microbial behavior in
drinking water systems. The specific computer programs evaluated were BAM, a biofilm system
modeling program and EPANET, a hydraulic modeling program with some water quality modeling
capabilities.

Experimental data from microbial regrowth research was used in conjunction with BAM to develop
and calibrate a model descriptive of the processes occurring in a drinking water pipe. The key
processes affecting BAM predictions of microbial regrowth were then identified. This information was
utilized to configure EPANET to simulate microbial regrowth. The capabilities and limitations of the
EPANET water quality model for simulating regrowth events in a distribution system were then
evaluated by correlating the EPANET results to experimental data obtained from pilot scale pipe loop
experiments.

The BAM program was successfully used to duplicate pilot scale experimental results for HPC
bacterial populations and to determine values of the unknown kinetic parameters. The development of
the BAM model yielded important information regarding biofilm systems in a water pipeline. Through
the model calibration process it was discovered that although the BAM program requires numerous
input terms, many of them do not have a significant influence on the simulation results.

Analysis of the modeling equations determined that detachment of cells from the biofilm into the bulk
fluid is the most significant process resulting in bulk fluid bacterial population increases, and is a first
order function of the film thickness. An important result, which relates to the accuracy of the EPANET
model of regrowth, was the conclusion that rate of regrowth is a function of the substrate concentration
since the biofilm growth and subsequent detachment depend on the available substrate.

Although EPANET was capable of simulating microbial populations, the model does not accurately
simulate regrowth since it does not account for the substrate limitation to microbial growth. Further
development of the EPANET model for the simulation of bacterial populations should include the
following improvements: o The ability to simulate substrate and bacterial populations concurrently.

- Development of more complex equations for modeling reaction rates.
- Modifications to the mass transfer calculation methods.
COMPARISON OF MODELS FOR BACTERIAL REGROWTH
IN WATER DISTRIBUTION SYSTEMS

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A thesis submitted in partial fulfillment
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This thesis has been read by each member of the thesis committee and has been found to be satisfactory regarding content, English usage, format, citations, bibliographic style, and consistency, and is ready for submission to the College of Graduate Studies.

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ABSTRACT

This thesis evaluates dynamic water quality modeling programs for predicting microbial behavior in drinking water systems. The specific computer programs evaluated were BAM, a biofilm system modeling program and EPANET, a hydraulic modeling program with some water quality modeling capabilities.

Experimental data from microbial regrowth research was used in conjunction with BAM to develop and calibrate a model descriptive of the processes occurring in a drinking water pipe. The key processes affecting BAM predictions of microbial regrowth were then identified. This information was utilized to configure EPANET to simulate microbial regrowth. The capabilities and limitations of the EPANET water quality model for simulating regrowth events in a distribution system were then evaluated by correlating the EPANET results to experimental data obtained from pilot scale pipe loop experiments.

The BAM program was successfully used to duplicate pilot scale experimental results for HPC bacterial populations and to determine values of the unknown kinetic parameters. The development of the BAM model yielded important information regarding biofilm systems in a water pipeline. Through the model calibration process it was discovered that although the BAM program requires numerous input terms, many of them do not have a significant influence on the simulation results.

Analysis of the modeling equations determined that detachment of cells from the biofilm into the bulk fluid is the most significant process resulting in bulk fluid bacterial population increases, and is a first order function of the film thickness. An important result, which relates to the accuracy of the EPANET model of regrowth, was the conclusion that rate of regrowth is a function of the substrate concentration since the biofilm growth and subsequent detachment depend on the available substrate.

Although EPANET was capable of simulating microbial populations, the model does not accurately simulate regrowth since it does not account for the substrate limitation to microbial growth. Further development of the EPANET model for the simulation of bacterial populations should include the following improvements:

- The ability to simulate substrate and bacterial populations concurrently.
- Development of more complex equations for modeling reaction rates.
- Modifications to the mass transfer calculation methods.
CHAPTER 1

INTRODUCTION

The quality of drinking water supplied to the 253 million Americans currently served by public water systems has long been a concern of Federal and State regulatory agencies and water utilities. This interest has become focused on the changes in quality which occur within water distribution systems and has led to increased research efforts to characterize and understand the hydraulic, chemical and bacteriological behavior of these systems.

Federal control of drinking water quality is the responsibility of the United States Environmental Protection Agency (EPA). The EPA, through the Safe Drinking Water Act and its 1986 Amendments (SDWAA), regulates water quality by specifying maximum contaminant levels (MCLs) for contaminants in drinking water. Water utilities, entities that supply water to the public, are required by law to provide drinking water that does not contain contaminants at levels that exceed the MCLs specified by the SDWAA.

One result of compliance with the drinking water quality regulations is an increase in water system operation and maintenance costs. In order to conform with the current regulations, and avoid violating any of the numerous MCLs, treatment techniques have become more complex. The regulations also require extensive monitoring of the treatment operations and sampling within the distribution system to ensure the
preservation of water quality. The necessary treatment, sampling, testing and analyses result in substantial expenses.

EPA estimates the cost to comply with the current SDWAA regulations to be $1.4 billion annually. Monitoring accounts for $253 million of this expense (JAWWA, Feb. 1994). Although the costs associated with complying with the SDWAA are considerable, the alternative can also be very expensive. In 1993, the largest civil fine ever collected under the SDWAA, $900,000, was paid by the Butte Water Company for supplying the city of Butte, MT with water that contained unacceptable levels of particulates (JAWWA, Feb. 1994). Particulates can harbor bacteria, viruses and/or parasites which cause a variety of severe health problems.

In addition to the fines levied by EPA for non-compliance, water utilities are often faced with lawsuits as a result of supplying low-quality drinking water. Over 1,400 legal claims, totalling approximately $25 million, have been filed against the city of Milwaukee, WI as a result of a 1993 cryptosporidiosis outbreak (JAWWA, May 1994).

The Milwaukee outbreak, which caused diarrhea in approximately 403,000 people, was caused by the protozoal parasite Cryptosporidium. The parasite entered the water distribution system as a result of decreased filtration efficiency and deteriorated raw water quality. Turbidity measurement at the time indicated inefficient filtration but none of the EPA regulations were violated (JAWWA, May, 1994).

The ongoing advance of technology has elevated the importance of the ability to predict drinking water quality. As the tools to measure contaminants in drinking water (indicators of water quality) become more sophisticated, detection limits become lower
and the list of regulated substances expands. The increasingly strict regulations on drinking water have led to increases in water system operational costs. In order to optimize treatment operations and thus minimize costs, water utility operators need the ability to predict the results of their treatment techniques beyond the treatment plant.

The cases in Butte and Milwaukee were both situations where a computer model of the distribution system water quality may have predicted the presence of pathogens at the point of consumption. However, there are currently no computer programs available that have the capabilities to simultaneously model all the physical, chemical and biological processes occurring within a drinking water distribution system.

There are numerous instances of waterborne disease outbreaks in municipal drinking water distribution systems every year. The cause of the outbreak can often be identified as an inadequate treatment or a distribution system deficiency, such as a water main break or a cross-connection to the sewer system. However, there are many cases where the cause is unknown.

Coliform bacteria are used as an indicator of the presence of disease-causing microorganisms in a water system. An indicator organism is one whose presence infers that contamination has occurred and suggests the nature and extent of the contaminant (Peavy, Rowe and Tchobanoglous, 1985).

As a measure to prevent disease outbreaks, EPA promulgated the Total Coliform Rule in June 1989, which states that coliform bacteria should not be detected in more than 5% of the samples of finished drinking water. However, coliform bacteria are
known to inhabit the biofilms present on pipe walls, and their presence within the biofilm environment complicates efforts to monitor bacterial quality (EPA, 1992).

A biofilm is defined as an accumulation of cells immobilized at a substratum (such as a pipe wall) and frequently embedded in an organic polymer matrix of microbial origin (Characklis and Marshall, 1989). Once microorganisms have entered a distribution system, they can attach to the pipe surfaces and grow within a biofilm. Biofilms can harbor a variety of microorganisms, including coliform bacteria and other opportunistic pathogens, organisms that cause disease in individuals with weak immune systems, but do not infect healthy people (EPA, 1992).

Biofilm growth, which has not been linked to disease outbreaks, can hide the presence of pathogenic bacteria that enter the water system through a loss of integrity of the treatment or distribution systems (EPA, 1992). As the biofilm accumulates, eventually portions of it will be sloughed off the pipe wall. The process of the microorganisms re-entering the bulk water is known as regrowth. When elevated levels of microorganisms in drinking water samples are observed, the occurrence of regrowth makes it difficult to determine the source of contamination.

In an effort to predict the complex behavior of biofilm systems, computer programs were developed in the 1980’s to model many of the processes occurring in a biofilm system (Wanner, 1989). The Swiss Federal Institute for Water Resources and Water Pollution Control has made a significant contribution to the biofilm modeling field with the program BIOSIM, written by Reicher, Ruchti and Wanner.
The BIOSIM model was modified by the Center for Biofilm Engineering at Montana State University to create a more user-friendly program and to allow the simulation of the effects of a biocide. The modified program is BAM, the acronym for Biofilm Accumulation Model (Goldstein, 1992).

BAM simulates the evolution of a mixed culture biofilm system within a series of "units". The BAM program is capable of modeling the development and maintenance of a biofilm within a single drinking water pipe, but does not have the ability to model a network of pipes, such as a drinking water distribution system.

Water distribution system modeling was initiated in the 1950s and 1960s with the development of computerized hydraulic models. Hydraulic models are capable of simulating the physical properties of water systems over extended time periods, with varying demand and operational conditions.

In recent years, the development of hydraulic modeling programs has expanded to include source tracing and water age. Source tracing identifies, at every point in the distribution system, the percentages of the total flow from each water supply source. The water age feature determines the total travel time from each source to every point in the system.

KYPIPE and CYBERNET, based on the KYPIPE algorithm, are two programs which contain these capabilities. Another model, PICCOLO, has been developed in France. EPANET, a program developed at the EPA Risk Reduction Engineering Laboratory by Lewis Rossman in the Drinking Water Research Division combines hydraulic modeling and dynamic water quality modeling. EPANET has source tracing
and water age features, as well as the ability to track the concentration of a substance throughout a network over time.

Although EPANET was not developed to simulate microbial growth, its modeling equations account for reactions in the bulk fluid and at the pipe wall. Therefore, the potential exists for using EPANET to model a biofilm system.

The application of EPANET's dynamic water quality module for simulating bacterial regrowth was investigated as presented herein. The BAM program was utilized to simulate the formation of a biofilm on the wall of a drinking water pipeline, and then coupled with EPANET in an effort to model regrowth of bacteria within a drinking water distribution system. The modeling results were compared to actual water quality data to assess the feasibility of combining dynamic and biofilm system modeling.

The water quality data was obtained from pilot scale experiments that use a pipe loop system to create typical water distribution system conditions. The pipe loop system consists of a length of mild steel pipe which is fed treated drinking water. The water recirculates through the pipe loop to simulate relevant distribution system conditions. Bacterial growth is monitored by measuring the bacterial populations in the influent water, effluent water and at the pipe wall. The concentration of cells in the biofilm at the pipe wall are measured from samples taken from removable circular sections of the pipe (coupons).

The pipe loop experimental data was used to estimate initial conditions for the BAM input file and to develop data sets to correlate with BAM and EPANET results.
CHAPTER 2

PROBLEM STATEMENT

The ability to predict water quality is useful for providing public health protection and for maximizing water system operation economy. However, current methods for predicting water quality are limited by the capabilities of the available computer models. The goal of this thesis is to evaluate dynamic water quality models for predicting microbial behavior in drinking water systems. This goal will be accomplished by completing the following objectives.

Objective I

Using experimental data from microbial regrowth research and a biofilm modeling computer program (BAM), develop and calibrate a model descriptive of the biofilm processes occurring in a drinking water pipe.

Objective II

Identify the key processes affecting BAM predictions of microbial regrowth.

Objective III

Evaluate the accuracy of EPANET modeling results. Assess the capabilities and limitations of the EPANET water quality model for simulating regrowth events in the distribution system.
A biofilm is a thin layer of microorganisms attached to a solid surface. Biofilms can develop on almost any surface exposed to an aqueous environment (Reichert, Ruchti and Wanner, 1989). A biofilm system, represented in Figure 1 (adapted from Biofilms), consists of different compartments, generally a solid substratum, the biofilm, bulk water and possibly gas. Analysis and prediction of biofilm behavior is complicated by the heterogeneous nature of the biofilm and the variety of physical, chemical and biological processes that occur within the biofilm system (Characklis and Marshall, 1989).

**FIGURE 1**
BIOFILM SYSTEM
The following conceptual and mathematical descriptions of a biofilm system are based on Chapter 11 of *Biofilms*, contributed by Gujer and Wanner (Characklis and Marshall, 1989). Numerous variables are contained in the equations presented in the following text, which are defined as they are introduced. A summary table of these terms is provided by the Nomenclature section.

The biofilm compartment consists of a continuous liquid phase, which contains different dissolved and suspended particles, and solid phases of attached particulate materials such as microorganisms and extracellular material, as depicted in Figure 1. Each phase \( k \) occupies a fraction of the total biofilm volume. The sum of the volume fractions for the liquid phase and the solid phases must equal 1, expressed by Equation 1:

\[
\sum_k e_k = e_l + \sum_s e_s = 1
\]

where \( e \) = local volume fraction of the total biofilm volume

**Description of Processes**

The many processes which affect the formation and subsequent behavior of a biofilm can be classified into three general categories: transport processes, transformation processes and interfacial transfer processes. These processes are summarized below:

**Transport processes.** Biofilm transport processes include molecular diffusion, turbulent or eddy diffusion and advection.
Transformation processes. These processes are characterized by a molecular rearrangement and may be chemical, biochemical or microbial in nature (Characklis and Marshall, 1989). For the biofilm systems present in a water distribution pipeline, these processes include growth, decay and inactivation of microorganisms.

Interfacial transfer processes. Attachment and detachment are included in this category, as well as the physical processes of adsorption, absorption and desorption which occur within the biofilm matrix.

In order to develop a model of biofilm behavior, these processes must be described in mathematical terms, through a series of equations. The primary equation is the biofilm mass balance equation, which is written as:

\[
\frac{\partial \varepsilon_k C_{ki}}{\partial t} = -\frac{\partial J_{ki}}{\partial z} + r_{ki}
\]

where  \( C_{ki} \) = mass of component \( i \) contained within a unit volume of phase \( k \) (M/L^3)

\( J_{ki} \) = flux of component \( i \) within phase \( k \) per unit total cross-sectional area of biofilm (transport process rate) (M/L^2T)

\( r_{ki} \) = rate of production of component \( i \) within phase \( k \) per unit total volume of biofilm (transformation process rate) (M/L^3T)
In addition to the mass balance on the biofilm, boundary or continuity conditions must be defined for the interfaces between the compartments of the biofilm system. The continuity condition is expressed by the following equation for the interface between two compartments:

$$u_I \cdot (e_{k1} \cdot C_{k1} - e_{k2} \cdot C_{k2}) = J_{k1} - J_{k2} + r''_{ki} \tag{3}$$

where

- $u_I$ = velocity of the interface relative to the fixed coordinate $z$ (L/T)
- $r''_{ki}$ = amount of component $i$ produced per unit total cross-sectional area of the interface (interfacial transfer process rate) (M/L²T)

The indices 1 and 2 refer to the sides of the interface, side 2 has higher $z$ coordinates.

**BAM Program**

Based on the fundamental equations presented above and assumptions concerning the various processes, the computer model BIOSIM was developed which allows simulation of the dynamics of biofilm systems (Reichert, Ruchti and Wanner, 1989). The assumptions made and the resulting simplified equations used by the BIOSIM model are included as Appendix A. In general, the BIOSIM model simulates the processes of molecular diffusion, advection, attachment, detachment and any transformation processes defined by the user.

The BAM model, a modified version of the BIOSIM program, was used in this research to simulate the formation and performance of a biofilm on the wall of a drinking
water pipeline. The simulated processes included the growth and decay of microorganisms within a biofilm and the bulk fluid, the consumption of substrate (food) for microbial growth, the detachment of the microorganisms from the wall into the bulk fluid, the advection of microbial cells as a result of net growth and the diffusion of substrate into the biofilm. Figure 2 is a representation of the processes modeled by the BAM program.

\[ Q = \text{FLOW RATE} \]
\[ S = \text{SUBSTRATE (AOC)} \]
\[ X = \text{PARTICULATES (CELLS)} \]
\[ L_r = \text{BIOFILM THICKNESS} \]
\[ r_i = \text{PROCESS RATE} \]
\[ j_i = \text{MASS FLUX} \]

**PROCESSES SIMULATED**
- \( r_x \): Growth of cells in bulk fluid and biofilm
- \( r_x \): Decay of cells in bulk fluid and biofilm
- \( j_x \): Detachment of cells from biofilm
- \( j_s \): Advecive flux of biofilm cells
- \( r_s \): Substrate consumption
- \( j_s \): Substrate diffusion

**FIGURE 2**
BAM REPRESENTATION OF PROCESSES
BAM models the transport, transformation and interfacial transfer of \( i \) components which are either particulate (X) or dissolved (S) species. For the distribution system application, the particulate species are microorganisms (heterotrophic plate count and coliform bacteria) and the dissolved species is the substrate, assimilable organic carbon (AOC). The equations used by BAM for representing the various processes are presented in the BIOSIM users manual and described below:

**Transformation Processes.** The growth of microorganisms within the biofilm and the bulk fluid was assumed to follow Monod kinetics, written as:

\[
\mu = \mu_m \left( \frac{C_S}{K_S + C_S} \right)
\]

where
- \( \mu \) = specific growth rate (1/T)
- \( \mu_m \) = maximum specific growth rate (1/T)
- \( C_S \) = substrate (AOC) concentration (M/L³)
- \( K_S \) = half-saturation constant (M/L³)

Assuming that microbial decay is a first order process, the net transformation rate of the particulate species (microorganisms) was defined as:

\[
r_X = \mu \cdot C_X - b \cdot C_X
\]

where
- \( b \) = decay rate coefficient (1/T)
- \( C_X \) = concentration of particulates (bacteria) (M/L³)

The transformation rate of the dissolved species (substrate) was defined as:
where \( Y_{X/S} \) = Yield coefficient (grams of microorganisms produced per gram of substrate) (M/M)

These transformation process rate \( (r_d) \) equations apply to both the bulk fluid and the biofilm. The rate equations are differentiated by subscripts for the bulk fluid (B) and the biofilm (F) which apply to the process rate and to the component concentrations. For example, the equation for the transformation of microorganisms (X) within the biofilm (F) is written as:

\[
(r_x)_F = \left( \frac{\mu_m (C_s)_F - b}{K_s + (C_s)_F} \right) \times (C_x)_F
\]

where \((C_s)_F\) = concentration of substrate in the biofilm

\((C_x)_F\) = concentration of cells in the biofilm = \(q_x \times \epsilon_s = q_F \) (M/L³)

\(q_x\) = density of cells (M/L³)

\(q_F\) = biofilm mass density (M/L³)

Interfacial Transfer Processes. The interfacial transfer rate was modeled as a net detachment rate, the sum of detachment and attachment, since the magnitude of the individual processes cannot be identified. The modeled detachment rate \(r_d\) is the product of a detachment velocity, \(u_{de}\), and the concentration of microorganisms in the film, \((C_x)_F\).

The BAM program can model detachment velocity in a variety of ways: as a function of biofilm thickness \((L_p)\), biofilm thickness squared, growth rate, or
concentration of microorganisms. The detachment can also be equal to a constant. A negative value for the detachment velocity would be used to represent attachment. As presented in Chapter 5 of this thesis, each of these methods was investigated to determine the most accurate representation of detachment from the pipe wall.

Transport Processes. The transport processes modeled by BAM are the advective flux of microbial cells and the diffusion of substrate into the biofilm. The advective flux, \( j_X \), is described by the following equation:

\[
j_X = u_F \cdot (C_X)_F
\]

where \( u_F \) = velocity by which particulate mass is displaced relative to the solid surface (substratum) (L/T)

The diffusion of substrate is calculated using Fick’s first law to determine the mass flux of the substrate, \( j_S \):

\[
j_S = -f \cdot D \cdot \frac{\partial (C_S)_B}{\partial z}
\]

where \( D \) = diffusivity in pure water (L²/T)

\( f \) = ratio of the diffusivities in the biofilm and in pure water

Biofilm. The development of the biofilm is described by:

\[
\frac{d(L_p)}{dt} = u_L = u_F(z = L_p) - u_{de}
\]

where \( L_p \) = biofilm thickness (L)
\[ u_L = \text{velocity by which biofilm surface is displaced relative to the substratum (L/T)} \]

\[ u_R = \text{velocity particulate mass is displaced relative to the solid surface (L/T)} \]

\[ u_{de} = \text{net detachment velocity (L/T)} \]

**Bulk Fluid.** The dynamics of both the dissolved and the suspended particulate components \((i)\) in the bulk fluid are described by the mass balance equation:

\[
\frac{d(V_B(C_i)_B)}{dt} = Q((C_i)_B - (C_i)_0) + A_F * j_i + V_B *(r_i)_B
\]  

(11)

where \((C_i)_B\) = concentration of component \(i\) in the bulk fluid \((M/L^3)\)

\((C_i)_0\) = concentration of component \(i\) in the influent \((M/L^3)\)

\(V_B\) = bulk fluid volume \((L^3)\)

\(Q\) = volumetric inflow rate \((L^3/T)\)

\(A_F\) = area in the unit covered by biofilm \((L^2)\)

\(j_i\) = mass flux between biofilm and bulk fluid per unit area of film \((M/L^2T)\)

\((r_i)_B\) = net transformation rate in the bulk fluid \((M/L^3T)\)

**Water Quality Models**

The existing dynamic water quality models that simulate the movement and transformation of substances in water under time-varying conditions use simplified
mathematical relationships to represent the physical, biological and chemical processes occurring in a distribution system. The concentration of substances is assumed to follow the first order decay function; that is, the rate of consumption of a substance is proportional to its concentration.

This relationship has been generally accepted as a model of chlorine decay. However, the application of this function to modeling the regrowth of microorganisms within a drinking water system is highly questionable.

**EPANET Program**

EPANET, developed by the Environmental Protection Agency’s Drinking Water Research Division of the Risk Reduction Engineering Laboratory, can perform extended period simulations of hydraulic and water quality behavior within drinking water distribution systems. The water quality module has been successfully used to simulate chlorine decay, fluoride tracer analysis, and source tracing.

The following summary of the EPANET algorithm has been condensed from the EPANET Users Manual.

The EPANET program represents water pipes as links and the endpoints of the pipes as nodes. The hydraulic model used by EPANET for extended period simulations solves the following set of equations for each link, with nodes a and b, and for each node n:

\[ h_a - h_b = f(Q_{ab}) \]  \hspace{1cm} (12)
\[ \sum_a Q_{an} - \sum_b Q_{nb} - Q_n = 0 \]  

(13)

For each storage node \( s \), which represents a tank or reservoir, the following equations are used:

\[ \frac{\partial y_s}{\partial t} = \frac{Q_s}{A_s} \]  

(14)

\[ Q_s = \sum_a Q_{as} - \sum_b Q_{sb} \]  

(15)

\[ h_s = E_s + y_s \]  

(16)

where \( h_a \) = hydraulic grade line elevation at node \( a \) (elevation head plus pressure head) (L)

\( Q_{ab} \) = flow in pipe connecting nodes \( a \) and \( b \) (L\(^3\)/T)

\( Q_s \) = flow in or out of storage node \( s \) (L\(^3\)/T)

\( f(Q_{ab}) \) = functional relation between head loss and flow in a link, can be the Hazen-Williams, Darcy-Weisbach or Chezy-Manning formula (L)

\[ f(Q_{ab}) = \frac{(4.72 L_{ab} Q_{ab}^{1.85})}{(C^{1.85} d^{4.87})} \] for Hazen-Williams formula, when \( L_{ab} \) and \( d \) are expressed in feet and \( Q \) is expressed as ft\(^3\)/s

\( L_{ab} \) = pipe length (L)

\( C \) = Hazen-Williams roughness coefficient

\( d \) = pipe diameter (L)
\[ Q_n = \text{flow consumed or supplied at node } n \ (L^3/T) \]

\[ y_s = \text{height of water stored at node } s \ (L) \]

\[ A_s = \text{cross-sectional area of storage node } s \ (\text{infinite for reservoirs}) \ (L^2) \]

\[ E_s = \text{elevation of node } s \ (L) \]

From the specified storage node elevation and initial water height, equation 16 is used as a boundary condition for iteratively solving equations 12 and 13 for all flows \( Q_{ab} \) and heads \( h_a \) at time zero. The initial network hydraulic solution is utilized with equation 16 to calculate the storage node flow \( Q_s \) and the new storage water height for the next time step is determined from Equation 14. The solution process is repeated for each subsequent time step.

The results of the hydraulic simulation are used by the water quality simulator to track the fate of a dissolved substance flowing through the network over time. The flows generated by the hydraulic solution are utilized to solve the following conservation of mass equation for the substance within each link:

\[
\frac{\partial(C_i)_B}{\partial t} = \nu \frac{\partial(C_i)_B}{\partial L} + r_i 
\]

where \( \nu = \text{velocity} = Q / \text{cross-sectional area} \ (A) \ (L/T) \)

\[ A = \pi d^2/4 \ (L^2) \]

\[ r_i = \text{rate of reaction of component } i \text{ within link} \ (M/L^3T) \]

Equation 17 is solved with a specified initial substance concentration and the following boundary condition from conservation of mass at the beginning of a link (designated node \( o \)), with \( P \) links joining at node \( o \):
\[ (C_i)_{Bo} = \frac{\sum P Q_p + (C_i)_{Bp}}{\sum P Q_p + Q_E} + (C_i)_{Bk} \]  

where \((C_i)_{BE}\) = substance mass introduced by any external source at node \(o\) (M)  

g = flow rate of external source (M/L³)

The numerical method used by EPANET for solving these equations is known as the Discrete Volume Element Method (DVEM). For each hydraulic time period (of a duration specified by the user), a shorter water quality time step is calculated and each pipe is divided into a series of completely mixed volume segments. Within each water quality time period, the substance contained in every pipe segment is transferred to the next downstream segment. When the next segment is a node, conservation of mass is used to compute the resulting concentration leaving that node. The resulting concentrations at each node are then released into the head end segment of pipes with flow leaving the node. Following the transport phase, the mass within each pipe segment is reacted. This sequence is repeated for the subsequent water quality time steps until the next hydraulic time step, when new pressures and flow rates are calculated, and the entire process is repeated.

Equation 17 calculates the change in substance concentration as the result of hydraulic transport and reaction in the bulk fluid. The equation used for modeling the growth of a substance is given below:
\[ r_i = + k_B \cdot (C_i)_B \cdot \left( \frac{k_f}{R_H} \right) \cdot (C_i)_B - c_w \]  \hspace{1cm} (19)

where  
- \( k_B \) = first-order bulk reaction rate constant (1/T)  
- \( k_f \) = mass transfer coefficient between bulk fluid and pipe wall (L/T)  
- \( R_H \) = hydraulic radius of pipe = \( d/4 \) (L)  
- \( c_w \) = substance concentration at the wall (M/L^3)

EPANET determines the concentration at the wall from the following mass balance equation:

\[ k_f \cdot (C_i)_B - c_w = k_w \cdot c_w \]  \hspace{1cm} (20)

where \( k_w \) = wall reaction rate constant (L/T)

Equation 20 equates the mass transfer to a first order reaction rate at the pipe wall. For modeling the microbial population, the wall reaction rate corresponds to the mass flux at the biofilm surface, \( j_X \) at \( z=L_P \), modeled in the BAM program. This relationship will be further analyzed in Chapter 4.

Based on Equation 20, the reaction rate equation can be rearranged to eliminate the \( c_w \) term:

\[ (r_i)_B = k_B \cdot (C_i)_B + \frac{k_w \cdot k_f}{R_H \cdot (k_w + k_f)} \cdot (C_i)_B = (K_1 + K_2) \cdot (C_i)_B \]  \hspace{1cm} (21)

where \( K_1 \) and \( K_2 \) represent overall first order rate constants for the bulk fluid and the pipe wall reactions, respectively.

The elimination of the wall concentration simplifies the comparison of the EPANET and BAM modeling equations, as presented in Chapter 4.
CHAPTER 4

METHODS

Experimental Data

The basic approach to modeling water quality in drinking water distribution systems was to use experimental results to develop a Biofilm Accumulation Model (BAM) of biofilm growth and detachment. The BAM model parameters were used to predict rate constants for EPANET. EPANET was then used to model the regrowth of microorganisms in a distribution system network. Finally, the accuracy of the EPANET model was assessed to determine the feasibility of using the EPANET water quality module for regrowth phenomena.

The American Water Works Association Research Foundation (AWWARF) provided the Center for Biofilm Engineering at Montana State University with funds to conduct a project investigating regrowth in water distribution systems, Factors Limiting Microbial Growth in the Distribution System. As a major part of this project, experiments to study the development of biofilms and the related regrowth of microorganisms within water pipelines were initiated in 1992 and completed in 1995.

The pilot scale experimental setup includes annular reactors and pipe loops, designed to model the hydraulic conditions of a water distribution system pipeline. The two systems operate as continuous flow stirred tank reactors (CFSTR), in which no
concentration gradients exist within the bulk fluid volume, and are useful for observing and evaluating biofilm processes (Characklis, 1989).

The annular reactors and pipes, which are both mild steel, include removable mild steel circular sections known as coupons. The coupons are used to determine the concentration of microorganisms within the biofilm and thereby monitor biofilm development.

Compared to pipe loops, rotating annular reactors are more desirable to use as a monitor of biofilm processes, mainly due to their size. However, the degree of accuracy of annular reactors for modeling water distribution pipeline conditions has not been established. The results of the AWWARF project will determine if the annular reactor results duplicate the pipe loop results and can therefore be used as a more convenient monitoring device.

Since the pipe loops have been shown to be reasonable physical models of pipeline distribution conditions, the data obtained from the pipe loop experiments was used to develop the BAM model (Camper, 1991). Therefore, the following discussion of the experiments will refer to the pipe loops, although the annular reactors were operated under the same conditions.

The experiments completed at the time of this writing are briefly described below:

Description of Experiments

Experiment 1. Five pipe loops were configured in series to simulate 2, 4, 8 and 16 hour residence times in order to determine the most favorable residence time for
growth of microorganisms. All subsequent experiments were performed with parallel loops and at a 2 hour residence time since this was found to be the optimum time.

Experiment 2. Assimilable organic carbon (AOC) and temperature were varied in the 5 different loops to determine the effect of varying AOC and temperature on biofilm accumulation/microorganism growth.

Experiment 3. Chlorine and temperature were varied to determine the effect on the biofilm and the microorganisms. Experiment 3 was performed in the summer.

Experiment 4. Duplicate of experiment 3, performed in the winter.

A schematic of the two hour residence time pipe loop is included as Figure 3. The pipe loops were set up at the Bozeman Water Treatment Plant and fed water from the treatment plant clearwell. The loop influent water was dechlorinated through a GAC column and the AOC reduced by passage through filters containing biologically active carbon. Analysis of the influent water has shown that assimilable organic carbon was present in concentrations which average between .02 to .2 mg/L. Average heterotrophic plate count (HPC) bacterial concentrations showed annual variations of 4,000-30,000 CFU/mL and coliform bacteria were not detected.

The water was supplied to the 4" diameter loops at a rate of 3.7 m³/d (39 gpm) in order to maintain a 2 hour residence time. The recycle rate was set to achieve a flow through the pipe of 213 m³/d, corresponding to a velocity of 1 ft/s. For the pipe loops with substrate addition, assimilable organic carbon was added to the influent water at a
constant concentration of 0.5 mg/L. Nitrate and phosphate (0.1 mg/L) were always present so the AOC was the limiting substrate. To inoculate the system with coliform bacteria, they were added at a concentration of approximately 10,000 CFU/mL at the beginning of each experiment. After the inoculation of coliforms, the system operated until a biofilm on the pipe wall had developed, then data collection began.
The data collected included the AOC concentrations in the loop effluent water (hereafter referred to as the bulk fluid), the concentration of heterotrophic plate count bacteria (HPCs) and viable coliform bacteria in the biofilm and in the bulk fluid.

AOC concentrations were obtained using the p17 and NOX measurement techniques presented by van der Kooij in Determining the Concentration of Easily Assimilable Organic Carbon in Drinking Water, (JAWAA, 1982).

Microbial populations were determined using plate count techniques. The bulk fluid population are reported as colony forming units (CFU) per mL of sampled bulk fluid. The biofilm population is determined by counting the number of CFUs from a coupon sample and is reported as CFU/cm². For modeling purposes, the bulk fluid populations were converted to concentrations in mg/L and the area-averaged biofilm population was converted to a biofilm thickness. The methods used to perform these conversions are presented below.

Microbial Population Conversion Techniques

Equations to convert the populations into concentrations with appropriate units for modeling were developed using the following assumptions and definitions.

Assumptions:

1. A colony is formed from 1 cell: 1 CFU = 1 cell
2. A cell is 90% water, the density of a cell is the same as the density of water:
\[ \rho_{\text{dry cell}} = 0.1 \times \rho_{\text{wet cell}} = 0.1 \times \rho_{\text{H2O}} = 0.1 \times 10^6 \text{ g/m}^3 = 10^5 \text{ g/m}^3 \]
3. A cell is approximately the shape of a cylinder and is 1 µm high and 0.5 µm in diameter.
4. According to experimental results from lab studies at the Center for Biofilm Engineering (for the same substrates and residence time conditions), a biofilm with $10^6$ CFU/cm$^2$ covers 60% of the total pipe surface area. Thus, 100% coverage (a monolayer) would contain $1.6 \times 10^6$ CFU/cm$^2$.

5. The biofilm solid phase consists of only microorganisms and occupies 2.5% of the biofilm volume (Characklis and Marshall, 1989).

Calculated Cell Properties:

Cell volume:

$$V_{cell} = \frac{\pi \times (0.5\mu m)^2 \times 1\mu m}{4} = 1.96 \times 10^{-19} m^3 \quad (22)$$

Dry cell mass:

$$M_{drycell} = V_{cell} \times \rho_{drycell} = 1.96 \times 10^{-19} m^3 \times 10^5 \frac{g}{m^3} = 1.96 \times 10^{-14} g \quad (23)$$

Conversion Equations:

Bulk Fluid Concentration - $(C_{\chi})_B$ in mg/L:

$$\frac{mass \ of \ cells}{fluid \ volume} = \frac{CFU}{fluid \ volume} \times \frac{1 \ cell}{CFU} \times \frac{mass \ per \ cell}{mass} \quad (24)$$

$$\frac{mg \ cells}{L \ fluid} = \frac{CFU}{mL} \times 1 \ cell \times 1.96 \times 10^{-14} g \times 10^3 mg \times 10^3 mL}{g \ L} \quad (25)$$
Biofilm Thickness - $L_F$ in $\mu m$:

$$L_F(\mu m) = \frac{X''_{100} \times V_{cell}}{\varepsilon_S}$$

$$= \frac{\left(\frac{X''}{.6}\right) \times 1.96 \times 10^{-19} \, m^3}{.025}$$

$$= \frac{CFU \, cell \, 1.96 \times 10^{-19} \, m^3 \times 10^4 \, cm^2 \times 10^6 \mu m}{cm^2 \, CFU \, cell \, m^2 \, m \times .6 \times .025}$$  \hspace{1cm} (26)$$

**Model Development**

**BAM Model**

Experimental data were used as the basis for developing a computer model of the experiments in the BAM program. As discussed previously, BAM is capable of modeling the processes of biofilm development and detachment, microbial growth and decay, substrate consumption, and substrate and particulate transport. BAM requires numerous input parameters, which were determined by a variety of methods, including literature review, known conditions, experimental results, as well as trial and error.
The BAM base model was developed and calibrated using the heterotrophic bacteria population results of Experiments 1 and 2. The measured concentrations of heterotrophic bacteria were much higher than the coliform bacteria concentrations; coliforms in the bulk fluid were either not detected or were present at concentrations of only a few CFU per mL. Since so few coliforms were present, the BAM base model was initially developed to model the measured HPC concentrations. This model was then adjusted to model the coliform population. From the known conditions (flow rates, average influent concentrations, pipe loop surface area and volume, etc.), a preliminary model was created of a single pipe loop.

The 2 hour residence time pipe loop was represented as two BAM "units", one for the 40' long, 4" diameter pipe and a second unit for the recirculation piping, as depicted by Figure 4. The physical parameters used to describe the BAM units are their volume and surface area. These were calculated from the known dimensions of the pipe and recirculation system.

\[
\begin{align*}
A_{F1} &= \pi*d*L = \pi*4^2*40' = 3.89 \text{ m}^2 \\
V_{B1} &= (\pi*d^2/4)*L = (\pi*4^2/4)*40' = .0988 \text{ m}^3 \\
Q_{in} &= (V_{B1} + V_{B2})/\Theta = 306 \text{ L/2 hrs} = 3.7 \text{ m}^3/\text{d} \\
Q &= \frac{A * v}{1 \text{ ft/s}} = (\pi*d^2/4) * 1 \text{ ft/s} = 213 \text{ m}^3/\text{d}
\end{align*}
\]

**FIGURE 4**

BAM UNITS
The numerous unknown constraints are summarized in Table 1, along with a description of the various methods used for determining their values for the model. Table 4, which is presented in Chapter 5, lists the specific references used for estimating values of the unknown parameters.

<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>METHOD</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\mu_m, K_S, Y_{X/S}, b$</td>
<td>Estimated from literature review of studies for growth kinetics of heterotrophs in drinking water environments</td>
</tr>
<tr>
<td>Density of microorganisms ($Q_X$)</td>
<td>Estimated from reported values</td>
</tr>
<tr>
<td>Ratio of diffusivity biofilm/bulk ($f$)</td>
<td>Estimated from reported values</td>
</tr>
<tr>
<td>Diffusivity of substrate ($D_s$)</td>
<td>calculated for the specific assimilable organic carbon used as a substrate source</td>
</tr>
<tr>
<td>Liquid boundary layer thickness ($\delta_{11}$)</td>
<td>Fit to data</td>
</tr>
<tr>
<td>Biofilm volume fraction ($\epsilon_s$)</td>
<td>estimated from measured cell concentrations in the biofilm</td>
</tr>
</tbody>
</table>

Estimates of the various unknown input parameters were made and the base model was then calibrated by adjusting the various parameters to obtain results that matched the experimental data for bulk fluid HPC bacteria counts, substrate concentrations and estimated film thickness.

Once an accurate model of the processes occurring in a drinking water pipeline had been developed using BAM, the model was used as a basis for evaluating the water quality modeling capabilities of the EPANET program.
EPANET Model

The EPANET input file requires specifications for the pipe sizes and lengths, system demands, water sources and network elevations, which describe the distribution system physical characteristics for hydraulic modeling. The water quality simulation results depend on the specified bulk fluid and wall reaction rate constants, which can be applied throughout the network (global) or varied for individual pipes.

For the simulation of two pipes in series, the EPANET model was developed as shown by Figure 5, and the reaction rate constants were varied to determine their effect on the EPANET simulation results.

\[ Q = 39 \text{ gpm} \]
\[ (C_{X, \text{in}}) = .0001 \text{ mg/L} \]

\[ L_1 = 7200' \]
\[ L_2 = 7200' \]
\[ d = 4" \]
\[ d = 4" \]

**FIGURE 5**
EPANET REPRESENTATION

As presented in the Theory Section, the following equation is used by EPANET to model the growth rate of a substance \( i \).

\[ r_i = +k_b*(C_i)_B*\left( \frac{k_f}{R_H} \right)^*(C_i)_B - c_w \] (19)

For modeling microbial regrowth, the EPANET substance concentration, \((C_i)_B\), is the concentration of cells in the bulk fluid. The bulk fluid reaction rate constant, \(k_b\),
was set to zero since, as presented in Chapter 5, the growth rate of bacteria in the bulk fluid is negligible compared to the detachment rate. The hydraulic radius, \( R_H \), and the mass transfer coefficient, \( k_f \), are both calculated internally by the EPANET program based on the physical parameters used to describe the pipeline network (pipe diameter, pipe length, flow rate, water viscosity and temperature).

The only unknown variable in this equation is \( c_w \), which can be eliminated from the reaction rate equation as shown in Chapter 3 by Equation 21. The rearrangement of EPANET’s reaction rate equation to remove the concentration at the wall requires the user to specify the wall reaction rate constant \((k_w)\), an empirical parameter, in the model input file.

To make estimates of \( k_w \) for comparing the BAM and EPANET programs, the reaction rate equations used to model bulk fluid HPCs in BAM are compared to the terms of Equation 21 in Table 2.

<table>
<thead>
<tr>
<th>TABLE 2</th>
<th>MODEL EQUATIONS FOR ( d(C_X)_B/dT )</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><strong>BAM</strong></td>
</tr>
<tr>
<td><strong>Transformation rate</strong></td>
<td>( \left( \frac{u_m*(C_S)_B}{K_s+(C_S)_B} - b \right) * (C_X)_B )</td>
</tr>
<tr>
<td><strong>Transport rate</strong></td>
<td>( j_X \left( \frac{A_F}{V_B} \right) = \frac{u_d*(C_X)_F}{R_H} )</td>
</tr>
</tbody>
</table>
An equation for calculating the wall reaction rate constant was generated by equating the two different expressions used for modeling transport as shown in the following development:

\[
\frac{k_w \cdot k_f}{R_H \cdot (k_w + k_f)} \cdot (C_{x})_B = \frac{u_{de} \cdot (C_{x})_F}{R_H}
\]  \hspace{1cm} (27)

\[
k_w = \frac{(k_w + k_f) \cdot u_{de} \cdot (C_{x})_F}{k_f \cdot (C_{x})_B}
\]  \hspace{1cm} (28)

The detachment velocity and microbial concentrations can be determined from both the experimental data and the BAM model results and \(k_f\) is calculated from the hydraulic conditions. The fact that both experimental and model results can be used to make estimates of the only unknown input variable \((k_w)\) is very important. In the absence of actual data, the BAM program can be used to generate the information necessary to determine the input parameters for EPANET.

For the calculation of \(k_w\) from BAM model results, \(u_{de}\) and \((C_{x})_F\) are determined from the model input parameters for the detachment rate formula, volume fractions \((\varepsilon)\) and particulate component densities. \((C_{x})_B\) is the resulting bulk fluid HPC concentration.
The experimental results can be used to calculate a $k_w$ value based on the assumption that detachment velocity is a function of film thickness, defined as:

$$u_{de} = k_d L_F$$  \hspace{1cm} (29)$$

This expression for detachment velocity was confirmed through the BAM modeling phase, as presented in Chapter 5. Values for the specific detachment rate, $k_d$, have been calculated as part of the AWWARF regrowth project. By substituting equation 29 and using the definition for biofilm mass density ($C_x = \epsilon_f = X''/L_p$), Equation 28 becomes:

$$k_w = \frac{(k_d L_F) \left( \frac{X''}{L_F} \right)}{(C_x)_B - (k_d L_F) \left( \frac{X''}{L_F} \right) \left( \frac{1}{k_f} \right)}$$

$$= \frac{k_d X''}{(C_x)_B - \frac{k_d X''}{k_f}}$$  \hspace{1cm} (30)$$

The area-averaged concentration of attached cells, $X''$, is measured as a part of the experimental data collection and can also be calculated from the BAM modeled film thickness, using Equation 26.

**BAM/EPANET Comparison**

For the comparison of the BAM and EPANET programs, two pipes in series were simulated with each program. Each pipe had a two hour residence time and a velocity
of 1 ft/s. The EPANET model using English units, was created with pipes having a 4" diameter, 7200 ft length and a flow rate of 39 gpm.

The base BAM model was modified to represent the two pipes in series as two identical units, without recycling. Each pipe was represented as a BAM unit with a surface area of 698.8 m² and a volume of 17.75 m³, to correspond to the 4" diameter and 7,200 ft length of the EPANET model pipes. The flow rate was set at 213 m³/d (39 gpm) to create a 1 ft/s velocity and 2 hour residence time in each pipe.

Typical BAM model results were used to calculate $k_w$ from Equation 30, using Equation 26 to convert the modeled $L_F$ to $X''$. For the EPANET simulation, $k_w$ was adjusted to yield a HPC concentration identical to the BAM model results. The required $k_w$ to produce a similar HPC concentration in EPANET was then compared to the BAM value. In addition to $k_w$, the diffusivity and the bulk fluid reaction rate coefficient, $k_B$, were also varied in the EPANET input file to evaluate their effect on the bulk fluid concentrations. The comparison of bulk fluid concentrations was made using the values at the end of the EPANET pipes and the concentrations in the completely mixed BAM units.
CHAPTER 5

RESULTS AND DISCUSSION

Objective I - BAM Model Calibration

A preliminary model was created using the known physical configuration of the pipe loop and estimated values of the parameters which characterize the chemical and biological processes occurring within a biofilm system. The model was calibrated by adjusting these various unknown input parameters to obtain results that matched the experimental data for HPC bacteria counts, AOC concentrations and estimated biofilm thicknesses.

Table 3 summarizes the values of the input parameters used in the calibrated BAM model. To assess the BAM model, literature review was performed to determine expected ranges for the unknown parameters. Table 4 is a comparison of the unknown BAM values to the possible ranges as determined from review of studies concerning microbial activity in drinking water environments.

The model results are demonstrated in Figures 6-8 which show the consumption of AOC, the development of a biofilm on the pipe wall and microbial regrowth for a typical BAM simulation. The range of the bulk fluid AOC concentrations and HPC bacterial populations measured during pipe loop experiments 1 and 2 are indicated on the graphs for comparison of the model and actual results.
<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>VALUE</th>
<th>NOTES</th>
</tr>
</thead>
<tbody>
<tr>
<td>Surface area of unit (A_f)</td>
<td>3.89 m²</td>
<td>4&quot; φ pipe, 40' long</td>
</tr>
<tr>
<td>Volume of unit (V_b)</td>
<td>.0988 m³</td>
<td>S.A./V = 12 ft⁻¹</td>
</tr>
<tr>
<td>Influent flow rate (Q_{in})</td>
<td>3.67 m³/d</td>
<td>306 L/2 hrs</td>
</tr>
<tr>
<td>Total flow rate (Q)</td>
<td>213 m³/d</td>
<td>1 ft/s in a 4&quot; pipe</td>
</tr>
<tr>
<td>Particulate dry density (ρ_x)</td>
<td>1.0e5 g/m³</td>
<td></td>
</tr>
<tr>
<td>Solid phase volume fraction (ε_x)</td>
<td>.025</td>
<td></td>
</tr>
<tr>
<td>Diffusivity ratio (f)</td>
<td>.8</td>
<td></td>
</tr>
<tr>
<td>Substrate diffusivity (D)</td>
<td>8.84e⁻⁵ m²/d</td>
<td></td>
</tr>
<tr>
<td>Liquid layer thickness (δ_{ll})</td>
<td>10 μm</td>
<td></td>
</tr>
<tr>
<td>Maximum specific growth rate (μ_m)</td>
<td>2.88 d⁻¹</td>
<td>.12 hr⁻¹</td>
</tr>
<tr>
<td>Yield (Y)</td>
<td>.005 g dry cells/g C</td>
<td>2.6e8 cells/mg C</td>
</tr>
<tr>
<td>Half-saturation constant (K_s)</td>
<td>.05 g/m³</td>
<td></td>
</tr>
<tr>
<td>Decay coefficient (b)</td>
<td>.5 d⁻¹</td>
<td></td>
</tr>
<tr>
<td>Detachment rate (u_{dc})</td>
<td>k_d*L_F</td>
<td></td>
</tr>
</tbody>
</table>

**INITIAL CONDITIONS**

<p>| Film thickness (L_p)                  | .05 μm  |                             |
| Inlet and bulk AOC (C_s)_{B₀} and (C_s)<em>B | .5 g/m³ | Loops with AOC addition.    |
|                                       |         | Background AOC: .02-.09 g/m³ |
| Inlet and bulk HPCs (C_X)</em>{B₀} and (C_X)_B | .0001 g/m³ | Based on measured bacterial counts |</p>
<table>
<thead>
<tr>
<th>Parameter</th>
<th>Range of Reported Values</th>
<th>BAM Model Value</th>
<th>Reference Numbers</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_s$</td>
<td>.01-.24 g/m$^3$</td>
<td>.05 g/m$^3$</td>
<td>4,7,19,20</td>
</tr>
<tr>
<td>$\mu_m$</td>
<td>.09-.38 hr$^{-1}$</td>
<td>.12 hr$^{-1}$</td>
<td>7,19,20</td>
</tr>
<tr>
<td>$Y$</td>
<td>$2.4\times10^8$-$1\times10^{10}$ CFU/mg C</td>
<td>$2.6\times10^8$ CFU/mg C</td>
<td>19,20</td>
</tr>
<tr>
<td>$D$</td>
<td>$1.4\times10^{-6}$-$1.4\times10^{-5}$ cm$^2$/sec</td>
<td>$1.0\times10^{-5}$ cm$^2$/sec</td>
<td>23</td>
</tr>
<tr>
<td>$f$</td>
<td>.5-.9</td>
<td>.8</td>
<td>9,23</td>
</tr>
<tr>
<td>$b$</td>
<td>.01-.08 hr$^{-1}$</td>
<td>.02 hr$^{-1}$</td>
<td>4</td>
</tr>
<tr>
<td>$k_d$</td>
<td>.001-.1 hr$^{-1}$</td>
<td>.004-.02 hr$^{-1}$</td>
<td>6</td>
</tr>
<tr>
<td>$\mu$</td>
<td>.001-.1 hr$^{-1}$</td>
<td>.03-.05 hr$^{-1}$</td>
<td>6</td>
</tr>
<tr>
<td>$\delta_{LL}$</td>
<td>50-100 $\mu$m</td>
<td>10 $\mu$m</td>
<td>9</td>
</tr>
<tr>
<td>$e_s$</td>
<td>.01-.13</td>
<td>.025</td>
<td>9</td>
</tr>
</tbody>
</table>
FIGURE 6
SIMULATED AOC VS TIME

ACTUAL: .014–.046 mg/L
FIGURE 7
SIMULATED BIOFILM THICKNESS VS. TIME

TIME (DAYS)

0 1 2 3 4 5 6 7 8

FLM THICKNESS (um)

0 0.05 0.1 0.15 0.2 0.25 0.3 0.35 0.4

ACTUAL: .14-.37 um
FIGURE 8
SIMULATED BULK HPCs VS. TIME

ACTUAL: 4.3E4 - 1.1 E5
Objective II - Identify Key Processes for BAM Model of Regrowth

Bulk Fluid Concentration Changes

The BAM modeling equations were analyzed to compare the importance of detachment and bulk solution growth to an increase of bacteria in the bulk fluid. The change in bulk fluid cell concentration is calculated from equation 11, the mass balance equation:

$$\frac{d(V_B \times (C)_B)}{dt} = Q \times ((C)_B - (C)_B) + A_F \times j_B + V_B \times r_B$$ (11)

Analysis of the mass balance equation showed that the mass flux from the biofilm to the bulk fluid is much more significant than the net growth of cells within the bulk fluid:

$$A_F \times j_B > V_B \times r_B$$

The mass flux and transformation rate were compared by substituting the equations for the $A_F$, $V_B$, $r_B$, and $j_B$ terms. At the BAM steady-state conditions, the biofilm thickness is constant, and therefore the mass flux equals the detachment flux. Equation 10 becomes:

$$\frac{d(L_f)}{dt} = u_F - u_{de} = 0$$ (32)
Using equation 8, the mass flux is written as:

$$ j_x = u_F \cdot (C_x)_F = u_{de} \cdot (C_x)_F $$  \hspace{1cm} (33) 

Consequently the comparison reduces to:

$$ A_F \cdot (u_{de} \cdot (C_x)_F) \hspace{0.5cm} VS. \hspace{0.5cm} V_B \cdot ((\mu - b) \cdot (C_x)_B) $$

Using the values of the various terms obtained from the BAM modeling results,

$$ A_F \cdot j_x = 1.52 \cdot 10^{-3} \text{g/day} $$
$$ V_B \cdot r_{xB} = 9.56 \cdot 10^{-5} \text{g/day} $$

This comparison shows that the detachment process contributes approximately 15 times more to microbial regrowth than the net growth in the bulk fluid.

**Detachment Velocity**

Detachment velocity is an important parameter that was determined through the BAM modeling process. BAM can model detachment in a variety of ways: as a function of biofilm thickness ($L_f$), biofilm thickness squared ($L_f^2$), growth rate, or concentration of microorganisms. The detachment velocity can also be equal to a constant. During the model calibration process, all of these methods were evaluated as the detachment velocity was adjusted until a thin biofilm (under 1 μm) was developed and maintained under conditions identical to the pipe loop experiments.
Initial results indicated that detachment velocity \( (u_{de}) \) could be modeled with any of the following equations, with the same resulting film thickness and bulk fluid microorganism concentration.

\[
\begin{align*}
  u_{de} &= c_1 \quad (34) \\
  u_{de} &= c_2 L_F \quad (35) \\
  u_{de} &= c_3 L_F^2 \quad (36)
\end{align*}
\]

When the BAM model was used to duplicate the results from the pipe loop Experiments 1 and 2, a detachment velocity of \( 1 \times 10^7 \) m/d was used. Each of the three methods for representing \( u_{de} \) yielded identical results, which led to the conclusion that \( u_{de} \) was actually a constant value.

However, when detachment velocities were calculated for all of the data (Experiments 1-4), using \( u_{de} = k_d L_F \), the range of velocities was \( 1.29 \times 10^8 \) to \( 1.22 \times 10^6 \) m/d. Although the \( u_{de} \) value of \( 1 \times 10^7 \) m/d could be valid, attempts were made to vary \( u_{de} \) and then make a comparison of the model results with the experimental data.

When detachment velocity was represented as a constant (Equation 34), the model would not reach a steady state at velocities greater than \( 1 \times 10^7 \) m/d. However, Equations 35 and 36 were successfully used to model the pipe loop experiments for detachment rates greater than \( 1 \times 10^7 \) m/d. The three methods of representing \( u_{de} \) were evaluated by adjusting the constant used in each equation in order to maintain the same bulk effluent HPC bacteria concentration. The effect on the model was assessed by comparing the resulting film thickness and substrate concentration. These comparisons
are made in Figures 9 and 10, which show the problem that occurs when a constant value of $u_{de}$ is used.

**FIGURE 9**
EFFECT OF VARIATIONS IN CONSTANT $U_{de}$

![Figure 9](image-url)
FIGURE 10
EFFECT OF VARIATIONS IN Ude EQUATIONS

\[ Ude = 1.1 \times 10^6 \times LF^2 \]
The behavior of the model confirmed that $u_{de}$ is not a constant, but a function of $L_F$. Although either Equation 35 or 36 could be used with essentially identical results, Equation 35 was used for subsequent BAM modeling.

Representing the detachment velocity as $u_{de} = c_2 * L_F$ allows the BAM constant $c_2$, to be compared to $k_d$, the detachment rate coefficient determined from the experimental results. The methods of calculating $k_d$ from the data were first presented in the AWWARF project *Factors Limiting Microbial Growth in the Distribution System - Quarterly Report 6* and are summarized in Appendix B.

**Sensitivity Analysis**

One result of the pipe loop model development was an awareness of the relationships between the various BAM input parameters. A summary of the effect that changing a variable had on the model results is contained in Table 5. As noted in Table 5, the importance of the individual parameters to the overall model accuracy was variable. The key parameters noted in Table 5 are the ones that had the most significant effect on the model results.

**Additional BAM Modeling Results**

Once the BAM base model was developed and calibrated, the influent concentrations of AOC and microorganisms were varied to evaluate their effect on the model results.

The results from varying the influent substrate concentration, the influent heterotrophic bacteria concentration and the detachment rate are summarized below:
### TABLE 5
SENSITIVITY ANALYSIS OF BAM INPUT PARAMETERS

<table>
<thead>
<tr>
<th>Input parameter</th>
<th>Effect of decreasing parameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\mu_m$</td>
<td>$(C_s)_B$ increases, $L_f$ and $(C_X)_B$ decrease</td>
</tr>
<tr>
<td>$K_s$</td>
<td>$(C_s)_B$ decreases, $L_f$ and $(C_X)_B$ increase slightly</td>
</tr>
<tr>
<td>$b$</td>
<td>$L_f$ increases, $(C_s)_B$ decreases, $(C_X)_B$ decreases</td>
</tr>
<tr>
<td>$Y$</td>
<td>$L_f$ and $(C_X)_B$ decrease</td>
</tr>
<tr>
<td>$\delta_{LL}$</td>
<td>$L_f$ increases, $(C_s)_B$ decreases</td>
</tr>
<tr>
<td>$D$</td>
<td>$L_f$ decreases, $(C_s)_B$ and $(C_X)_B$ increase</td>
</tr>
<tr>
<td>$f$</td>
<td>No effect</td>
</tr>
<tr>
<td>$k_d$</td>
<td>$(C_X)_B$ decreases, $L_f$ increases</td>
</tr>
</tbody>
</table>

Key parameters: $k_d$, $\delta_{LL}$, $Y$, $\mu_m$

1. Variations in the influent substrate concentrations, to the degree observed in the experiments, did not significantly affect the BAM model results.

2. Variations in the influent bacteria concentrations had virtually no effect on the BAM model results.

3. Variations in the detachment coefficient $k_d$ did not significantly affect bulk substrate concentrations, but did affect bulk fluid bacteria concentrations and the film thickness.

These observations agree with the conclusions reached from the analysis of the modeling equations - that the most significant process affecting bulk fluid concentrations is detachment of cells from the biofilm into the bulk fluid.
Modeling Coliform Bacterial Populations

The BAM model of the processes occurring in a drinking water pipeline was developed using heterotrophic bacteria as the particulate species. The HPC data were used since the measured CFUs in the bulk fluid and in the pipe wall biofilm are approximately 10,000 times greater than the coliform populations.

After the BAM model was developed for HPCs, efforts to model the coliform bacteria were undertaken. The BAM program was not capable of modeling both heterotrophs and coliforms simultaneously, so a separate model for the coliforms was created. The original BAM model of the heterotrophs was used as a base model and the changes were made to simulate the growth, decay and detachment of coliforms within the pipe loop.

The solid phase fraction ($e_s$) was reduced to reflect the minute fraction of coliforms in the biofilm. Since the concentration of coliforms in the biofilm is approximately 10,000 times lower than the heterotroph population, $e_s$ for coliforms was set at $2.5 \times 10^{-7}$ ($0.025/10,000$). The bulk substrate concentration was fixed as $0.04$ mg/L to reflect the remaining AOC after the heterotrophs consumed the bulk of the substrate. The initial film thickness was set at $0.4 \mu m$, the resulting thickness when the BAM model performs a typical run. The initial bulk coliform concentration was set at $0.002$ mg/L, to represent the inoculation of the pipe loop with $10,000$ CFU/mL of coliforms at the beginning of each experiment. The influent coliform concentration was zero since coliforms have not been detected in the pipe loop influent water.
With these changes made, the model was evaluated to determine what additional modifications would be necessary to keep the film thickness essentially constant and to reduce the bulk coliform concentration to below 10 CFU/mL, (the average experimentally measured quantity). Initially, the same kinetic and stoichiometric parameters used in the heterotrophic BAM model were used for the coliform BAM model. Under this scenario, the detachment rate coefficient had to be increased from .4 to .83 d⁻¹.

Alternatively, the detachment rate coefficient was kept constant at .4 d⁻¹ and necessary changes to the kinetic and/or stoichiometric parameters were assessed. When the detachment rate coefficient was unchanged, the maximum specific growth rate \( \mu_m \), had to be changed from 2.88 d⁻¹ to 1.95 d⁻¹ to maintain a constant film thickness.

Although the coliform populations were modeled using the BAM program, the accuracy of the model is doubtful. The measured coliform populations were generally very small and observed increases in the coliform CFU counts could not be predicted by the model.

**kₚ Calculations**

With the measured bacterial counts on the biofilm coupons and calculated values of \( k_d \), \( k_p \) values were computed for the experimental data. These values were compared to \( k_p \) values calculated from BAM model results to confirm that the two \( k_p \) calculation methods agreed. Table 6 summarizes the results of this comparison for various data sets.
 Objective III - BAM and EPANET Evaluation

The Biofilm Accumulation Model developed from experimental data was used to make an evaluation of the capability of the EPANET program for modeling microbial growth.

For the comparison of the BAM and EPANET programs, two pipes in series were simulated with each program. Each pipe had a two hour residence time and a velocity of 1 ft/s. The BAM model results were used to calculate the wall reaction rate constant $k_w$ from the modeled HPC bacteria concentration and film thickness, as discussed in Chapter 4. The value of $k_w$ that was required to achieve similar HPC bacteria concentrations in EPANET was then compared to the value predicted from the BAM program.
The results of the BAM and EPANET simulations are summarized in Table 7 and Table 8.

### TABLE 7
**BAM RESULTS**

<table>
<thead>
<tr>
<th></th>
<th>UNIT 1</th>
<th>UNIT 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>$(C_b)_x$ (mg/L)</td>
<td>.00117</td>
<td>.0012</td>
</tr>
<tr>
<td>$L_F$ (μm)</td>
<td>.305</td>
<td>.01</td>
</tr>
<tr>
<td>$k_w$ (ft/day)</td>
<td>.58</td>
<td>.016</td>
</tr>
<tr>
<td>$(C_b)_s$ (mg/L)</td>
<td>.0381</td>
<td>.0142</td>
</tr>
</tbody>
</table>

### TABLE 8
**EPANET RESULTS**

<table>
<thead>
<tr>
<th>RUN NO.</th>
<th>D (ft²/sec)</th>
<th>$k_b$ (1/day)</th>
<th>$k_w$ (ft/day)</th>
<th>$(C_b)_x$ (mg/L)</th>
<th>$(C_b)_s$ (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.64x10⁻⁸</td>
<td>0</td>
<td>2.80</td>
<td>.00035</td>
<td>2.80</td>
</tr>
<tr>
<td>2</td>
<td>1.64x10⁻⁸</td>
<td>0</td>
<td>280</td>
<td>.00102</td>
<td>280</td>
</tr>
<tr>
<td>3</td>
<td>1.64x10⁻⁸</td>
<td>0</td>
<td>28000</td>
<td>.00104</td>
<td>28000</td>
</tr>
<tr>
<td>4</td>
<td>1</td>
<td>0</td>
<td>1.72</td>
<td>.00035</td>
<td>1.72</td>
</tr>
<tr>
<td>5</td>
<td>1</td>
<td>0</td>
<td>4.85</td>
<td>.00117</td>
<td>4.85</td>
</tr>
<tr>
<td>6</td>
<td>1</td>
<td>0</td>
<td>4.85</td>
<td>.00117</td>
<td>.02</td>
</tr>
<tr>
<td>7</td>
<td>1</td>
<td>1.323</td>
<td>0</td>
<td>.00011</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>1</td>
<td>1.323</td>
<td>4.72</td>
<td>.00117</td>
<td>.02</td>
</tr>
</tbody>
</table>

For the EPANET modeling, the diffusivity and the wall reaction rate were modified to obtain different bulk fluid concentrations. The results show that although EPANET can simulate the detachment of microorganisms from the pipe wall, there are
significant problems with the modeling techniques when used for the application of simulating microbial regrowth.

The discrepancies between the EPANET and the BAM models which occurred although the same reaction rate coefficients were used may be related to the different hydraulic calculation methods. BAM considers each unit as a completely mixed reactor while EPANET divides each pipe into a series of mixed volumes. Since the EPANET approach is actually a representation of plug flow, the calculated concentrations may vary significantly from those obtained in a completely mixed system.

The following conclusions were made from a comparison of the EPANET model behavior to the BAM results.

1. The reaction rate constant, $K_2 = k_w \cdot k_f / (R_H \cdot (k_w + k_p))$, calculated to model mass transfer from the pipe wall to the bulk fluid, reaches a maximum value, determined by the value of the mass transfer coefficient $k_f$. As $K_2$ approaches a maximum, it is insensitive to increases in $k_w$. The BAM concentration of .0012 mg/L at a 4 hour residence time could be matched by EPANET, but increasing $k_w$ by 10,000 times (runs 2 & 3) was not effective in modeling the BAM concentration in the first unit. The only way of obtaining a concentration change from .0001 mg/L to .00117 mg/L in a 2 hour residence time (pipe 1) was to increase the mass transfer by specifying a very high diffusivity value (1 ft$^2$/sec).

2. Although EPANET and BAM $k_w$ values do not agree, they show the same trend of decreasing values for successive pipes. BAM results show that as substrate is consumed, biofilm growth decreases along a pipeline, resulting in less regrowth
and smaller $k_w$ values. To correspond with the BAM concentrations, the EPANET wall reaction rate constant must be changed in each pipe.

3. Referring to runs 7 and 8, bulk fluid growth is insubstantial compared to mass transfer from the wall. When $k_b$ was set equal to the growth rate $\mu$ determined from the BAM results, it accounted for the production of only .00001 mg/L of microorganisms in a 2 hour residence time.

The EPANET program has potential as a tool to accurately model the transport and fate of microorganisms within a distribution system. However, the equations used for simulating the complex processes occurring within a pipeline must be further developed. Although the water quality equation includes reactions in the bulk fluid and at the pipe wall, the reaction rates in the bulk fluid (growth and decay) are insignificant compared to the detachment process.

EPANET can model mass transport between the pipe wall and the bulk fluid but the modeling techniques are not representative of the true detachment process. As determined from the BAM simulations, detachment rate is a function of film thickness, which in turn depends on the available substrate concentration. The EPANET capabilities should be expanded to allow the simulation of more than one component and the relationships between different components.

Modeling Substrate

To simulate the regrowth of microorganisms, a computer program needs to concurrently model substrate and microorganisms. Regrowth is the result of detachment
of cells from the biofilm, which depends, indirectly, on the substrate concentration. From a mass balance on cells in the biofilm:

Net growth rate - Net detachment rate = Rate of accumulation

As discussed in Chapter 3, the net detachment rate, \( r_{de}'' \), accounts for the individual processes of attachment and detachment. Cell growth and decay are also combined in the overall transformation rate expressed per area of biofilm, \( r_x'' \). At steady state, the mass balance equation is:

\[
\frac{\partial n}{\partial t} = 0
\]

From the definition of \( u_{de} \),

\[
(\mu - b) * X'' = u_{de} * (C_x)_F
\]

(38)

From the definition of \( u_{de} \),

\[
(\mu - b) * X'' = k_d * L_F * (C_x)_F
\]

(39)

Since \( X'' = L_F * (C_x)_F \), equation 10 reduces to:

\[
\mu - b = k_d
\]

(40)

This equation shows that the detachment rate coefficient \( k_d \) is directly related to the growth rate \( \mu \) of cells in the biofilm. Since Monod kinetics were used to define the cell growth rate, with growth as a function of available substrate, detachment depends on the substrate concentration within the biofilm. The BAM model includes the option of expressing the detachment rate formula as a function of growth rate. However, this method of representing the detachment rate resulted in inaccurate results during the
model development phase, which may have occurred since the equation does not use the
net growth rate ($\mu - b$).

The concentration of substrate in a biofilm system depends on transport and
transformation processes. Table 9 is a comparison of the equations used by EPANET
and BAM to model the net change in substrate concentration over time. The EPANET
equations for substrate consumption are the same form as for the growth of a substance,
except with negative signs.

**Transformation Process**

The EPANET equation for the transformation rate is incorrect for modeling
substrate consumption since it has no dependence on cell growth rate. It is well
established that cell growth and substrate consumption rates are related. The relationship

| TABLE 9  |
|-------------------------|-------------------------|
| Model Equations for dS/dt |
|-------------------------|-------------------------|
| **Transformation rate** | **BAM**                  | **EPANET**              |
|                         | $\left( \frac{\mu_m \cdot (C_s)_B}{K_s + (C_s)_B} \right) Y_{X/L} \cdot (C_x)_B$ | $-k_f \cdot (C_s)_B$ |
| **Transport rate**      | $j_s \cdot \left( \frac{A_F}{V_B} \right) = \frac{-f \cdot D \cdot \frac{\partial C_s}{\partial z}}{R_H}$ | $-\frac{k_f}{R_H} \cdot (C_s)_B - (C_s)_F$ |
|                         | $= -\frac{k_f}{R_H} \cdot (C_s)_B - (C_s)_F$ | |


between substrate consumption and biomass production is represented by the yield \((Y)\), the ratio of cell growth and substrate removal rates. EPANET should model substrate consumption using the same equation as BAM \((r_s = - r_x/Y)\).

**Transport Process**

BAM calculates the substrate flux using the same mass transport equation as EPANET. However, the two methods for calculating the mass transfer coefficient, \(k_f\), are different, as summarized in Table 10. The EPANET program calculates the mass transfer coefficient using the Sherwood Number while the BAM program calculates \(k_f\) using the user specified thickness of the boundary layer between the biofilm and the bulk fluid, the liquid layer thickness \((\delta_{LL})\).

<table>
<thead>
<tr>
<th>TABLE 10</th>
<th>COMPARISON OF MASS TRANSFER MODELS</th>
</tr>
</thead>
<tbody>
<tr>
<td>(k_f) formula</td>
<td>BAM</td>
</tr>
<tr>
<td>(k_f = \frac{f \times D}{\delta_{LL}})</td>
<td></td>
</tr>
<tr>
<td>calculated (k_f) value</td>
<td>22.75 ft/day</td>
</tr>
<tr>
<td>corresponding (\delta_{LL})</td>
<td>10 (\mu)m</td>
</tr>
</tbody>
</table>

where \(Sh\) = Sherwood Number = \(0.023 \times Re^{.83} \times Sc^{.33}\) for \(Re \geq 2300\)  
\(Re\) = Reynolds Number = \(Q \times d \div (A \times v) = 30555\) for the modeled pipe  
\(Sc\) = Schmidt Number = \(\tau \div D\)  
\(\tau\) = fluid kinematic viscosity = \(1.05 \times 10^{-5}\) ft\(^2\)/s (water at 20°C)
Although both of these methods are valid, it should be noted that the BAM $k_f$ is not a function of velocity. The EPANET calculation does consider fluid velocity, through the Sherwood and Reynolds numbers. However, several versions of the formula used to calculate the Sherwood number have been developed from experimental investigations. Different coefficients used in these empirical formulas account for variations in the pipe roughness. A useful modification to EPANET would be an incorporation of a change in the mass transfer coefficient with different pipe materials.

The EPANET mass transfer coefficient corresponds to a liquid boundary layer thickness of 80 $\mu$m, which is significantly larger than the thickness determined during the BAM model development process. If the BAM $\delta_{LL}$ was increased to 80 $\mu$m, the simulated bulk fluid substrate concentration does not agree with measured values, even if the kinetic parameters are modified. Changes in the kinetic and stoichiometric coefficients had insignificant effects on bulk fluid substrate concentration which indicates that the model is mass transport limited when the EPANET liquid layer thickness of 80 $\mu$m is used.

To confirm the mass transport limitation, the limiting regime for substrate consumption was calculated following the methods presented in *Biological Wastewater Treatment* (Grady and Lim, 1980). The limiting regime is determined by the Damköhler number, $D_a$, the ratio of the substrate removal rate at the biofilm to the maximum possible substrate transfer rate across the liquid boundary layer. When $D_a$ is greater than 1 the substrate removal rate exceeds the transfer rate and the situation is transport
limited. For $D_a$ less than 1, the conditions are reaction limited since the transfer rate exceeds the substrate removal rate.

The Damköhler number is written mathematically as:

$$D_a = \frac{q''_m}{k_f * (C_g)_B}$$  \hspace{1cm} (38)

$D_a$ was calculated using the different $k_f$ values determined for the two models. The kinetic parameters used in the BAM input model file were used to calculate $q''_m$:

$$q''_m = \frac{\mu_m * X''}{Y} = \frac{2.88d^{-1}}{.005mg/mg} * (2500g/m^3 * .305\mu m)$$ \hspace{1cm} (39)

$$= .4392 \frac{g}{m^2*day}$$

For the EPANET model:

$$D_a = \frac{.4392 \frac{g}{m^2*day}}{2.9 \frac{ft}{day} * .0381mg/L} = 13.04$$

For the BAM model:

$$D_a = \frac{.4392 \frac{g}{m^2*day}}{22.75 \frac{ft}{day} * .0381mg/L} = 1.66$$
From Figure 14.2 in Grady and Lim, at $K_r/S_b = 1.3$ (included as Appendix C) the substrate removal rate is limited by mass transfer effects when the EPANET $k_r$ value is used. Although the BAM model results also indicate transport limitation, slight changes in the kinetic parameters used to calculate $D_a$ would change the regime to reaction limited.

Although the equations used by the two models to simulate consumption of substrate differ, as a final analysis of EPANET, model runs were performed to compare the resulting substrate concentrations. The results of the EPANET simulations, which show agreement with the BAM results (refer to Table 8), are summarized in Table 11.

<table>
<thead>
<tr>
<th>RUN NO.</th>
<th>$k_b$ (1/day)</th>
<th>$k_w$ (1/day)</th>
<th>PIPE 1 ($C_{sb}$) (µg/L)</th>
<th>PIPE 2 ($C_{sb}$) (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0</td>
<td>-2.6</td>
<td>39.8</td>
<td>3.2</td>
</tr>
<tr>
<td>2</td>
<td>-17.2</td>
<td>0</td>
<td>40.1</td>
<td>3.2</td>
</tr>
<tr>
<td>3</td>
<td>-8</td>
<td>-1</td>
<td>40.8</td>
<td>3.3</td>
</tr>
</tbody>
</table>

Although both models show a decrease in substrate concentration with successive pipes, this trend does not agree with laboratory and field observations. This is demonstrated by Table 12, developed from the experimental results for pipe loops with varying residence times. The discrepancy between measured and simulated substrate concentrations with increasing residence time should be further investigated to determine possible sources of error.
### TABLE 12
**EXPERIMENT 1**
**BULK FLUID SUBSTRATE CONCENTRATIONS**

<table>
<thead>
<tr>
<th>LOOP</th>
<th>AVERAGE AOC (µg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 - 2 hr. residence time</td>
<td>23.5</td>
</tr>
<tr>
<td>2 - 2 hr. residence time</td>
<td>23.0</td>
</tr>
<tr>
<td>3 - 4 hr. residence time</td>
<td>19.5</td>
</tr>
<tr>
<td>4 - 8 hr. residence time</td>
<td>44.1</td>
</tr>
<tr>
<td>5 - 16 hr. residence time</td>
<td>76.2</td>
</tr>
</tbody>
</table>
CHAPTER 6

CONCLUSIONS

Data from pilot scale pipe loop experiments was used to develop a BAM model of the biofilm system present in a drinking water pipe. The results of this process were employed to evaluate the water quality module of the EPANET program for simulating bacterial regrowth. This investigation has served to distinguish both the capabilities and limitations of the BAM and EPANET programs. As a result, the following conclusions are made:

1. Although the BAM program requires numerous input terms, many of them do not have a significant influence on the simulation results when modeling the conditions of a water distribution pipeline. The key parameters for the model of a biofilm system in a drinking water pipe were the yield, detachment rate coefficient, liquid layer thickness and maximum specific growth rate.

2. Detachment of cells from the biofilm into the bulk fluid is the most significant process resulting in bulk fluid bacterial population increases, and is first order function of the film thickness.
3. Rate of regrowth is a function of the substrate concentration since the biofilm growth and subsequent detachment depend on the available substrate.

4. The BAM program was successfully used to duplicate pilot scale experimental results for HPC bacteria populations and to determine values of the unknown kinetic parameters. The accuracy of the model should be confirmed through field data.

5. EPANET does not accurately simulate regrowth since it does not account for the substrate limitation to microbial growth.

6. Further development of the EPANET model for the simulations of bacterial populations should include the following improvements:
   - The ability to simulate substrate and bacterial populations concurrently.
   - Development of more comprehensive equations for modeling reaction rates.
   - Modifications to the calculation methods for the mass transfer coefficient.
NOMENCLATURE

Definitions

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>$A_F$</td>
<td>area in the unit covered by biofilm</td>
</tr>
<tr>
<td>$A$</td>
<td>cross-sectional area = $\pi d^2/4$</td>
</tr>
<tr>
<td>$b$</td>
<td>respiration rate coefficient</td>
</tr>
<tr>
<td>$C$</td>
<td>concentration</td>
</tr>
<tr>
<td>$d$</td>
<td>diameter</td>
</tr>
<tr>
<td>$D$</td>
<td>diffusivity in pure water</td>
</tr>
<tr>
<td>$f$</td>
<td>ratio of the diffusivities in the biofilm and in pure water</td>
</tr>
<tr>
<td>$E$</td>
<td>elevation</td>
</tr>
<tr>
<td>$\varepsilon$</td>
<td>local volume fraction of the total biofilm volume</td>
</tr>
<tr>
<td>$h$</td>
<td>hydraulic grade line elevation</td>
</tr>
<tr>
<td>$J$</td>
<td>mass flux per unit total cross-sectional area</td>
</tr>
<tr>
<td>$k_B$</td>
<td>first-order bulk reaction rate constant</td>
</tr>
<tr>
<td>$k_d$</td>
<td>detachment coefficient</td>
</tr>
<tr>
<td>$k_f$</td>
<td>mass transfer coefficient</td>
</tr>
<tr>
<td>$K_S$</td>
<td>half-saturation constant</td>
</tr>
<tr>
<td>$k_w$</td>
<td>wall reaction rate constant</td>
</tr>
<tr>
<td>$L_F$</td>
<td>biofilm thickness</td>
</tr>
<tr>
<td>$L_p$</td>
<td>pipe length</td>
</tr>
</tbody>
</table>
\( \delta_{LL} \)  liquid layer thickness

\( \mu \)  specific growth rate

\( \mu_m \)  maximum specific growth rate

\( \varrho_x \)  density of cells

\( \varrho_F \)  biofilm mass density = \( \varrho_x \epsilon_s = (C_x)_F = X''/L_F \)

\( Q \)  volumetric flow rate = \( A^*v = V/t \)

\( r \)  transformation rate of component per unit total volume

\( r'' \)  transformation rate per unit total cross-sectional area

\( R_H \)  hydraulic radius of pipe \( d/4 \)

\( \text{Re} \)  Reynolds Number

\( \text{Sc} \)  Schmidt Number

\( \text{Sh} \)  Sherwood Number

\( u_i \)  velocity of the interface relative to the fixed coordinate \( z \)

\( u_r \)  velocity by which particulate mass is displaced relative to the film surface

\( u_L \)  velocity biofilm surface is displaced relative to the solid surface

\( u_{de} \)  detachment velocity

\( v \)  fluid velocity = \( Q/A \)

\( \tau \)  fluid kinematic viscosity

\( V_B \)  bulk fluid volume

\( X'' \)  area-averaged concentration of biofilm cells

\( y \)  height of water

\( Y \)  Yield coefficient
### Key Subscripts

<table>
<thead>
<tr>
<th>Subscript</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>B</td>
<td>bulk fluid</td>
</tr>
<tr>
<td>F</td>
<td>biofilm</td>
</tr>
<tr>
<td>de</td>
<td>detachment</td>
</tr>
<tr>
<td>I</td>
<td>liquid</td>
</tr>
<tr>
<td>o</td>
<td>influent</td>
</tr>
<tr>
<td>s</td>
<td>solid</td>
</tr>
<tr>
<td>S</td>
<td>substrate</td>
</tr>
<tr>
<td>X</td>
<td>microorganisms</td>
</tr>
</tbody>
</table>
REFERENCES


Notes: Submitted to Water Research for review March, 1994


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Appendix A

BIOSIM Assumptions and Simplified Equations
Assumptions Made in the Biofilm Model

1. The biofilm consists of one liquid phase and several solid phases, of which each is constituted by the mass of one particulate component.

2. A solid phase carries no dissolved or suspended particulate components.

3. Advective transport of the solid phases is the result of changes in the volume of the constituent particulate components.

4. The advective velocities of $u_p$ of all solid phases are equal.

5. The density of the particulate components is constant: $\rho = \text{constant}$. 

6. The liquid phase carries only dissolved components.

7. Transport of the dissolved components in the liquid phase is due to molecular diffusion.

8. The dissolved components in the liquid phase are electrically neutral and show ideal behavior: Fick’s first law of molecular diffusion applies.

9. The ratio of the diffusivities in the biofilm and in pure water is constant: $\frac{D_f}{D} = \text{constant}$. 

10. The volume fraction of the liquid phase is constant: $\epsilon_L = \text{constant}$. 

11. Transport and gradients of system properties in directions other than $z$ are negligibly small: A model that is one-dimensional in space applies.
2. THE BIOFILM MODEL

2.1 Model Assumptions

The mixed culture biofilm model used in BIOSIM is a mechanistic model (Wanner, 1989). It is based on the assumption that the continuum concept applies. This means that biofilm components, such as microbial species, are not described by the shape and size of their individual cells, but by averaging quantities such as volume fractions and concentrations. The model equations are derived from fundamental physical laws and principles. They represent a set of consistent hypotheses on the processes which take place inside a biofilm and are based on only a minimum number of a priori assumptions. A list of the assumptions made in the model is included in the Appendix. Some of these assumptions are definitions of the quantities used in the model and others are simplifications which reflect missing experimental data. The volume fraction of the liquid phase in the biofilm, e.g., is very likely to change with time and space, however, in practice it is usually assumed to be constant just because the needed data is not available.

2.2 Model Equations

The dynamics and spatial distribution of particulate components in the biofilm compartment is described by the one-dimensional mass balance equation (Gujer and Wanner, 1989)

\[
\frac{\partial C_x}{\partial t} = -\frac{\partial j_x}{\partial z} + r_x
\]

where \( C_x \) is the concentration of the particulate components per unit biofilm volume, \( j_x \) is the mass flux per unit biofilm volume in the direction perpendicular to the solid surface, \( r_x \) is the observed transformation rate, \( t \) is time, and \( z \) is the perpendicular distance from the solid surface. The concentration \( C_x \) relates to the volume fraction \( \varepsilon_s \) (\( \varepsilon_{ss} \)) of the biofilm solid phase which forms by a specific particulate component, by \( C_x = \rho \varepsilon_s \). The mass flux \( j_x \) originates from the production of particulate mass which leads to volume expansion and subsequent displacement of neighboring cells. Thus, \( j_x \) formally is an advective flux and is described by

\[
j_x = u_x C_x
\]

where \( u_x \) is the velocity by which particulate mass is displaced relative to the solid surface. The velocity \( u_x \) is calculated by

\[
u_x = \frac{1}{\rho} \int_0^{\varepsilon_{ss}} \frac{z \cdot r_x}{1 - \varepsilon_s} dz
\]

The boundary condition for equation (1) is

\[
j_x(z=0) = 0
\]
since no particulate mass can move into or out from the solid surface \((u_f(z=0) = 0)\). If attachment to the biofilm surface of cells or particles from the bulk fluid is greater than detachment of particulate components from the film surface, a second boundary condition is required for the solution of equation (1). By this boundary condition the concentration of the particulate components at the biofilm surface \((z=L_p)\) is specified as a function of the attachment rate \(r_{at}\) and attachment velocity \(u_{at}\) as

\[
C_X(z=L_p) = \frac{r_{at}}{u_{at}} \quad \text{required, if} \quad u_{at} > u_{dc}
\]

The detachment velocity \(u_{de}\) must be specified by the user as a constant or as a function of time \(t\), biofilm thickness \(L_f\) or bulk fluid velocity \(u_B\). Attachment is described as a first-order process, with the rate

\[
\frac{1}{r_{at}} = k_{at} C_{L,X}
\]

where \(C_{L,X}\) is the concentration of particulate component \(X\) in the liquid boundary layer at its interface to the film and \(k_{at}\) is the first-order rate coefficient. The attachment velocity is calculated as

\[
u_{at} = \frac{\sum \frac{r_{at}}{1-e_i X}}{\rho}
\]

The dynamics and spatial distribution of dissolved components in the biofilm compartment is described by the one-dimensional mass balance equation

\[
\varepsilon_i \frac{\partial C_s}{\partial t} = \frac{\partial j_s}{\partial z} + r_s
\]

where \(C_s\) is the dissolved component concentration, defined as mass per unit liquid phase volume, \(\varepsilon_i\) is the liquid phase volume fraction in the biofilm, \(j_s\) is the mass flux per unit biofilm volume in the direction perpendicular to the solid surface, \(r_s\) is the observed transformation rate, \(D\) is the diffusivity in pure water, and \(f\) is the ratio of the diffusivities in the biofilm and in pure water. The mass flux \(j_s\) is described by Fick's first law as

\[
j_s = -f D \frac{\partial C_s}{\partial z}
\]

The two boundary conditions required for the solution of equation (8) are the no-flux condition for the solid surface

\[
j_s(z=0) = 0
\]

and the continuity condition for the concentration at the biofilm surface
\[ C_S(z=L_F) = C_{L,S}(z=L_F) \]  

(11)

where \( C_{L,S} \) is the concentration of the dissolved component in the liquid boundary layer at its interface to the film.

The development of the biofilm thickness \( L_F \) is described by the ordinary differential equation

\[ \frac{dL_F}{dt} = u_L \]  

(12)

where the velocity \( u_L \) at which the biofilm surface is displaced relative to the solid surface is calculated by

\[ u_L = u_F(z=L_F) + u_{at} - u_{de} \]  

(13)

The bulk fluid compartment is assumed to be completely mixed. The dynamics of both the dissolved and the suspended particulate components in the bulk fluid is described by

\[ \frac{d(V_B C_B)}{dt} = Q_{in}(C_{in} - C_B) + A_F j_B + V_B r \]  

(14)

where \( C_B \) and \( C_{in} \) are the concentration of dissolved or suspended particulate components in the bulk fluid and in the influent, respectively, \( V_B \) is the volume of the bulk fluid, \( Q_{in} \) is the volumetric inflow rate, \( A_F \) is the area in the unit covered with biofilm, \( j_B \) is the mass flux from the biofilm to the bulk fluid per unit area of \( A_F \), and \( r \) is the net transformation rate.

In the liquid boundary layer transport is controlled by molecular diffusion. Biochemical transformation reactions are neglected and the concentration profiles of both the dissolved and the suspended particulate components are calculated for steady state.

Modeling a non steady-state situation requires that initial and boundary conditions are given. The former include values for the biofilm thickness \( L_F \), for the bulk fluid concentrations \( C_B \) and for the spatial profiles of the particulate components \( C_X \) at the initial time \( t_0 \) of the simulation (usually \( t_0=0 \)). The initial spatial profiles of the dissolved components \( C_S \) are calculated by the program.
Appendix B

AWWARF Project $k_d$ Calculation Methods
A simplified material balance on cells of a particular type (either HPC or coliforms) over the reactor yields the following:

\[
\frac{dX_b}{dt} = D(X_{b0} - X_b) + q_d a
\]

(1)

\[
\frac{dX_f}{dt} = r_g - q_d
\]

(2)

where

- \( a \) = specific surface area = surface area/volume
  - 2.73 cm\(^{-1}\) for the annular reactor
  - 0.393 cm\(^{-1}\) for the pipe loop
- \( D \) = dilution rate (.5 hr\(^{-1}\)) for 2 hr. residence time
- \( q_d \) = net detachment rate (cells cm\(^{-2}\) hr\(^{-1}\))
- \( r_g \) = net growth rate of attached cells (cells cm\(^{-2}\) hr\(^{-1}\))
- \( X_b \) = bulk fluid cells (cells cm\(^{-3}\)) in effluent
- \( X_{b0} \) = bulk fluid cells (cells cm\(^{-3}\)) in influent
- \( X_f \) = attached cells (cells cm\(^{-2}\))

The bulk cell balance (equation 1) assumes that negligible growth of suspended cells occurs in the reactor. Provided that the specific growth rate of cells in the system is less than 0.05 hr\(^{-1}\), this assumption is reasonable. The growth and attachment rates can be expressed as:

\[ q_d = k_d X_f \quad \text{and} \quad r_g = \mu_f X_f \]

(3)

where

- \( k_d \) = specific detachment rate (hr\(^{-1}\))
- \( \mu_f \) = specific growth rate (hr\(^{-1}\))

By expressing the time derivatives as finite differences and averaging measurements over each individual time difference, the following expressions are developed for net detachment rate, net growth rate, specific detachment rate and specific growth rate:
\[ q_{di} = \frac{1}{a} \left( \frac{X_{bi} - X_{bi-1}}{t_i - t_{i-1}} \right)^* \left( \frac{D}{2} \right)^* \left( X_{boi} + X_{boi-1} - X_{bi} - X_{bi-1} \right) \] (4)

\[ r_{gh} = q_{di} \frac{X_i - X_{i-1}}{t_i - t_{i-1}} \] (5)

\[ k_{di} = \frac{q_{di} \times 2}{X_i + X_{i-1}} \] (6)

\[ \mu_{fi} = \frac{r_{gh} \times 2}{X_i + X_{i-1}} \] (7)

where the subscripts \( i \) and \( i-1 \) refer to the current and previous time-step values, respectively.
Appendix C

Figure 14.2 from *Biological Wastewater Treatment*
Figure 14.2. Effect of Damköhler number of the effectiveness factor for external mass transfer resistance as predicted by Eq. 14.15.