

BACTERIAL GROWTH IN WATER DISTRIBUTION SYSTEMS

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INTRODUCTION

Occurrences of excessive bacterial populations in distribution systems, sometimes referred to as events or blooms in the literature, have troubled utilities because of their possible implications for the hygienic safety of their product. The water utility industry uses "regrowth" and "aftergrowth" synonymously to describe the processes contributing to this increase in cell numbers with travel time away from the treatment plant. It has often been suggested that cells growing on the inside walls of water mains, as biofilms, play an important role in the regrowth phenomenon. Because the immobilized cells do not wash out as fast as their suspended counter-parts, relatively large biofilm cell populations can accumulate in the system. Biofilm environments also offer ecological advantages like partial protection against disinfectants. The concentration of planktonic cells increases as a result of biofilm erosion and biofilm sloughing.

OBJECTIVES

- 1) Quantify the contribution of detached biofilm cells and the replication of planktonic cells to the total planktonic cell concentration in drinking water. Determine if biofilm growth is significant to the regrowth problem.
- 2) Determine the effect of low concentrations of chlorine on biofilm accumulation and bacteriological water quality.

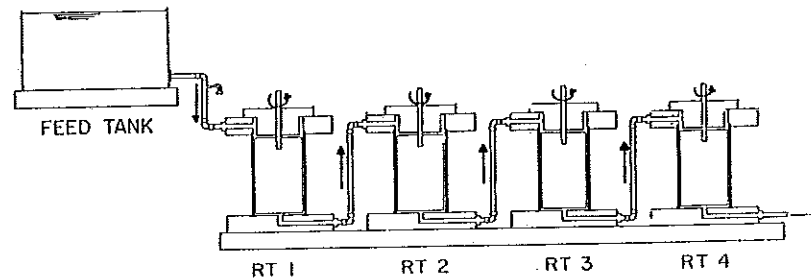


Fig. 1. RotoTorque system.

EQUIPMENT

The experimental apparatus consisted of a newly developed RotoTorque System consisting of four (4) RotoTorque reactors (RT) in series (Figure 1). Each RT is a continuous flow stirred tank reactor (CFSTR) consisting of a removable water pipe section with a solid pvc drum spinning inside it. The volumetric flow rate determines the hydraulic residence time of the RTS. The rotational speed of the drum simulates the shear stress imposed on the inside pipe wall surface of a water main. The RTS simulates the plug flow nature of a water main. Mains of variable length carrying water at variable flow rates are modelled by selecting the appropriate combination of rotational speed and volumetric water flow rate. Since the system allows the independent control of hydraulic residence time and shear stress, their effects on biofilm processes can be studied independently, too. The four (4) RT's serve as "windows" at fixed positions along the modelled main. Water and biofilm samples can be collected at these "windows". The RTS influent in this study consisted of treated effluent from the West River water treatment plant from the SCCRWMA at which the system was installed. Chlorine-free influent was used for the growth rate experiments. For the chlorine experiments, chlorine was added to the influent in a mixed feed tank prior to the RTS. Because chlorine is such a highly reactive compound, a declining chlorine concentration gradient developed through the RTS with increasing residence time.

ANALYTICAL METHODS

Biofilm samples were collected by means of a vacuum technique. A 50 ml plastic sterilized erlenmeyer was supplied with a bent, glass, suction tube and connected to a vacuum pump through a second tube. Two (2) sq in of biofilm were vacuumed off each time with using a rinse of sterile phosphate buffer. 20-40 ml of this buffer were dripped on the remaining biofilm during the last phase of sampling and also vacuumed off. The number of cells in the biofilm samples were determined by means of a spread plate technique. The samples were homogenized and dilution series were made in phosphate buffer. 0.1 ml of each dilution was then plated out on R2A agar in triplicate and the plates were incubated at room temperature for 5 days. After this incubation time all the colonies visible were counted with a 10x magnifying glass to obtain the number of heterotrophic plate count (HPC) bacteria. Water samples from the RT's were diluted with sterilized tap water where necessary. Plating and counting of the samples was done similar as with the biofilm samples.

GROWTH RATES OF BIOFILM CELLS AND PLANKTONIC CELLS

The experimental design was based on the assumption that the replication of planktonic cells would be negligible if the hydraulic residence time in the RTS was significantly shorter than the generation time of the planktonic cells. The determination of biofilm and planktonic cell numbers in the RTS for two (2) suitable hydraulic residence times, one (1) short and one (1) long, could then reveal the growth rates with the help of material balances. Consider the following mathematical expressions which describe the balance of cells in the RTS:

Planktonic Cell Balance in RT1:

$$\frac{dX_1}{dt} = \frac{F}{V} [X_0 - X_1] + \mu X_1 + r_d \frac{X_b}{V} - \frac{A}{V} \quad [1]$$

- where X_1 = planktonic cell concentration in RT1 ($\# \text{ L}^{-3}$)
 X_0 = planktonic cell concentration entering RT1 ($\# \text{ L}^{-3}$)
 X_b = biofilm cell concentration ($\# \text{ L}^{-2}$)
 F = volumetric flow rate through RT1 ($\text{L}^3 \text{ t}^{-1}$)
 V = liquid volume in RT1 (L^3)
 A = surface area for biofilm accumulation in RT1 (L^2)

μ = specific growth rate for suspended cells (t^{-1})
 r_d = specific biofilm detachment rate (t^{-1})

Biofilm cell balance:

$$A \frac{dX_b}{dt} = \mu_b X_b A - r_d X_b A \quad [2]$$

where μ_b = specific growth rate of biofilm cells (t^{-1})

For steady state, the equations simplify to

$$\frac{F}{V} [X_1 - X_0] = \mu X_1 + r_d X_b \frac{A}{V} \quad [3]$$

$$\mu_b = r_d \quad [4]$$

The equations contain three (3) unknowns: μ , μ_b and r_d . However, for the short hydraulic residence time μ will be negligible and the set of equations is determinant.

$$\frac{F}{V} [X_1 - X_0] = r_d X_b \frac{A}{V} \quad [5]$$

Now μ_b and r_d are known and μ can be found using the data from the long hydraulic residence time in Eq. [3].

Results

Most microbial growth occurred in the first RT and specific growth rate of the biofilm cells was calculated as $\mu_b = 0.06 \text{ day}^{-1}$ or a biofilm turnover time of approximately 17 days. The specific growth rate of the planktonic cells in this reactor was -2.9 day^{-1} . The negative value may be explained by analytical error or decreasing cell viability/cell dieoff in RT 1A fluid. Dieoff is quite possible as suggested by the declining viable bacterial numbers in the consecutive RT's operating at the low flow rate. Although the calculated specific growth rate of biofilm cells is low, it is large enough to permit cell numbers in the water of the RIS to increase significantly because of detachment processes. The low specific growth rate in the biofilm is countered by a high concentration of biofilm cells which results in a high cell production rate.

LOW CHLORINE CONCENTRATION GRADIENT

The results for the low chlorine concentration are shown in figure 2 for the exposure times of 17 and 45 days. Biofilm cell activity increases through the RIS as indicated by the distribution of biofilm cells. The numbers of suspended cells increased in proportion to biofilm cell numbers while they reached approximately the same levels as in the chlorine-free system.

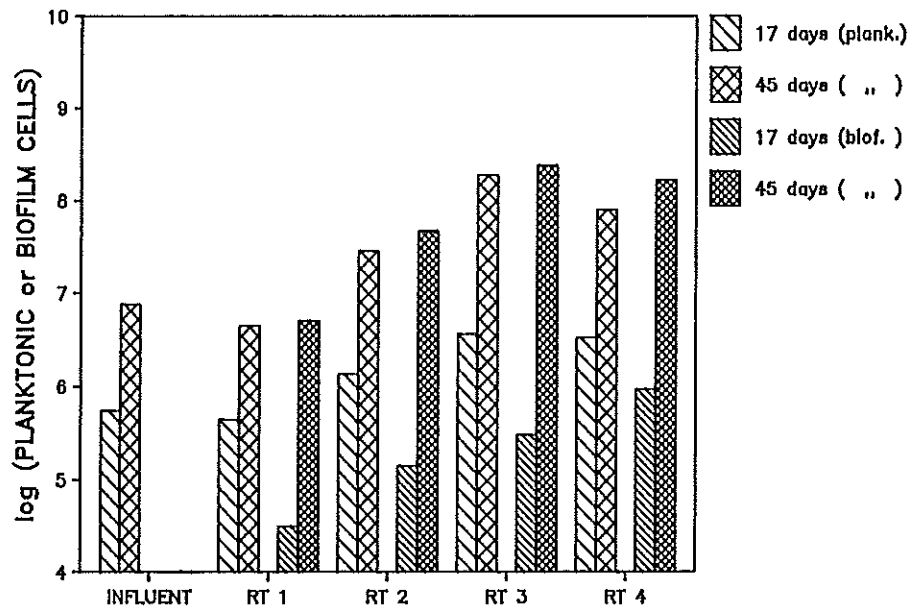


Fig. 2. Planktonic and biofilm cells in RIS with a [chlorine] gradient from .2 to .05 mg/l.