

# Biofilm Floc Nanostructures in Biological Wastewater Treatment in Fisheries

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## ABSTRACT

Biological wastewater treatment modelling has become an important tool in process engineering. There are state of the art activated sludge models (ASMs) available, which have found wide application in the engineering community. Biofilm nanomodels have found less application in engineering practice so far, and a gap has developed between biofilm research and engineering practice in the biofilm modelling community. In this context biofilm and floc nanostructures have played different roles in biological wastewater treatment modelling. Activated sludge models (ASMs) do not explicitly take floc structure into account. In contrast biofilm nanostructure has been strongly emphasized in biofilm models over the past decades. Biofilm models have as a result evolved with increasing complexity from one- to two- to three-dimensional models. The biofilm nanostructure is crucially linked to diffusion by Fick's laws of diffusion in biofilm systems and multidimensional biofilm models with increased model complexity. The biofilms have a complex, heterogeneous three-dimensional nanoscale structures. The increased application of biofilm models in engineering practice is drifting towards simplified (e.g. zero-dimensional) models for this purpose. The diffusion and nanostructure both play an important role in activated sludge systems. The role of activated sludge structure has recently led to the development of multidimensional activated sludge models in activated sludge research. Still, the state of the art ASMs for engineering practice are becoming available as floc structures in waste water treatment.

*Keywords: nanostructures, waste water treatment, fisheries*

## 1 INTRODUCTION

Process modelling has become an important tool in biological wastewater treatment engineering and models are being used for design, control, teaching and research. Biofilm and floc structure have played different roles in biological wastewater treatment modelling. Biofilm structure has been strongly emphasized over the past decades in biofilm research whilst state of the art activated sludge models (ASMs) do not take floc structure into account. It is the aim of this article to analyse and discuss the reasons for the different importance dedicated to biofilm and floc structure in biological wastewater treatment modelling. The role of diffusion as described by Fick's laws of diffusion is discussed and the use and meaning of Monod kinetics in biological wastewater treatment modelling is outlined, since biofilm and floc structure are interrelated to diffusion and

Monod kinetics when modelling microbial growth. Both concepts, i.e. Monod kinetics and Fick's laws of diffusion are introduced in the following paragraphs.

## 2 MONOD KINETICS

Monod kinetics are widely used for modelling microbial growth on a given substrate. The Monod equation (Eq. 1) gives a functional relation between substrate uptake rate ( $\mu$ ), substrate concentration ( $S$ ), Monod affinity constant or half-saturation coefficient ( $K_S$ ), and maximum specific uptake rate ( $\mu_{max}$ ). The functional relation given by the Monod equation is illustrated in figure 1.

$$\mu = \mu_{max} \cdot \frac{S}{K_S + S} \quad (1) \text{Eq.1}$$

Although there have been attempts to give a mechanistic explanation to the Monod equation and although it has formal similarities to Michaelis-Menten kinetics, the Monod equation remains essentially empirical, i.e. it is based on experimental observations and curve fitting of experimental data. In activated sludge models Monod type kinetics are used for mathematical convenience rather than conformity to any fundamental rate law. This has implications for the interpretation of the kinetic parameters, in particular the Monod affinity constant, since wastewater and activated sludge represents a multisubstrate/multispecies system. The value of Monod's affinity constant in activated sludge systems has been discussed in the literature and some principle implications are given here. In single substrate single species systems the affinity constant takes on values of a few mg/l. In multisubstrate/multispecies systems the value of  $K_S$  is typically around 50 mg/l. This difference can be attributed to diffusion within and exterior to flocs. It is thus important to note that diffusion plays an important role in activated sludge processes, and that diffusional mass transport limitations affect the value of Monod's affinity constant when modelling activated sludge systems. Since diffusion is related to floc structure, floc structure plays an implicit role when modelling activated sludge systems. In biofilm models Monod kinetics are generally used to model the intrinsic kinetics of biofilm systems, since diffusion is usually explicitly described using Fick's laws of diffusion.

## 3 FICK'S LAWS OF DIFFUSION

Diffusion processes are governed by Fick's laws of diffusion. Fick's first law (Eq. 2) is used in steady-state diffusion, i.e., when the concentration within the diffusion volume does not change with respect to time. Fick's first law states, that the flux ( $J$ ) of a given substance ( $S$ ) is

proportional to the concentration gradient of the substance along the respective dimension (x), the proportionality factor being the diffusion coefficient (D). Fick's second law (Eq. 3) is used in non-steady or continually changing state diffusion, i.e., when the concentration within the diffusion volume changes with respect to time (t).

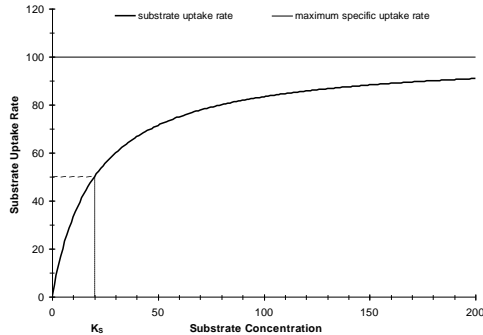


Figure 1. Functional relation between substrate uptake rate and substrate concentration according to the Monod equation ( $v_{max} = 100$ ,  $K_s = 20$ ).

$$J = \frac{D \partial S}{\partial x} \quad Eq.2$$

Eq 2 and 3 are as:

$$J = -D \frac{\partial S}{\partial x} = \frac{\partial S}{\partial t} \quad Eq.3$$

In two or more dimensions Fick's second law can be formulated in generalized form using the gradient operator  $\nabla$  (Eq. 4).

$$\frac{\partial S}{\partial t} = D \nabla^2 S \quad Eq.4 (4)$$

It is apparent from equation 2 – 4 that biofilm and floc structure must have a crucial importance when applying Fick's laws of diffusion to biological wastewater treatment systems, since the spatial dimensions are part of Fick's laws. It will be shown in the following paragraphs that structure and diffusion play an important role in both biofilm and activated sludge systems. In contrast it will be outlined, how diffusion and biofilm structure have been strongly emphasized in biofilm modelling by using Fick's law of diffusion, whilst structure and diffusion is not explicitly described in state of the art activated sludge models.

#### 4 FLOC STRUCTURE IN ACTIVATED SLUDGE MODELLING

The composition of activated sludge flocs is complex, but in a simplified way, three types of bacteria can be considered, which determine the structure of an activated sludge floc: floc-forming bacteria, non-floc-forming bacteria and filamentous bacteria. The filamentous bacteria form the backbone of the activated sludge floc whilst the floc forming bacteria function as the glue that holds the floc together. Settleability is an important sludge property since it determines the solid separation in an activated sludge plant

and it therefore directly affects effluent quality. The settleability is directly linked to floc structure and the causes of settleability problems can be diagnosed and solved using analysis of floc structure. Large flocs will generally settle faster than small flocs of similar density. Floc strength is thus also important, because activated sludge flocs are exposed to a number of shear stresses that can cause the floc to break apart. Weak flocs may break apart resulting in poor settleability and poor dewatering properties.

Floc structure is thus an important operational parameter in activated sludge processes, since it determines sludge properties such as flocculation, settling and dewaterability. Diffusion processes and resulting mass transport limitations are linked to floc structure by Fick's laws of diffusion. In activated sludge systems diffusional mass transport limitations are known to occur and influence process quality; they lead to concentration gradients in the sludge floc and play for example an important role in the formation of bulking sludge. The process kinetics in activated sludge systems are also affected by floc structure, diffusion and diffusional mass transport limitations. The nitrification rate for example is strongly dependent up-on the oxygen concentration in the bulk liquid. This is because the nitrifying organisms are agglomerated in large flocs and the dissolved oxygen concentration within the floc may be considerably less than the bulk fluid concentration.

The presence of concentration gradients in activated sludge flocs has been confirmed by measurement with microelectrodes. These measurements have revealed the presence of anoxic zones inside sludge flocs under aerobic conditions. Diffusion and floc structure are hence significant factors in activated sludge systems. However, the state of the art activated sludge models of the ASM model family do not explicitly integrate diffusion of dissolved species into the sludge floc. Instead diffusional mass transport limitations in the sludge floc are modelled using half-saturation coefficients in the Monod expression which are typically one order of magnitude greater than those reported for single suspended cells.

Diffusion and floc size play thus an important role when estimating kinetic parameters in activated sludge models, and the relation between floc diffusion and kinetic parameters, in particular half-saturation coefficients. Essentially it has been observed and suggested that mass transport limitations are more important in larger flocs than in smaller flocs and that therefore the observed half-saturation coefficients are bigger for larger flocs than for smaller flocs.

Kinetic parameters, and in particular half-saturation coefficients, do thus not represent the intrinsic kinetics of activated sludge flocs in activated sludge modelling. Activated sludge models are used to simulate the macrokinetic behaviour of activated sludge systems, rather than the intrinsic kinetics and micro-environment of the sludge floc. For this reason floc structure is not taken into account in state of the art activated sludge models belonging to the ASM model family.

#### 4.1 THE ROLE OF BIOFILM STRUCTURE IN BIOFILM MODELLING

The picture of biofilm structure has changed over the past decades. Initially biofilms were thought to have a homogeneous structure that covers the substratum as a film, i.e. a thin layer (Fig. 2). This picture was supported by low resolution light microscopy and scanning electron microscopy (SEM). The picture of homogenous biofilm structure has led to the so called continuum approach in biofilm modelling. In the continuum approach it is assumed that the biomass is evenly distributed in the biofilm; concentration gradients are supposed to occur perpendicular to the substratum only.

Biofilm models based on the continuum approach are thus one-dimensional. Until the mid nineties practically all biofilm models were based on the continuum approach. The most prominent example of a one-dimensional biofilm model based on continuum approach is the multispecies biofilm model. This model was later extended and implemented in the computer program AQUASIM. The picture of a homogenous biofilm structure has changed since the mid-nineties, because advanced microscopic examination tools, namely the confocal scanning laser microscope (CSLM), have revealed that biofilms have a complex, heterogeneous three-dimensional structure with pore channels and cell clusters. The picture of homogenous biofilm structure painted by 1D biofilm models in the continuum approach appears therefore to be inadequate. Mass transport into biofilms is not purely diffusional as assumed in 1D biofilm models; in contrast convective mass transport can also take place through pores and channels of the biofilm.

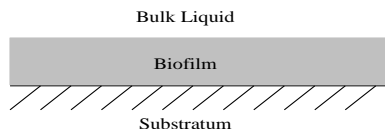


Figure 2. Schematic representation of homogenous biofilm (continuum approach).

It was thus noted that biofilms also have a complex three-dimensional structure like activated sludge flocs. Biofilms are composed of cell clusters and pore channels and biofilm structure can be affected by filamentous bacteria. This biofilm structure suggested two- and three dimensional biofilm models. Two- and three-dimensional biofilm models explain the formation of biofilm structures. However, the picture of biofilm structure painted by 2D and 3D models remains schematic only – the same accounts for the 1D models. On the other hand the development of multi-dimensional biofilm models has not led to more applications of biofilm models in engineering practice.

### 3.2 PROBLEMS AND CURRENT TRENDS: ACTIVATED SLUDGE AND BIOFILM MODELLING

Biofilm models have evolved over the past decades with increasing complexity going from one-dimensional to two-dimensional to three-dimensional models. Unlike in activated sludge modelling, structure has been strongly

emphasized in the biofilm modelling community. A major reason and driving force for this development was on one hand side the importance of diffusion and the related application of Fick's law, which crucially links diffusion to biofilm structure. On the other hand microscopic advances using the confocal laser scanning microscope (CLSM) have revealed that biofilms have a complex three-dimensional structure. This revelation indicates that one-dimensional (stratified) biofilm models paint an inadequate picture of biofilm structure, which has been an additional driving force towards two- and three-dimensional biofilm models. However, a gap has developed in the biofilm modelling community between engineering applications and biofilm research, i.e. biofilm models have been primarily used for research whilst they have found little application in engineering practice because of:

- Biofilm models are perceived as complicated mathematical entities.
- Simplifications and assumptions used in 1D models are often not supported by experimental observations.
- There are many phenomena not considered in the models, such as the fate of particulate substrate, the activity of higher organisms, and the role of exopolymeric substance (EPS) production.
- There is a general lack of trust in the capability of the models to make accurate and reliable predictions.
- The usefulness of biofilm models for the design of full scale systems is not fully appreciated. Many engineers prefer to use simple empirical correlations for design, while models are mostly used as troubleshooting tools when operational problems arise.
- Biofilm models have not been adequately distributed or commercialized.
- Parameters used in biofilm models are sometimes difficult to estimate.

The difficulties encountered when applying biofilm models:

- Biofilm models are too detailed but leave out important factors. Biofilm models have become more and more complex, taking into account an increasing amount of details at the micro-scale. Practitioners are not interested in micro-scale details unless the detailed information becomes directly important for the macro-scale performance of the plant. Thus, for practical applications simpler biofilm models have to be derived from the more complex models. On the other hand, the available models often disregard important processes like attachment and detachment.
- The purpose of mathematical modelling is unclear and many practitioners do not see the need for using mathematical models.
- Too many biofilm models are available. A variety of modelling approaches are available and even biofilm modellers are sometimes in doubt what model to apply for what purpose.
- Model calibration is difficult. A large number of input parameters is required by most models but parameters are often very difficult to determine. Guidance for model calibration is often not provided.

- Models are needed for predictions of dynamic responses to influent variations.
- Operators of wastewater treatment plants need models for trouble shooting and plant optimization.
- Models are needed that integrate the multiple processes, e.g. particle removal, carbon oxidation, nitrification, denitrification, and biological phosphorous removal, thereby helping to understand the complex interactions between these processes.
- Models are needed for reactor design and testing of reactor configurations. These models could be used to evaluate data from pilot-scale plants and to predict the performance of planned full-scale plants.

### 3.3 BIOFILM MODELS

There is a trend towards simplified biofilm models with less complexity, which can overcome the difficulties and which can satisfy the needs described above. In the past this trend has pointed from three-dimensional (3D) towards one-dimensional (1D) biofilm models, although it was known, that 1D biofilm models with purely diffusional mass transport may paint an inadequate picture of biofilm structure and performance, since biofilms have a complex 3D structure with pores and channels, which might allow convective mass transport into the biofilm. 1D model was suited to approximate average concentration profiles of 3D simulations. This points towards 1D models for engineering applications, because simplified models are needed for this purpose. Zero-dimensional (0D) biofilm model has recently been developed for dynamic simulation of moving bed bioreactor (MBBR) i.e. the model was able to dynamically predict effluent quality parameters in response to influent variations. The proposed 0D biofilm model is based on the activated sludge model no. 1 (ASM1). The ASM1 processes were extended with attachment of particulates to the biofilm and detachment of biofilm into the bulk liquid. A key feature of the proposed model is that it does not incorporate biofilm structure in any form. The model does not describe a biofilm structure (for example in layers) across which molecular mass transport is explicitly modelled by means of diffusion. Diffusional mass transport limitations are taken into account implicitly by adapted half-saturation coefficients in the Monod terms of the model instead. Kinetic parameters including half-saturation coefficients were obtained from respirometry. In the proposed approach the macro-kinetic behaviour of the biofilm system is modelled rather than the intrinsic kinetics and the micro-environment of the biofilm itself. On a complexity scale the 0D biofilm model situates prior to the biofilm model evolution from 1D to 3D. The 0D biofilm model situates in the gap between the bottom edge of the complexity scale and biofilm model evolution from 1D to 3D. The 0D biofilm modelling approach appears to be a complementary alternative to the practicing engineer, not at last because it is in-line with current trends, which point towards biofilm models with less complexity for applications in engineering practice shown in Fig. 2. Further a fundamental difficulty has been overcome by the 0D biofilm model, i.e. modelling biofilm structure. However, the 0D

biofilm model needs validation on real-scale plants, and further testing, including application to other biofilm systems.

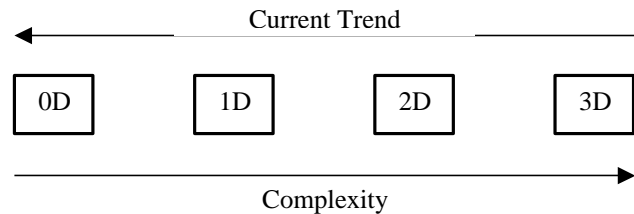


Figure 2. Biofilm model complexity and current trend in the biofilm modelling community.

In activated sludge modelling the state of the art models of the ASM model family [5] have proven to be robust tools in engineering practice over the past decades. The ASM models are zero-dimensional since they do not take activated sludge structure into account, although it is known that floc structure and diffusional mass transport limitations play an important role in activated sludge processes (see chapter 2). The ASM models account for the macro-kinetic behaviour of activated sludge systems rather than the micro-environment of activated sludge flocs and activated sludge structure. Only recently a model, originally developed for a biofilm system, was adapted to simulate three-dimensional (3D) formation of activated sludge flocs [43]. This model takes into account floc-forming and filamentous bacteria; diffusion is the only mass transport mechanism. This type of model can be an important tool in activated sludge research, since it helps to understand floc formation. It appears thus, that knowledge from biofilm research, where structure has been strongly emphasized, can be transferred to activated sludge systems. This approach is promising in order to obtain a better fundamental understanding of activated sludge formation and the activated sludge process itself.

## 4 CONCLUSION

The models should be as simple as possible and as complex as needed for both activated sludge and biofilm systems. Activated sludge and biofilm models have evolved with different degrees of complexity over the past decades. Structure has been strongly emphasized in biofilm modelling. Recent developments points towards simplified (zero-dimensional) biofilm models for engineering application whilst three-dimensional activated sludge models appear to be interesting tools in activated sludge research. New info on activated sludge and biofilm modelling appears to develop: methods from activated sludge modelling (ASM1) have been applied to biofilm systems and methods from biofilm modelling (3D biofilm model) to activated sludge systems. Hopefully biofilm research will give a better activated sludge process.

## 5 REFERENCES

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