A Mathematical Model of a High Sulphate Wastewater, Anaerobic Treatment System

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Synopsis

High sulphate wastewaters, originating from industrial activity or from the biological oxidation of sulphide ores (acid mine drainage), cannot be discharged into the environment untreated. Apart from the high sulphate levels, these waters may be very acidic and have high dissolved heavy metal concentrations. One promising treatment technology is biological sulphate reduction in anaerobic reactors. During anaerobic treatment, sulphate is reduced to sulphide and alkalinity is generated, raising the pH and precipitating many of the heavy metals. The process requires a carbon source as an electron donor. This may be simple organics such as ethanol or volatile fatty acids, which are directly utilized by the sulphate reducing bacteria, or complex organics such as sewage sludge which must first undergo solubilization and fermentation by a different microbial group.

As an aid to the design and operation of this treatment process, a mathematical model describing an anaerobic digester treating high sulphate waste waters has been developed. Apart from sulphate reduction, the model includes those reactions which occur either prior to sulphate reduction, or in competition with it. These include, hydrolysis of solid substrates, acidogenesis, beta oxidation of long chain fatty acids, acetogenesis and methanogenesis. By incorporating terms for these reactions the model is able to simulate sulphate reduction using a wide range of carbon sources.

A comprehensive literature survey of the kinetic parameters for the above reactions was undertaken. Apart from the Monod equation describing substrate uptake the kinetic expressions used in the model also includes terms for: unionized fatty acid inhibition; unionized or total sulphide inhibition; hydrogen inhibition and hydrogen product regulation where appropriate; pH inhibition; and dual substrate uptake where appropriate.

Acid/base equilibrium chemistry has been included in order to predict the pH and unionized component concentrations (needed for calculating inhibition). The weak acids, H_2CO_3 , H_2S , a number of SCFAs, NH_3 , and their ions, as well as the strongly dissociating sulphates Na_2SO_4 and H_2SO_4 are included. An activity based model was used, with the activity coefficients calculated using Debye-Hückle theory. The mass transfer rates of hydrogen, methane, carbon dioxide and hydrogen sulphide from the liquid to the vapour phase are also included.

A final aspect of the model is the equations describing the reactor geometry. A number of different reactors may be simulated, including a dynamic batch, steady state CSTR and dynamic CSTR. By separating the hydraulic and solids residence times, high rate reactors such as UASB and packed bed reactors may also be simulated.

The model has been used to successfully predict the dynamic and steady state behaviour of a number of different reactor types, utilizing both simple and complex carbon sources.

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Nomenclature

As far as possible, the nomenclature adopted for this thesis, is that recommended by the International Association on Water Research and Control and by the Commission on Water Quality of the International Union of Pure and Applied Chemistry (Grau *et al.*, 1982). An important exception to this is the use of the term r rather than r_v to denote volumetric reaction rate. The reason for this is that, all reaction rates presented in this work are volumetrically based.

Where the symbols are typeset in bold, they represent a columns vector of a number of values.

Table 1: Abbreviations

Abbreviation	Definition
ACE	Acetogenic bacteria (acetogens)
<i>p</i> ACE	Propionate utilizing acetogens
<i>b</i> ACE	Butyrate utilizing acetogens
AcH	Acetic acid (acetate)
BOB	Long chain fatty acid beta oxidizing bacteria
BuH	Butyric acid (butyrate)
COD	Chemical oxygen demand
FER	Fermenters (Acidogenic bacteria)
<i>g</i> FER	Fermenters utilizing glucose
<i>l</i> FER	Fermenters utilizing lactic acid
GI	Glucose
LaH	Lactic acid (lactate)
LCFA	Long chain fatty acid
MPB	Methane producing bacteria
<i>a</i> MPB	Acetate utilizing methane producing bacteria
<i>h</i> MPB	Hydrogen utilizing methane producing bacteria
PrH	Propionic acid (propionate)
SRB	Sulphate reducing bacteria
<i>a</i> SRB	Acetate utilizing sulphate reducing bacteria
<i>b</i> SRB	Butyrate utilizing sulphate reducing bacteria
<i>h</i> SRB	Hydrogen utilizing sulphate reducing bacteria
<i>l</i> SRB	Lactate utilizing sulphate reducing bacteria
<i>p</i> SRB	Propionate utilizing sulphate reducing bacteria
UASB	Upflow anaerobic sludge blanket reactor
VFA	Volatile fatty acid

Table 2: Roman Symbols

Symbol	Quantity name	Units	Defined in
A	Constant used to calculate I from Davies equation	-	Equation 3.10
b	Specific biomass decay rate	d^{-1}	Equation 2.3
c	Molar concentration in liquid phase	$mmole \cdot l^{-1}$	
FS	Free sulphide concentration	$mg \cdot l^{-1}$	Equation 4.1
$\mathcal{H}$	Henry's law constant	$atm/(mg/l)$	
HRT	Hydraulic residence time	days	Equation 3.27
I	Concentration of inhibitory compound	$mg \cdot l^{-1}$	Equations 2.9–2.12
I	Ionic strength	-	Equation 3.7
k	Substrate utilization rate constant	day^{-1}	Equation 2.7
k_h	First order hydrolysis rate constant	day^{-1}	Equation 2.16
k_{la}	Product of gas transfer coefficient and specific interfacial area	day^{-1}	Equation 3.1
K_a	Acid dissociation constant	$mmole \cdot l^{-1}$	Equation 3.4
K_I	Inhibition constant	$mg \cdot l^{-1}$	Equations 2.9–2.12
K_S	Half velocity constant	$mg \cdot l^{-1}$	Equation 2.6
K_{sp}	Solubility product	varies	Equations 3.10–2.12
m	Solids retention factor in high rate digester model	-	Equation 3.22
N	Gas mass transfer rate per unit volume	$mmole \cdot l^{-1} d^{-1}$	Equation 3.1
P	Total pressure	atm	
p	Partial pressure of species	atm	
Q	Volumetric flow rate of liquid	l/day	
Q_G	Volumetric flow rate of gas at STP	l/day	
r	Rate of substrate utilization per unit volume	$mg \cdot l^{-1} \cdot d^{-1}$	Equation 2.5
r_{cell}	Rate of biomass production per unit volume	$mg \cdot l^{-1} \cdot d^{-1}$	Equation 2.3
R	Rate of substrate utilization per unit volume (cell energy production only)	$mg \cdot l^{-1} \cdot d^{-1}$	
R	Universal gas constant (8.206×10^{-5})	$(atm \cdot l)/(mmole \cdot K)$	
S	Concentration of dissolved component	$mg \cdot l^{-1}$	
SRT	Solids residence time	days	Equation 3.27
T	Temperature	K	
V_l	Liquid phase volume of reactor	l	
V_v	Vapour phase volume of reactor	l	
VFA	Total concentration of unionized volatile fatty acids	$mmole \cdot l^{-1}$	Equation 4.1
X	Concentration of insoluble component	$mg \cdot l^{-1}$	
X_{cells}	Concentration of biomass	$mg \cdot l^{-1}$	Equation 2.3
X_{deg}	Concentration of biodegradable portion of insoluble species	$mg \cdot l^{-1}$	Equation 2.2
X_{∞}	Concentration of nondegradable portion of insoluble species	$mg \cdot l^{-1}$	Equation 2.17
Y	Biomass yield coefficient (based on overall substrate consumption)	$mg \cdot mg^{-1}$	Equation 2.5
Y'	Biomass yield coefficient (based on the production of a compound)	$mg \cdot mg^{-1}$	
z	Ionic charge	-	Equation 3.7

Table 4: Greek Symbols

Symbol	Quantity name	Units	Defined in
α	Parameter in on/off switching function	-	Equation 2.13
α_{LL}	Parameter in pH inhibition function	pH	Equation 2.14
α_{UL}	Parameter in pH inhibition function	pH	Equation 2.14
β	Parameter in on/off switching function	-	Equation 2.13
γ	Activity coefficient	-	Equation 3.5
μ	Specific biomass growth rate	$\text{mg} \cdot \text{mg}^{-1} \cdot \text{d}^{-1}$	Equation 2.3
μ_{max}	Maximum specific biomass growth rate	$\text{mg} \cdot \text{mg}^{-1} \cdot \text{d}^{-1}$	Equation 2.6
μ_{obs}	Observed or net specific biomass growth rate	$\text{mg} \cdot \text{mg}^{-1} \cdot \text{d}^{-1}$	Equation 2.4
ξ_j	Factor modifying specific biomass growth rate due to condition j		Equation 2.15 and table 2.1

Chapter 1

Introduction

1.1 Background

High sulphate waste waters originate from a number of different industrial activities. These include:

- The manufacture of pulp and paper.
- Minerals processing.
- Explosives, fertilizers and other petro-chemical industries.
- Mining, i.e. acid mine drainage.

Of these four points, the formation of acid mine drainage (AMD) is—at least in a South African context—potentially the most significant. Two factors combine to make this so: Firstly, South Africa is a semi-arid to arid country and as such its water resources are particularly valuable. In addition South Africa has a large minerals producing industry, and as a consequence has a large number of mines, either still in operation or abandoned.

Acid mine drainage (also known as acid mine water or acid rock drainage) results when sulphide containing rock is exposed to water and oxygen as a result of mining activity. The sulphide is biologically oxidized by species such as *Thiobacillus ferrooxidans*. The reaction for pyrite is (Thompson, 1980)

further acid is produced by subsequent chemical oxidation and hydrolysis of the iron sulphate, i.e.

Acid mine drainage may originate within mine dumps (the water and oxygen being provided by rainfall percolating into the dump) or deep underground where the water is naturally occurring subsurface water. Underground AMD may be pumped to the surface in order to prevent the mine from flooding, or could emerge at the surface as a result of natural drainage.

1.2 Ecological, Industrial and Social Impact of Sulphate Waste Waters

High sulphate waste water streams discharged into the environment have a number of associated environmental and social penalties. Sulphate itself, results in an increase in the total dissolved solids content of the water, which may affect its usefulness downstream. (e.g., as drinking water, irrigation water or industrial water). In the environment sulphate may be biologically reduced, resulting in the formation of hydrogen sulphide with concomitant odour problems. If the pH of the stream is low, its discharge can result in the complete sterilization of the receiving waters.

A particular problem, associated with acid mine drainage, is its high heavy metals content. Even those metals which are essential for life in small quantities, such as zinc and copper, can become toxic at high concentrations. The low pH of AMD results in metals being leached from the surrounding rock. Iron is usually present in significant quantities while the presence of other heavy metals is dependant on the nature of the ore. Table 1.1 gives the composition of some typical AMD streams around the world.

Table 1.1: Composition of raw AMD streams world wide

Species	Concentration (mg/l)				
	Grootvlei ¹	Anaconda Copper ²	Avoca ³	Wallenberg ⁴	Gammelgruva ⁵
pH	6.25	2.5	2.7	5.52	2.40
TDS	3028				
SO ₄	1383	3510	10579	2940	6600
Cl	350				
F	1.8				
Na	291	72.0			
K	12.9	21.7			
Fe	187	300	1031	139	1160
Zn	0.077	155	362	33.5	89.2
Cu	0.5	29	243	1.45	99.2
Ni	3.2	0.5			
Cr	0.087				
Al	0.025	125		1.38	263
Pb	0.077				
Cd	0.025		1.198		
Ca		215		500	382
Mn	5.3	88		13.6	11.7
Mg		225		350	212
Si		22.0			
Flow (Ml/d)		22.6			

¹Grootvlei mines, Springs, South Africa (Department of Mineral and Energy Affairs, 1995)

²Anaconda Copper Mining Company, Butte, MT, USA (Jenke and Diebold, 1983)

³Avoca mines, County Wicklow, Ireland (Gray, 1996)

⁴Wallenberg mine, Norway, (Christensen *et al.*, 1996)

⁵Gammelgruva mine, Norway, (Christensen *et al.*, 1996)

Very often it is desirable to reuse industrial waste streams—possibly after treatment—in order to reduce water consumption and effluent output. Particular problem associated with sulphate are scaling (e.g., as MgSO₄), biocorrosion of metals, and weakening of concrete structures due to calcium leaching.

1.3 Overview of Existing and Emerging Treatment Technologies

A number of different treatment technologies have been developed for the treatment of low pH, high sulphate streams. These range from mature technologies, for which a number of full scale plants exist, to emerging technologies which may only have been tested on a laboratory or pilot plant scale.

The most widely used process is neutralization with slaked lime ($\text{Ca}(\text{OH})_2$), followed by solids removal by settling and filtration. During neutralization, the metals precipitate as their hydroxides, and sulphate precipitates as gypsum. Soda ash (Na_2CO_3) may be added in a second step to raise the pH further in order to precipitate any calcium remaining in solution. Calcium carbonate formed in this stage can be recycled to the first neutralization stage. Significant disadvantages of this process are: the large chemical requirements, and the large quantities of sludge produced. In addition the sludge is difficult to dewater and needs to be disposed of by landfill.

Membrane technologies, such as reverse osmosis, may be used as a final polishing step. For example RGC Mineral Sands Ltd (Australia) operate a reverse osmosis plant, for final sulphate and sodium removal after a neutralization sequence (Cinanni *et al.*, 1996). Disadvantages of membrane plants, include the potential for membrane fouling and the high costs of the membrane itself. In addition, the concentrated waste stream from the plant requires disposal—possibly by evaporation and crystallization.

Other nonbiological treatment options which have been proposed are electroprecipitation and ion exchange.

Biological treatment of high sulphate waters has received increased attention recently. Two process options exist, sometimes termed 'passive' and 'active'. In a passive system, the water is passed through a man-made wet land. Under the anaerobic conditions present, the sulphate is reduced to sulphide, by sulphate reducing bacteria (SRB). The pH of the water is increased, as the strong acid H_2SO_4 is reduced to the weak acid H_2S , and by the production of CO_2 . The increase in pH and the generation of sulphide results in the precipitation of the heavy metals present.

The sulphate reducing bacteria require a carbon source, both for biomass growth and as an electron donor. This may be added to the wetland as a high COD stream, or can result from the growth and death of plant matter in the wetland.

A particular attraction of wetlands is the low effort and cost required for their operation. Passive systems also have several disadvantages. Firstly they require a large land area which may be expensive or unavailable. The formation of hydrogen sulphide results in odour problems, and since the gas is not collected there is no potential for sulphur recovery. A potential long term problem is the build up of metal sludge in the wetland. This must ultimately be disposed of, since should the wetland become aerobic—possibly in several generations time—there is the potential for the metal sulphide to reoxidize and leach out.

In contrast, active systems refer to biological sulphate reduction occurring in a conventional anaerobic digester. While only a few full scale plants treating high sulphate streams exist (Colleran *et al.*, 1994; Scheeren *et al.*, 1992; Barnes *et al.*, 1992), numerous laboratory and pilot plant studies have been done. Table 1.2 lists the reactor configurations, carbon sources used and removal rates obtained, of some of these experiments. A number of different carbon sources and electron donors

Table 1.2: Overview of operating conditions and effluent quality of a number of sulphate reducing reactors. Exit concentrations and specific removal rates shown are those obtained during stable reactor operation.

Reference	Reactor	Scale	Carbon Source	HRT days	Feed g SO ₄ ²⁻ /l	Exit g SO ₄ ²⁻ /l	Specific removal rate g SO ₄ ²⁻ · l ⁻¹ · d ⁻¹
Maree <i>et al.</i> (1989)	Upflow packed bed	Laboratory	Molasses	2	2.4	0.2	1.1
Omil <i>et al.</i> (1996)	UASB	Laboratory	AcH/PrH/BuH	0.17-0.26	1.2-4.6	0.38-3.2	~ 4
Stucki <i>et al.</i> (1993)	Upflow packed bed	Laboratory	Acetate		0.17	0.77	up to 6.5
Oleszkiewicz and Hilton (1986)	UASB	Laboratory	Cheese whey		2.8-12.7		3.4-3.8
Pavlostathis and Giraldo-Gomez (1991)	Upflow packed bed	Laboratory	Propionate	2	2.8	0.5	1.2
Colleran <i>et al.</i> (1994)	Upflow packed bed	Full scale	Beet molasses	1.4	3.43	0.25	2.27
Colleran <i>et al.</i> (1994)	Upflow packed bed	Laboratory	Beet molasses	1.4	4.0	1.8	1.57
Maree and Strydom (1983)	Upflow packed bed	Laboratory	Pulp mill effluent	2	1.35	0.172-0.450	0.45-0.59
Maree and Strydom (1983)	Upflow packed bed	Laboratory	Sewage sludge	2	1.35	0.3	0.525
Maree and Strydom (1983)	Upflow packed bed	Laboratory	Sewage sludge & sugar	2	1.35	0.05	0.65
du Preez and Maree (1994)	Upflow packed bed	Laboratory	Producer gas (H ₂ /CO)		2.0	0.1	1.2-2.4
Moosa (1984)	Baffled plate	Laboratory	Unknown	0.5-4.0	13	2.6-9.1	2.6-3.9
Mizuno <i>et al.</i> (1994)	CSTR	Laboratory	Butyrate	5-20	5.0	2.4-3.1	0.13-0.38
van Houten <i>et al.</i> (1994)	Gas lift	Laboratory	H ₂ /CO ₂	0.19	3.36		14
Corbett (1998)	Valley reactor + Baffle plate	Pilot Plant	Sewage sludge	1.6 + 1.7	1.7	0.5	0.36
Scheeren <i>et al.</i> (1992)	UASB	Pilot Plant	Ethanol	0.33	1.4	0.12	3.8

have been shown to be suitable for sulphate reduction. These may be classified into three groups:

- Complex organics, such as sewage sludge, pulp mill effluent, molasses and cheese whey. The organic compounds present in these wastes are not directly used by sulphate reducing bacteria. In a system fed with complex organics, acidogenic bacteria are responsible for degrading the compounds to simpler molecules which can be utilized by sulphate reducers.
- Simple organic compounds, such as short chain fatty acids and ethanol. Sulphate reducing bacteria are able to directly utilize these compounds, although they may be in competition for these substrates with other bacteria.
- $\text{CO}_2/\text{CO}/\text{H}_2$ (Producer gas). Some sulphate reducing species are able to utilize hydrogen as the electron donor and carbon dioxide or carbon monoxide as a carbon source.

In the cases where the sulphate stream does not already have an organic component, such as AMD, the choice of carbon feed stock will be governed by economic considerations. Simple compounds are more easily degraded and hence require a smaller reactor. In addition the residual COD after treatment will be low. Since they must be produced industrially, and possibly transported to the site, their cost can be high. In contrast, complex organics are usually available as waste streams, such as sewage, and are consequently less costly. However, the fact that they are less easily degraded means that reactor sizes will be larger, and that a residual COD component may remain after anaerobic treatment—making further treatment necessary.

One attraction of active anaerobic systems is the possibility of sulphur and heavy metals recovery. Hydrogen sulphide stripped from the product water can partially oxidized to elemental sulphur, either by chemical or biological means. In the cases where only one metal is present in a significant concentration, the biological sludge with the precipitated metals may be fed to a refinery. In the case where more than one metal is present, by recycling the sulphide and alkalinity generated in the reactor, it is possible to sequentially precipitate the metals upfront of the reactor. Both biological sulphur recovery and metal recovery have been implemented in at least one plant. (Barnes *et al.*, 1992; Scheeren *et al.*, 1992).

1.4 Objectives and Outline of this Thesis

Given the enormous potential for anaerobic sulphate reduction, as a treatment for acid mine drainage and other high sulphate waste waters, it is desirable that a mathematical model of such a system be made available. It is anticipated that a model of an anaerobic sulphate reducing system would have several uses:

- *Process design.* For example the model could be used to evaluate different feed stocks and reactor configurations, without the need to run numerous experimental studies.
- *Process operation and control.* For example a dynamic simulation could be used to quantify the effects of a change in loading rates, and to evaluate different control strategies.

- *Process insight.* One (sometimes serendipitous) result of modelling a process in detail is that it often gives valuable insights, many of which are not apparent when the process is considered as a macroscopic, 'black box', whole.

While general anaerobic mathematical models, beginning with that of Graef and Andrews (1974), have been developed for several decades, only in recent years have models incorporating sulphate reduction emerged (Kalyuzhnyi and Fedorovich, 1997; Vasiliev *et al.*, 1993; Vavilin *et al.*, 1995). In some respects these models are not yet mature, in that they omit some of the more advanced features of the older non-sulphate models. In addition, not all of the possible electron donor substrates have been considered. This is usually the result of a deliberate simplification, but does limit the usefulness of the models in some cases.

The aim of this thesis is then to develop a comprehensive mathematical model describing the anaerobic treatment of high sulphate waste waters. No laboratory experimental work formed part of the project, data for the model's development and calibration being taken from literature. In addition this project was confined to the anaerobic stage of treatment—the further recovery of sulphur from the sulphide stream, by conventional or microbiological means, was not considered.

Gerber (1982) describes the development of a mathematical model as being composed of a number of stages, namely:

- A *conceptual stage* is which the cause-effect relationships of the system are established, and a framework for the model is proposed.
- The *functional representation* of the model in which the governing relationships are translated into mathematical expressions.
- Once the mathematical model has been formulated, a *computational scheme* for its solution is developed. This may be analytical, if the model is so amenable, or numerical, in which case a computer is employed.
- The final stage is the *validation* of the model. Gerber further divides this into two sub-stages. During *calibration* the model's parameters are adjusted so that the model fits experimental data. The model is then *verified* using separate independent data.

Gerber adds that "although the developer must negotiate each of the individual steps the use of models cannot be considered a step-by-step procedure. When dealing with real systems a model is never exact and complete data are never available, therefore model use is an iterative process to which one never achieves a final solution".

The outline of this thesis is as follows: In Chapter two the fundamental aspects of modelling the kinetic behaviour of an anaerobic system are considered. Kinetic constants compiled from a comprehensive literature survey are tabulated. In Chapter three, the non-microbial aspects of an anaerobic model are discussed. These include the solution chemistry, vapour/liquid mass transfer effects and the geometry of various reactors.

Chapter four takes a closer look the kinetic constants compiled and a procedure for selection of suitable constants for use in the model is suggested. This chapter also describes the implementation of the model using the simulation packages Aspen Plus¹, Matlab² and Octave³. Several test experiments are performed for calibration and model verification, and the results of this are discussed.

¹ Aspen Plus is a trademark of Aspen Technology, Inc.

² Matlab is copyright by the Math Works, Inc.

³ Octave is copyright by John W. Eaton

In Chapter five a simulation showing how the model may be used for design is given. Finally, conclusion and recommendations for further research—both numerical and experimental—are given in Chapter six.

Chapter 2

Microbiological Fundamentals

This chapter presents a detailed discussion of the fundamentals of modelling an anaerobic system. The first section presents an overview of the microbial reactions occurring in an anaerobic system. Following this, the various forms of soluble and solid substrate kinetics useful to the modeller are reviewed. The bulk of the chapter then deals with the biological reaction stoichiometry and a survey of kinetic data, as taken from a large selection of literature. Various aspects of the competition of sulphate reduction with methanogenesis are discussed at the conclusion of the chapter.

At this point an important note, regarding the notation used to describe the microbial stoichiometric reactions given below, is required. In the reactions presented all of the compounds are represented as their unionized forms. However, the formula should be interpreted as representing the *total* concentration of the species (i.e. both ionized and unionized). For example, the acetate sulphate reduction reaction:

means that one mole of total acetate (i.e. $\text{CH}_3\text{COOH} + \text{CH}_3\text{COO}^-$) and one mole of sulphate ($\text{HSO}_4^- + \text{SO}_4^{2-}$) will produce one mole of hydrogen sulphide ($\text{H}_2\text{S} + \text{HS}^- + \text{S}^{2-}$) and one mole of carbon dioxide ($\text{CO}_2 + \text{H}_2\text{CO}_3 + \text{HCO}_3^- + \text{CO}_3^{2-}$).

While in some cases, over the pH ranges expected, the components will only be present in significant quantities in one form (e.g. SO_4^{2-}), those that have pK_a values close to the system pH will be present in more than one form. This is particularly true for hydrogen sulphide and carbon dioxide.

Using the above approach, the biological rate reactions describe the consumption or production of the total amount of each species present. As is expanded on in the following chapter, the exact state of ionization of each species is then determined from the equilibrium solution chemistry.

2.1 Overview of the Biological Reactions in an Anaerobic System

Within a high sulphate anaerobic system, utilizing insoluble primary sludge as a carbon source, the following biological processes can be expected to occur:

- Colonization of the sludge particles by acid fermenting bacteria. The particles are hydrolyzed extracellularly by enzymes secreted by the bacteria. The carbohydrate fraction is broken down to sugars, the protein fraction to amino acids and the lipid portion to long chain fatty acids (LCFAs) and polyols such as glycerol.
- Acid fermentation (acidogenesis) of the sugars and amino acids, to hydrogen, carbon dioxide and short chain fatty acids (SCFAs) such as acetic, propionic, butyric and lactic acids.
- Beta oxidation of the long chain fatty acids to yield acetate, propionate and hydrogen.
- Anaerobic oxidation (acetogenesis) of propionate and other SCFAs to acetate, hydrogen and carbon dioxide.
- Methanogenesis using hydrogen and acetate as substrates.
- Biological sulphate reduction utilizing hydrogen, acetate, propionate and potentially other intermediate SCFAs as substrates.
- Homoacetogenesis, i.e. the production of acetate from hydrogen and carbon dioxide.

A diagram showing the interacting flows of substrates between each biological process is shown in figure 2.1.

2.2 Preliminary Discussion of Kinetics

Before reviewing the kinetic data and reaction stoichiometry of the various microbial processes involved in anaerobic digestion and sulphate reduction, it is convenient to provide an overview of the various mathematical expressions used to describe microbial biological kinetics. For reasons that will become apparent, the kinetic forms may be broken into two groups, namely, first order kinetics describing the degradation of insoluble components, and Monod and related kinetics describing the uptake of soluble substrates.

2.2.1 First Order Form Kinetics (Insoluble Substrates)

The breakdown of insoluble particles to soluble components represents the cumulative effects of a number of processes, including: the colonization of the particles by bacteria, the mass transport of enzymes into the particle, and the degradation of a number different components at varying rates. This is usually most easily represented by an empirical first order expression of the form:

$$r_X = -kX \quad (2.1)$$

where r_X is the volumetric rate of degradation ($\text{mg} \cdot \text{l}^{-1} \cdot \text{d}^{-1}$), k is a first order rate constant (d^{-1}) and X is the mass concentration (mg/l) of the insoluble component. In most cases a certain fraction of the substrate is resistant to breakdown. Equation 2.1 is modified to take this into account, giving:

$$r_X = -k(X - X_\infty) \quad (2.2)$$

where X_∞ is the mass concentration of the nondegradable fraction (mg/l).

Figure 2.1: Microbial ecology and major substrate flows in an anaerobic system. Dissociation and vapour/liquid mass transfer steps not shown. Depending on reactor condition not all pathways may be active.

2.2.2 Monod and Related Growth Kinetics (Soluble Substrates)

The volumetric net rate of biomass production r_{cells} ($\text{mg} \cdot \text{l} \cdot \text{d}^{-1}$) is usually determined from:

$$r_{cells} = (\mu - b)X_{cells} \quad (2.3)$$

$$= \mu_{obs}X_{cells} \quad (2.4)$$

where μ is the specific biomass growth rate (d^{-1}), b is the specific biomass loss rate due to cell death (d^{-1}), X_{cells} is the biomass concentration (mg/l) and μ_{obs} is the observed or net specific biomass growth rate (d^{-1}). The volumetric rate of substrate utilization r_S ($\text{mg} \cdot \text{l} \cdot \text{d}^{-1}$) is given by:

$$r_S = -\frac{\mu X_{cells}}{Y} \quad (2.5)$$

where Y is the biomass yield coefficient ($\text{mg cells/mg substrate}$).

The specific biomass growth rate (μ) is almost always a function of the substrate concentration, and is also potentially dependent on inhibitory or regulatory factors. Based on these considerations several expressions for determining μ have been put forward.

The most simple of these is to assume a constant value for μ , in which case equations 2.3 and 2.5 become first order expressions. In practice it is found that, while this is usually the case at high substrate concentrations, when the substrate concentration is low (saturation conditions) deviation from first order behavior is observed. This effect can be accounted for by the Monod model:

$$\mu = \frac{\mu_{max} S}{K_S + S} \quad (2.6)$$

where μ_{max} is the maximum specific biomass growth rate (d^{-1}), S is the soluble substrate concentration (mg/l) and K_S is the Monod half velocity constant (mg/l). K_S has the effect of providing a minimum substrate threshold below which substrate uptake ceases—a phenomenon which occurs in practice. Frequently the specific rate of substrate utilization k ($\text{mg substrate} \cdot \text{mg cells}^{-1} \cdot \text{d}^{-1}$) is reported instead of μ_{max} . The two are related by the expression:

$$k = \frac{\mu_{max}}{Y} \quad (2.7)$$

In some instances two or more substrates may be limiting, in which case a multiple term Monod equation (Bailey and Ollis, 1986) is useful:

$$\mu = \mu_{max} \frac{S_1}{K_{S,1} + S_1} \frac{S_2}{K_{S,2} + S_2} \cdots \frac{S_n}{K_{S,n} + S_n} \quad (2.8)$$

A reduced growth rate may occur due to the presence of inhibitory compounds in the system. These could be substances present in the feed (e.g. heavy metals), substances produced by the species under consideration (product inhibition), substances produced by a different, possibly competing, species or even high levels of substrate. Several models have been put forward to account for this.

Noncompetitive inhibition combines an inhibition factor with equation 2.6 giving:

$$\mu = \mu_{max} \frac{S}{K_S + S} \frac{K_I}{K_I + I} \quad (2.9)$$

where K_I is the inhibition constant (mg/l) and I is the concentration of the inhibitory substance (mg/l). In the case of noncompetitive inhibition K_I may be regarded as the concentration of the inhibitory compound which results in a 50% decrease in specific growth (under first order conditions). A modified form of equation 2.9 has been used by at least one group of workers (Reis *et al.*, 1990) and is given by:

$$\mu = \mu_{max} \frac{S}{K_S + S} \frac{1}{1 + (I/K_I)^n} \quad (2.10)$$

For the case $n = 1$ equation 2.10 reduces to equation 2.9. Other inhibition models are uncompetitive inhibition:

$$\mu = \mu_{max} \frac{S}{K_S + S \left(1 + \frac{I}{K_I}\right)} \quad (2.11)$$

and competitive inhibition:

$$\mu = \mu_{max} \frac{S}{S + K_S \left(1 + \frac{I}{K_I}\right)} \quad (2.12)$$

where K_I and I have the same meaning. A better understanding of the effect the various inhibitory equation have on the specific growth rate, can be obtained from examining plots of μ/μ_{max} versus substrate concentration. Plots of this sort along with a plot of a standard Monod equation for comparison are given in figure 2.2.

Three observations may be made. Firstly it is apparent that although all of the plots use the same values of K_I and K_S and the same concentration of I , the levels of inhibition are very different. In other words although the units of K_I are the same no comparison can be made between values relating to different inhibition functions. A second point worth noting is that a *lower* K_I results in a *greater* degree of inhibition. The third point relates to the concentration ranges at which the inhibition functions have their greatest effect. Noncompetitive inhibition exerts a constant inhibitory effect over the entire concentration span, uncompetitive inhibition results in greater inhibition at higher substrate concentrations, while the converse is true for competitive inhibition. This is illustrated in figure 2.3 where a plot of $\mu_{inhibited}/\mu_{noninhibited}$ against substrate concentration for the three functions is given.

An alternative way of accounting for inhibition is to include an 'on/off' switching function as a factor of μ . A suitable expression is the sigmoidal function:

$$\xi_{SF} = \frac{1}{1 + e^{\alpha(C-\beta)}} \quad (2.13)$$

where ξ_{SF} varies between 0 and 1; C is some reactor condition affecting the reaction rate (e.g. pH, concentration or partial pressure); β is the value of this condition at $\mu = \frac{1}{2}\mu_{max}$ (no other factors affecting μ); and α sets the stepness of the curve (the higher value the more rapid the change) and whether the function is switched on or off (a negative value sets ξ_{SF} to 1 as C is increased, and vice versa). An example plot of equation 2.13 is given in figure 2.4.

(a) $K_I = 100$ (b) $K_I = 50$ **Figure 2.2:** Illustrative plots of inhibition functions. $K_S = 50$, $I = 150$.

Figure 2.3: Inhibition as a function of substrate concentration for various inhibition functions. $K_I = 100$, $K_S = 50$, $I = 150$.

A particularly useful application of switching functions is to account for the inhibitory effects of pH. Most biological reactions occur only within a specific pH range—the reaction rate rapidly dropping to zero at too low or high pH values. The product of two switching functions (one on, one off) provides an easy way to account for this, i.e:

$$\xi_{pH} = \left(1 + e^{-\alpha_{LL}(pH - pH_{LL})}\right)^{-1} \left(1 + e^{\alpha_{UL}(pH - pH_{UL})}\right)^{-1} \quad (2.14)$$

where ξ_{pH} takes on a value between 0 and 1; pH_{LL} and pH_{UL} are upper and lower pH limits, defined as the values at which $\mu = \frac{1}{2}\mu_{max}$ due to pH effects; and α_{LL} and α_{UL} are positive values which effect the steepness of the curve. Higher α values result in a more rapid onset of inhibition.

An illustrative plot of equation 2.14 is shown in figure 2.5. Values of $\alpha_{LL} = 2$ and $\alpha_{UL} = 8$ are used resulting in a gradual onset of inhibition at the lower pH end but a more rapid onset at the upper values.

From the above it is apparent that the specific growth rate can be affected by a number of factors. For the purposes of developing a mathematical model it is convenient to express μ as a product of μ_{max} and several modifying factors, i.e:

$$\mu = \mu_{max} \prod_i \xi_i \quad (2.15)$$

where ξ_i are appropriate factors. A summary of ξ_i terms is given in table 2.1.

Figure 2.4: Plot of $\xi_{SF} = \frac{1}{1+e^{-\alpha(C-\beta)}}$ with $\alpha \approx 3, \beta = 5$.

Figure 2.5: Illustrative plot of the pH inhibition switching function (equation 2.14), with $\alpha_{LL} = 2, \alpha_{UL} = 8, pH_{LL} = 5$ and $pH_{UL} = 8$.

Table 2.1: ξ Factors modifying the specific biomass growth rate. Parameters are defined in the text.

Notation	Description	Form
ξ_{SU}	Substrate utilization	$\frac{S}{K_S + S}$
ξ_{NI}	Noncompetitive inhibition	$\frac{K_I}{K_I + I}$
ξ_{NI^n}	Noncompetitive inhibition (variation)	$\frac{1}{1 + \left(\frac{I}{K_I}\right)^n}$
ξ_{UI}	Substrate utilization with uncompetitive inhibition	$\frac{S}{K_S + S \left(1 + \frac{I}{K_I}\right)}$
ξ_{CI}	Substrate utilization with competitive inhibition	$\frac{S}{S + K_S \left(1 + \frac{I}{K_I}\right)}$
ξ_{SF}	On/off switching function	$\frac{1}{1 + e^{\alpha(C - \beta)}}$
ξ_{pH}	pH inhibition on/off switching function	$(1 + e^{-\alpha_{LL}(pH - pH_{LL})})^{-1} \times (1 + e^{\alpha_{UL}(pH - pH_{UL})})^{-1}$

2.3 Hydrolysis of Insoluble Substrates

2.3.1 Overview

Primary sludge originates from the solid component of raw sewage settled prior to any biological treatment. The sludge consists mainly of insoluble lipids, proteins and carbohydrates. During hydrolysis the exterior of the particles is colonized by acid forming bacteria. The bacteria secrete hydrolytic enzymes which are responsible for the extracellular hydrolysis of the sludge. Specifically the following reactions may be expected to occur:

- The hydrolysis of the amide bonds of proteins to yield amino acids.
- The hydrolysis of the ester bonds of lipids to yield long chain fatty acids, glycerol (and other polyols) and alcohols.
- The hydrolysis of the glycoside bonds of polysaccharides to yield dimeric and monomeric sugars.

With respect to the subsequent fermentation and anaerobic oxidation steps, the extracellular hydrolysis of sludge is rate limiting (Eastman and Ferguson, 1981). Consequently it is difficult to experimentally determine the rate of hydrolysis alone. For this reason the hydrolysis and acid fermentation steps are often modelled as a single step.

The composition of primary sludge as taken from several sources is shown in table 2.2.

Table 2.2: Composition of primary sludge as a percentage of dry matter. After Pavlostathis and Giraldo-Gomez (1991).

Component	O'Rourke (1968)	Eastman and Ferguson (1981)	Higgins <i>et al.</i> (1982)
Volatile solids	79.7	73.5	75.0
Lipids	18.6	21.0	10.3
Cellulose	18.2	19.9 ²	32.2
Hemicellulose	–	–	2.5
Lignin	–	–	13.6
Crude protein	17.2	28.7	19.0
Volatile acids	3.5 ¹	5.0	6.4 ³
Ash	20.3	26.5	25.0

¹Expressed as acetic acid, ²Measured as total carbohydrates, ³Total free fatty acids

2.3.2 Kinetics of Hydrolysis

The rate of hydrolysis has been shown to be dependent on a large number of factors. These include:

- Temperature. The rate of hydrolysis increases exponentially with an increase in temperature (Gujer and Zehnder, 1983).
- pH. Near neutral condition results in better hydrolysis than acid conditions. (Eastman and Ferguson, 1981).
- Microbial biomass and hence the level of hydrolytic enzymes.
- Particle geometry (surface area and size).
- The different rates of hydrolysis for the lipid, carbohydrate and protein fractions.
- The fact that the various components may be intimately bound.

The last two points are significant in that a slowly degrading component may shield an easily degraded component from enzymatic attack. In particular the lipid portion which degrades slowly may protect the protein and carbohydrate fractions.

Typically a first order function is used to model anaerobic sludge hydrolysis (Eastman and Ferguson, 1981; Lilley *et al.*, 1990; Elisosov and Argaman, 1995; Shimizu *et al.*, 1993), although other expression such as the Contois model have been used to describe hydrolysis in aerobic systems (Dold *et al.*, 1980). The first order approach has been shown to fit experimental data extremely well and it is doubtful if a more complicated expression is necessary. As Eastman and Ferguson have put it “The first-order hydrolysis function is an empirical expression that reflects the cumulative effects of all the microscopic processes occurring in the digesters.”

As discussed in section 2.2.1 the first order expression describing the rate degradation of a component of the sludge r_X ($\text{mg l}^{-1}\text{d}^{-1}$) is given by a form of equation 2.2:

$$r_X = -k_h(X - X_\infty) \quad (2.16)$$

$$= -k_h X_{deg} \quad (2.17)$$

where k_h is the overall hydrolysis rate constant (d^{-1}), X is the total concentration of a sludge component, X_∞ is the concentration of the 'nondegradable' fraction and X_{deg} that of the degradable fraction. (All concentrations in mg/l). The rate of hydrolysis is hence a function of the *degradable* particulate matter remaining and not of the total particulate matter remaining. Failure to account for the nondegradable portion of the substrate will lead to erroneously reduced rates (Pavlostathis and Giraldo-Gomez, 1991).

The nondegradable portion reported by workers differs (See table 2.3). It is suggested that several factors will influence what portion of the sludge can be regarded as degradable. These are:

- pH.
- Temperature.
- Whether the sludge component is genuinely nondegradable or just very slowly degradable. For example some workers (e.g. Eastman and Ferguson, 1981) regard the lipid portion as nondegradable while others (e.g. O'Rourke, 1968) regard it as a slowly degradable component.
- Sludge composition. As mentioned this is complicated by the fact that a nondegradable or slowly degradable component may protect a faster degrading component from hydrolysis.

Therefore, in any numerical comparison of the data of various workers both the hydrolysis rate constant and the portion of sludge reported as being nondegradable are required.

First order sludge hydrolysis kinetic data reported in the literature may broadly be divided into three categories:

- Rate data for the hydrolysis of sludge as a whole.
- Rate data for subportions (e.g. the lipid fraction) determined from the hydrolysis of sludge.
- Rate data for subportions but determined from artificial single component 'sludges'.

The last approach is not likely to be of much use since the effect of sludge morphology is not taken into account. The second approach has the advantage that more precise stoichiometric relationships linked to the rates of hydrolysis may be formulated.

First order hydrolysis rate data for a number of different sludge types, as taken from a broad selection of the literature, are given in table 2.3.

2.3.3 Stoichiometry of Hydrolysis

The poorly defined nature of sludge makes it difficult to write definite stoichiometric relationships for its hydrolysis. Although broadly sludge is said to consist of lipid, protein and carbohydrate fractions, within these fractions numerous different molecular species can be expected to occur. One solution to this problem is to use

a pseudo (or real) molecule to represent the entire fraction. For example the protein fraction has variously been represented by $C_{16}H_{30}O_8N_4$ (Vasiliev *et al.*, 1993), $C_{39}H_{63}O_{13}N_{10}$ (Shimizu *et al.*, 1993) and $(C_4H_6O_2N)_n$ (Eastman and Ferguson, 1981). Since the ultimate products of protein hydrolysis are amino acids, a better generic formula for protein would be one built from an 'average' amino acid. An average of the twenty amino acids gives the formula as approximately $(C_5H_9O_3N)_n$. (A better approach would be to use a weighted average based on an amino acid analysis of sludge). Protein hydrolysis is then represented by the reaction:

In a similar manner $(C_6H_{10}O_5)_n$ (i.e. starch or cellulose) may be taken as the formula for the carbohydrate fraction, the hydrolysis of which would then be given by:

Since the lipid fraction consists mostly of triglycerides a suitable generic triglyceride formula can be used. For example Eastman and Ferguson (1981) have used palmitic acid as the basis of the lipid fraction. McMurry (1992) gives palmitic acid, stearic acid, oleic acid and linoleic acid as being the most abundant naturally occurring fatty acids. Since the molecular formulae of these compounds are reasonably similar, a triglyceride of one of these can be taken as being representative of the degradable lipid fraction. If palmitic is used, the stoichiometric reaction of lipid hydrolysis is:

2.4 Fermentation (Acidogenesis)

The amino acids, sugars and polyols resulting from hydrolysis are broken down further by fermentation or acidogenesis. The main products of this are: hydrogen; carbon dioxide; and the short chain fatty acids, lactate, butyrate, propionate and acetate. Other higher order acids such as valeric acid and capronic acid are also produced to a lesser extent.

2.4.1 Glucose Fermentation

Substrate Uptake and Hydrogen Inhibition and Product Regulation

Acid forming bacteria ferment glucose to produce a mixture of acetic, propionic, butyric and lactic acids. The respective stoichiometric reactions are:

The first reaction is favoured by the bacteria as it provides the largest energy yield (Mosey, 1983). The other reactions are said to occur as a response to elevated hydrogen concentrations. A convincing theory to account for this has been proposed by Mosey (1983) with modifications by Costello *et al.* (1991a,b). The essence of the theory is that the different cell pathways metabolizing the substrate are regulated by the relative concentrations of NADH and NAD⁺. By assuming a constant internal cell pH of 7, and using the Nerst equation, Mosey was able to related the ratio of the two concentrations to the hydrogen partial pressure. His equation is: $[\text{NADH}]/[\text{NAD}^+] = 1500P_{\text{H}_2}$.

In Mosey's scheme an increase in hydrogen partial pressure shifts the fermentation reaction away from the production of acetic acid towards the production of butyric and propionic acids. In addition to this, elevated hydrogen levels result in a decreased rate of glucose uptake. The rate of glucose uptake for *energy production only*, R_{Gl} ($\text{mg l}^{-1} \text{d}^{-1}$) is given by a noncompetitive inhibition model:

$$R_{Gl} = -\frac{kXS}{K_S + S} \frac{1}{1 + \frac{[\text{NADH}]}{[\text{NAD}^+]}} \quad (2.25)$$

$$= -\frac{kXS}{K_S + S} \frac{1}{1 + 1500P_{\text{H}_2}} \quad (2.26)$$

$$= -\frac{kXS}{K_S + S} \frac{K_{I,\text{H}_2}}{K_{I,\text{H}_2} + P_{\text{H}_2}} \quad (2.27)$$

where the symbols have the definitions as given in section 2.2.2. The rate constant k is used rather than the standard μ due to the varying biomass yield. This is explained in more detail below. The reported values of the kinetic constants are $k = 268 \text{ mg glucose} \cdot \text{mg biomass} \cdot \text{d}^{-1}$, $K_S = 23 \text{ mg glucose/l}$ and $b = 0.02 \text{ d}^{-1}$.

The rates of acid production are given as functions of the glucose uptake rate, and of the NADH/NAD⁺ ratio or hydrogen partial pressure (in atm):

$$r_{\text{AcH}} = \frac{2R_{Gl}}{\left(1 + \frac{[\text{NADH}]}{[\text{NAD}^+]}\right)^2 \left(1 + \frac{[\text{NAD}^+]}{[\text{NADH}]}\right)} - \frac{2R_{Gl}}{\left(1 + \frac{[\text{NADH}]}{[\text{NAD}^+]}\right)^2} \quad (2.28)$$

$$= \frac{2R_{Gl}}{\left(1 + \frac{P_{\text{H}_2}}{K_{I,\text{H}_2}}\right)^2 \left(1 + \frac{K_{I,\text{H}_2}}{P_{\text{H}_2}}\right)} - \frac{2R_{Gl}}{\left(1 + \frac{P_{\text{H}_2}}{K_{I,\text{H}_2}}\right)^2} \quad (2.29)$$

$$r_{\text{PrH}} = \frac{-2R_{Gl}}{\left(1 + \frac{[\text{NADH}]}{[\text{NAD}^+]}\right) \left(1 + \frac{[\text{NAD}^+]}{[\text{NADH}]}\right)} \quad (2.30)$$

$$= \frac{-2R_{Gl}}{\left(1 + \frac{P_{\text{H}_2}}{K_{I,\text{H}_2}}\right) \left(1 + \frac{K_{I,\text{H}_2}}{P_{\text{H}_2}}\right)} \quad (2.31)$$

$$r_{BuH} = \frac{-R_{Gl}}{\left(1 + \frac{[NADH]}{[NAD^+]}\right)^2 \left(1 + \frac{[NAD^+]}{[NADH]}\right)} \quad (2.32)$$

$$= \frac{-R_{Gl}}{\left(1 + \frac{P_{H_2}}{K_{I,H_2}}\right)^2 \left(1 + \frac{K_{I,H_2}}{P_{H_2}}\right)} \quad (2.33)$$

In all equations the inhibition constant K_{I,H_2} is equal to 1500^{-1}atm^{-1} . Alternatively the temperature dependency of this constant can be calculated from the following expression derived by Costello *et al.*:

$$\log_{10}(K_{I,H_2}) = \frac{1139}{T} - 7 \quad (2.34)$$

where T is the temperature in Kelvin.

The scheme of Costello *et al.* differs in that they assume that acetic acid, butyric acid and lactic acid (and not propionic acid) are the three products of glucose fermentation. They also assume a second group of bacteria responsible for the further breakdown of lactic acid to either acetic acid (under low hydrogen conditions) or propionic acid (under high hydrogen conditions). Analyzing the metabolic pathways and relating the NADH/NAD⁺ ratio to the hydrogen partial pressure in the same manner as Mosey, they obtain the same expression for glucose uptake (equation 2.27) and the following expressions for the production of volatile fatty acids:

$$r_{AcH} = \frac{-2R_{Gl}}{\left(1 + \frac{P_{H_2}}{K_{I,H_2}}\right)^2} \quad (2.35)$$

$$r_{BuH} = \frac{-R_{Gl} \frac{P_{H_2}}{K_{I,H_2}}}{\left(1 + \frac{P_{H_2}}{K_{I,H_2}}\right)^2} \quad (2.36)$$

$$r_{LaH} = \frac{-2R_{Gl} \frac{P_{H_2}}{K_{I,H_2}}}{\left(1 + \frac{P_{H_2}}{K_{I,H_2}}\right)} \quad (2.37)$$

Substrate Uptake without Hydrogen Regulation

Alternatively, if the hydrogen partial pressure remains low normal Monod kinetics can be used to describe glucose fermentation. Zoetemeyer *et al.* (1982a) report $\mu_{max} = 7.9 \text{ d}^{-1}$ and $Y = 0.134$ giving $k = 59 \text{ mg glucose} \cdot \text{mg biomass}^{-1} \cdot \text{d}^{-1}$; and Moletta *et al.* (1986) give $\mu_{max} = 1.5 \text{ d}^{-1}$ and $Y = 0.82$ giving $k = 1.8 \text{ mg glucose} \cdot \text{mg biomass}^{-1} \cdot \text{d}^{-1}$.

Acid Product Inhibition

A further refinement to the kinetics of the acidogenesis of glucose is the inclusion of the inhibitory effects of the acids produced. The proposed mechanism assumes that only the *undissociated* acids are freely permeable to the cell membrane (Zoetemeyer *et al.*, 1982b). Once inside the cell the acids lower the pH and disrupt cell functioning. Since energy is required to expel the acids a lower growth rate results.

Costello *et al.* found that a noncompetitive model described the effects of product inhibition in acidogenesis. Equation 2.27 should then be modified to:

$$R_{GI} = -\frac{kXS}{K_S + S} \frac{K_{I,VFA}}{K_{I,VFA} + I_{VFA}} \frac{K_{I,H_2}}{K_{I,H_2} + P_{H_2}} \quad (2.38)$$

where I_{VFA} is the total concentration of all undissociated short chain fatty acids and $K_{I,VFA}$ is the inhibition constant (in molar rather than the usual mass units). It is assumed here that a single expression may be used to account for the effect all volatile fatty acids since the dissociation constants for the acids are similar. A value of $K_{I,gF,VFA} = 10$ mmole/l is reported by Costello *et al.*

Using the notation introduced earlier, equation 2.38 can be shortened to:

$$R_{GI} = \xi_{SU,GI} \cdot \xi_{NC,VFA} \cdot \xi_{NC,H_2} \quad (2.39)$$

pH Inhibition

Zoetemeyer *et al.* (1982a) measured the maximum specific growth rate of acidogenic bacteria fed glucose over a pH range of 4.5 to 7.9. They found that the optimal pH was around 6.0 with the maximum specific growth rate rapidly decreasing to 50% at pH 5 and gradually decreasing to 25% at pH 8.0. Hilton and Oleszkiewicz (1988) report a similar trend over a pH range of 6.0 to 8.0 for the utilization of lactose (a galactose-glucose disaccharide). Based on the data of Zoetemeyer *et al.* the parameters for the pH inhibition function as defined in equation 2.14 are estimated as $\alpha_{LL} = 4$, $\alpha_{UL} = 0.5$, $pH_{LL} = 5$ and $pH_{UL} = 10.5$. A plot of this function is given in figure 2.6 and shows a reasonable approximation to that given by Zoetemeyer *et al.* Equation 2.39 is then further modified to include pH inhibition to give:

$$R_{GI} = \xi_{SU,GI} \cdot \xi_{NC,VFA} \cdot \xi_{NC,H_2} \cdot \xi_{pH} \quad (2.40)$$

Hydrogen Sulphide Inhibition

Hilton and Oleszkiewicz (1988) examined the effect of sulphide concentration on lactose uptake over a pH range of 6.0 to 8.0. They concluded that unionized H_2S was inhibitory but did not report inhibition constants. They also found that the optimal pH shifted to around 7.0 in the presence of sulphide. This is due to the lower fraction of unionized H_2S at this pH. Hence in modelling a high sulphate reducing system the effect of sulphide concentration on sugar uptake needs to be included. A term for sulphide inhibition should be incorporated in equation 2.40.

Cell Synthesis

Due to the different amounts of energy produced in each pathway the biomass yield will vary. Using the scheme of Costello *et al.*, the overall rate of biomass production of glucose fermenters can be calculated from:

$$r_{gF} = Y'_{AcH} \cdot r_{AcH} + Y'_{BuH} \cdot r_{BuH} + Y'_{LaH} \cdot r_{LaH} \quad (2.41)$$

Mosey and Costello *et al.* estimated the yield coefficients Y'_i from a rule of thumb which states that approximately 10g of biomass are produced for each mole of ATP

Figure 2.6: Plot of pH inhibition of glucose fermentation based on data from Zoetemeyer *et al.* (1982a). $\alpha_{LL} = 4$, $\alpha_{UL} = 0.5$, $pH_{LL} = 5$ and $pH_{UL} = 10.5$.

generated (Bauchop and Elsdén, 1960). The respective yields given by these workers for reactions 2.21 to 2.24 are four, three, two and two ATP molecules per molecule of glucose consumed. These values correspond to product based yields of $Y'_{AcH} = 0.33$, $Y'_{PrH} = 0.13$, $Y'_{BuH} = 0.34$ and $Y'_{LaH} = 0.11$.

Assuming a cell formula of $C_5H_7O_2N$ the reaction describing the production of biomass from glucose is:

2.4.2 Lactic Acid Fermentation

Substrate Uptake and Hydrogen Inhibition and Regulation

Lactic acid has been shown to be a major intermediate in anaerobic digestion (Zeller *et al.*, 1994). In the model of Costello *et al.*, lactic acid produced by glucose fermentation is broken down to different ratios of acetic and propionic acids depending on the hydrogen partial pressure. The reactions given are:

Using a similar argument to that applied to glucose fermentation, the hydrogen regulated kinetics of lactate uptake (for energy production only) and acetic and propionic acid production were derived:

$$R_{LaH} = -\frac{kXS}{K_S + S_{LaH}} \frac{1}{1 + P_{H_2}/K_{I,H_2}} \quad (2.45)$$

$$\tau_{PrH} = -\frac{R_{LaH}P_{H_2}/K_{I,H_2}}{1 + P_{H_2}/K_{I,H_2}} \quad (2.46)$$

$$r_{AcH} = -\frac{R_{LaH}}{1 + P_{H_2}/K_{I,H_2}} \quad (2.47)$$

The kinetic constants used in the model were $k = 46$ mg glucose · mg biomass · d⁻¹, $K_S = 34$ mg/l and $b = 0.02$ d⁻¹. As before the inhibition constant K_{I,H_2} is calculated using equation 2.34.

Substrate Uptake without Hydrogen Regulation

If hydrogen levels can be assumed to be low the Monod equation on its own can be used. Zeller *et al.* (1994) give Monod kinetic parameters of $\mu_{max} = 16.8$ d⁻¹ and $K_S = 225$ mg/l for *Clostridium propionicum* growing on lactate.

Acid Product Inhibition

As with glucose fermentation, lactate acidogenesis is inhibited by undissociated fatty acids. As before a noncompetitive model has been used (Costello *et al.*, 1991a,b). Equation 2.45 should therefore be multiplied by a suitable factor ($\xi_{NC,VFA}$). Costello *et al.* used a value of $K_{I,VFA} = 10$ mmole/l.

pH Inhibition

Costello *et al.* assume that since the lactate acidogenic bacteria are in close symbiosis with the glucose acidogens that the optimal pH ranges must be similar. Based on this assumption, the pH inhibition parameters estimated for glucose fermenters may be used to calculate ξ_{pH} for lactate fermenters.

Hydrogen Sulphide Inhibition

No specific mention of hydrogen sulphide inhibition of lactate fermenting bacteria has been found in the literature. However, based on the fact that sulphide has been shown to be inhibitory to almost all of the other microbial processes occurring in an anaerobic system, it is reasonable to assume that lactate fermenters will be similarly effected. A suitable inhibition term should hence be included when calculating the uptake rate of lactate.

Cell Synthesis

Based on ATP yields of 0.5 and 1.0 mole per mole lactate consumed for reactions 2.43 and 2.44 respectively (Costello *et al.*, 1991a), the biomass product based yields are estimated as $Y'_{PrH} = 0.067$ and $Y'_{AcH} = 0.17$. The biomass synthesis reaction is:

2.4.3 Amino Acid Fermentation

Assuming that the formula $\text{C}_5\text{H}_9\text{O}_3\text{N}$ is a valid approximation for the average of all amino acids produced in hydrolysis, the reactions for the production of the four principle fatty acids are:

No data for the kinetics of amino acid acetogenesis are available in the literature. In the absence of any useful data, a rate similar to that of glucose fermentation can be tentatively assumed. Alternatively, since protein hydrolysis has been shown to be rate limiting in the conversion of protein to acids, a rate constant at least one order of magnitude greater than that for hydrolysis could be assumed.

Of the four reactions given above, only reaction 2.51 does not result in the production of hydrogen. Therefore, it could be assumed that a shift to butyrate production would result from an increase in hydrogen partial pressure. However, in the absence of any evidence it is simpler to assume a fixed ratio of acid production. In the study of Eastman and Ferguson (1981) on the hydrolysis and acidogenesis of primary sludge, it was found that the production of acetic and propionic acids were roughly equal (on a COD basis). Although their work dealt with a mixture on carbohydrate and nitrogenous material, it is assumed here that this ratio will also be valid for the acidogenesis of amino acids alone.

The relevant biomass synthesis equation is:

and a yield of 0.48 g cell COD/g COD utilized is assumed (Eastman and Ferguson, 1981). Based on a cell COD of 1.41 and a generic amino acid COD of 1.22, this corresponds to $Y = 0.55$.

2.4.4 Glycerol Fermentation

For the sake of completeness the fermentation of the small amount of glycerol generated by lipid hydrolysis needs to be accounted for. Since no data exist, the rate and yield are assumed to be the same as for glucose. The acidogenesis and cell synthesis reactions are (assuming for the sake of simplicity that acetic acid is the sole product):

2.5 Beta Oxidation of Long Chain Fatty Acids

According to *Knoop's Theory* (Sawyer and McCarty, 1989) the breakdown of long chain fatty acids occurs by oxidation of the beta carbon atom, resulting in the formation of acetic acid and hydrogen. The process is repeated and the fatty acid is shortened by two carbon atoms in each step. If the molecule has an even number of carbon atoms only acetic acid results. An odd numbered molecule will result in the formation of both acetic and propionic acids (McInerney and Bryant, 1981). The general stoichiometry for beta oxidation as given by Gujer and Zehnder (1983) is:

Table 2.4: Monod kinetic parameters for long chain fatty acid beta oxidation at 37°C. Values from Gujer and Zehnder (1983) based on data from Novak and Carlson (1970). Yields and decay rates averaged for all experiments. Yield values may include methanogenic biomass.

Fatty acid	μ_{max} d ⁻¹	Y mg biomass/ mg fatty acid	K_s mg acid/l	b d ⁻¹
Stearic	0.10	~ 0.3	143	~ 0.01
Palmitic	0.12	~ 0.3	49.8	~ 0.01
Myristic	0.11	~ 0.3	37.5	~ 0.01
Oleic	0.45	~ 0.3	1116	~ 0.01
Linoleic	0.56	~ 0.3	637	~ 0.01

For the specific case of palmitic acid the overall reaction is:

Monod kinetic parameters for the breakdown of a number of LCFAs, as calculated by Gujer and Zehnder (1983) based on the data of Novak and Carlson (1970), are given in table 2.4.

The large amount of hydrogen generated by beta oxidation has been shown to be inhibitory (Novak and Carlson, 1970). A noncompetitive model has been shown to describe the behaviour of hydrogen inhibition of acetogenesis (See below). Since beta oxidation of LCFAs is closely related to acetogenesis of SCFAs, it is hypothesized that a noncompetitive model is also valid here. It is further assumed that the internal cell mechanisms are similar enough to allow the calculation of the inhibition constant from equation 2.34.

If palmitic acid is taken as representative of all the LCFA present, the relevant cell synthesis reaction is:

2.6 Acetogenesis (Anaerobic Oxidation of Short Chain Fatty Acids)

2.6.1 Basic Kinetics and Stoichiometry

The process by which intermediate short chain fatty acids are degraded to acetate and hydrogen is termed acetogenesis. A number of bacteria capable of degrading butyrate and higher fatty acids have been identified (e.g. *Syntrophomonas wolfei* and *Syntrophomonas sapovorans*), however to date only one acetogenic species capable of degrading propionate (and only propionate) has been identified, *Syntrophobacter wolinii* (McCarty and Mosey, 1991). Hence, for the purpose of modelling acetogenesis these two groups should be kept separate.

Propionate Substrate

The reactions describing the anaerobic oxidation of propionate (McCarty and Mosey, 1991) and the production of biomass are:

A summary of Monod kinetic constants for propionate oxidation, compiled from a literature survey, is given in table 2.5.

Butyrate and Higher Fatty Acids

Butyrate oxidation and biomass production are represented by:

Kinetic constants for the anaerobic oxidation of butyrate, as taken from the literature, are given in table 2.6.

2.6.2 Inhibition Kinetics

Hydrogen Inhibition

Both the oxidation and biomass synthesis reactions result in the production of hydrogen. The oxidation reactions are thermodynamically unfavourable, and can only proceed if accompanied by efficient hydrogen removal (Mosey, 1983; Pavlostathis and Giraldo-Gomez, 1991). Using the same reasoning as applied to the metabolism of glucose, Mosey determined that hydrogen regulation of propionate and butyrate uptake could be accounted for by a noninhibition model. As before the inhibition constants are calculated from equation 2.34.

Fatty Acid Inhibition

As with acidogenic bacteria, high unionized fatty acid levels have been shown to be inhibitory to acetogens (Denac, 1986). Costello *et al.* (1991a,b), based on the results of Denac, used a competitive inhibition model to account for this, reporting inhibition constants of 3.0 and 30.0 mmole/l for propionate and butyrate utilizing acetogens respectively.

pH Inhibition

To date, no specific studies have been conducted to determine the optimal pH range for acetogenic bacteria. Costello *et al.* have argued that, in a two stage digester with the stages operated at pH 6 and pH 7 respectively, due to the high hydrogen partial pressure in the first stage, acetogenic bacteria are more likely to predominate

Table 2.5: Monod kinetic parameters for propionate utilizing acetogens. Adapted from Pavlostathis and Giraldo-Gomez (1991) and Oude Elferink *et al.* (1994).

Reference	Culture	T °C	Y mg biomass/ mg propionate	μ_{max} d ⁻¹	K_S mg PrH/l	K_{I,H_2S} mg H ₂ S/l	b d ⁻¹
Lawrence and McCarty (1969)	Propionic acid utilizing	25	0.077 ¹	0.36	40		0.04
Lawrence and McCarty (1969)	Propionic acid utilizing	35	0.064 ¹	0.31	758		0.01
Heyes and Hall (1983)	Propionic acid utilizing	35		0.13–1.20	11–331		
Maillacheruvu and Parkin (1996)	Propionic acid utilizing		0.063	0.15 ²	17.9	26.6 ³	0.021
Mosey (1983)	Propionic acid utilizing		0.13 ⁴				
Dörner (1992); Boone and Bryant (1980)	<i>Syntrophobacter wolinii</i>			0.02–0.21			
Stams <i>et al.</i> (1993)	Strain MPOB			0.15–0.17			
Dörner (1992)	Strain KoProp1			0.07			
Wu <i>et al.</i> (1992)	Culture PT			0.01			
Wu <i>et al.</i> (1992)	Culture PW			0.14			

¹Includes biomass formed in methanogenesis, ²Reported as k , ³Uncompetitive Inhibition, ⁴Based on ATP yield

Table 2.6: Monod kinetic parameters butyrate utilizing acetogens. Adapted from Pavlostathis and Giraldo-Gomez (1991) and Oude Elferink *et al.* (1994).

Reference	Culture	T °C	Y mg biomass/ mg butyrate	μ_{max} d ⁻¹	K_S mg BuH/l	b d ⁻¹
Lawrence and McCarty (1969)	Butyric acid utilizing	35	0.085 ¹	0.37	7.2	0.027
Massey and Pohland (1978)	Butyric acid utilizing	37		0.86	164	
Mosey (1983)	Butyric acid utilizing		0.13 ²			
Wu <i>et al.</i> (1992); Stieb and Schink (1985); Zhao <i>et al.</i> (1990)	<i>Syntrophospora (Clostridium) bryantii</i> BH			0.252		
Roy <i>et al.</i> (1986)	<i>Syntrophomonas sapovorans</i>			0.6		
McInerney <i>et al.</i> (1981); McInerney <i>et al.</i> (1979); Dörner (1992)	<i>Syntrophomonas wolfei</i>			0.19–0.31		
Zhao <i>et al.</i> (1993)	<i>Syntrophomonas</i> strain FSM2			0.32		
Zhao <i>et al.</i> (1993)	<i>Syntrophomonas</i> FSS7			0.34		
Zhao <i>et al.</i> (1993)	<i>Syntrophomonas</i> FM4			0.24		

¹Includes biomass formed in methanogenesis, ²Based on ATP yield

in the second methanogenic stage. They therefore assume that selection pressure would favour acetogenic bacteria with the same optimal pH range as methanogenic bacteria. While their argument is not complete, in the absence of any better data the same pH inhibition function as that for methanogenesis (see below) can be assumed.

Hydrogen Sulphide Inhibition

Maillacheruvu and Parkin (1996) found that an uncompetitive inhibition model best described the inhibition of propionate oxidation due to unionized hydrogen sulphide. The reported inhibition constant is 26.6 mg H₂S/l. No sulphide inhibition constants for butyrate and higher fatty acid utilizing acetogens are to be found in the literature.

2.7 Homoacetogenesis

Homoacetogenesis refers to the production of acetic acid from carbon dioxide and hydrogen. The reaction given by McCarty and Mosey (1991) is:

A suitable biomass synthesis reaction is:

No rate data for homoacetogenesis are to be found in the literature. According to Nozhevnikova and Kotsuyrbenko (1995), homoacetogenesis is only significant in relation to hydrogen consuming methanogenesis at temperatures below 20°C.

2.8 Methanogenesis

2.8.1 Basic Kinetics and Stoichiometry

In an anaerobic system the ultimate sink for carbon is methane. Although a number of species are methanogenic, only acetic acid or carbon dioxide and hydrogen are able to be utilized as substrates.

Hydrogen/Carbon Dioxide Substrates

Methanogenesis utilizing H₂ and CO₂ can be represented by the reaction:

and using cell synthesis by:

A large amount of kinetic data, describing methanogenesis using hydrogen and carbon dioxide as substrates, is present in the literature. A summary of Monod kinetic parameters as taken from a number of sources is given in table 2.9. The values have been converted to a common unit basis. In some cases some assumptions have had to be made for this to be possible. These are recorded in the footnotes.

Table 2.7: Monod kinetic parameters for methanogenesis using hydrogen as substrate. Adapted from Oude Elferink *et al.* (1994), Pavlostathis and Giraldo-Gomez (1991) and Harper and Pohland (1986).

Reference	Culture	Conditions	T °C	Y mg biomass/ mg hydrogen	μ_{max} d ⁻¹	K_S $\mu\text{g H}_2/\text{l}$	$K_{I, \text{H}_2\text{S}}$ mg H ₂ S/l	b d ⁻¹
Robinson and Tiedje (1984)	<i>Methanospirillum hungatei</i> JF-1	Batch	37	0.16–0.24 ¹	1.24–1.29	11.7–14.7		
Wu <i>et al.</i> (1992)	<i>Methanospirillum hungatei</i> BD				2.4–2.8			
Robinson and Tiedje (1984)	Mean of several species				1.3	13		
Hungate (1967)	Rumen bacteria		37		5.4	0.03		
Robinson and Tiedje (1982)	Digesting sludge					0.03		
Kaspar and Wuhrmann (1978)	Digesting sludge					166 ⁴		
Kristjansson <i>et al.</i> (1982)	<i>Methanobrevibacter arboriphilus</i> AZ	Continuous	35			12.1		
Zehnder and Wuhrmann (1977)	<i>Methanobrevibacter arboriphilus</i> AZ			0.3–0.35	1.4–3.4			
Zeikus and Henning (1975)	<i>Methanobrevibacter arboriphilus</i> DH1			0.5–0.6				
Pavlostathis <i>et al.</i> (1990)	<i>Methanobrevibacter smithii</i>		37	0.36	4.02	2.3		
Archer and Powell (1985)	<i>Methanobrevibacter smithii</i>		37		4.07			
Maillacheruvu and Parkin (1996)	Enriched MPB	Batch		0.39 ²	0.18 ^{2,3}	30 ^{2,4}	664 ²	0.013
Weimer and Zeikus (1978b)	<i>Methanosarcina barkeri</i> MS			0.8	1.4			
Smith and Mah (1978)	<i>Methanosarcina barkeri</i> 227			1.1				
Wu <i>et al.</i> (1992)	<i>Methanosarcina mazei</i> T18				1.4–1.7			
Jain <i>et al.</i> (1987)	<i>Methanobacterium ivanovii</i>			0.54	0.8–1.7			
Wu <i>et al.</i> (1992)	<i>Methanobacterium formicicum</i> T1N				2.1–2.8			
Schauer and Ferry (1980)	<i>Methanobacterium formicicum</i> JF1			0.4	1.2–2.8			
Jain <i>et al.</i> (1987)	<i>Methanobacterium bryantii</i> MOH			0.3	0.3–0.4			

¹Based on cell protein content of 0.5 g protein/g dry biomass, ² Uncompetitive inhibition, ³Reported as k , ⁴Converted using Henry's Law

Acetate Substrate

The overall reaction, for the biological production of methane from acetate, is given by:

Biomass synthesis can be described by:

As with hydrogen utilizing methanogenesis, a large amount of kinetic data can be found in the literature. This is summarized in table 2.8.

2.8.2 Inhibition Kinetics

Sulphide Inhibition

The inhibitory effect of sulphide on methanogenesis has been well documented. Hydrogen sulphide levels resulting in a 50% inhibition of methanogenesis range from 50 mg/l to 250 mg/l (Visser, 1995). Hilton and Oleszkiewicz (1988) studied the effect of sulphide on methanogenesis utilizing acetate as a substrate over a pH range of 6.5 to 8.0. They concluded that it is the unionized H_2S and not total sulphide which is inhibitory. They were also able to confirm the generally accepted level of 200 mg/l H_2S , above which methanogenesis is severely or completely retarded. Maillacheruvu and Parkin (1996) have reported kinetic constants for sulphide inhibition for a number of anaerobic reactions including methanogenesis (both acetate and H_2/CO_2 substrates). They concluded that an uncompetitive inhibition model gives the best fit to the data. The constants reported by these workers are given in tables 2.8 and 2.9.

Acid Inhibition

Another factor, which has been shown to be inhibitory to methane production, is the presence of high concentrations of unionized acetic and other volatile fatty acids. In their general model of an anaerobic digester, Moletta *et al.* (1986) used a noncompetitive inhibition model to describe the kinetics of acetate substrate methanogenesis. The reported inhibition constant is 40 mg AcH/l.

pH Inhibition

In contrast to acidogenic bacteria which have an optimal growth at pH 6.0, methanogenic bacteria are seriously inhibited below pH 6.5 (Sawyer and McCarty, 1989). Huser *et al.* (1982) report the optimal pH to be 7.4–7.8, with complete cessation of methane production below pH 6.8 and above pH 8.2. Visser (1995) reports similar results but notes that granular sludge is more tolerant of pH extremes. This is presumably because of concentrations gradients within the particles.

The narrow pH range indicates a rapid onset of inhibition, and consequently the inhibition parameters α_{LL} and α_{UL} should be large (Around 20 say). pH_{LL} and pH_{UL} were chosen to be 7.1 and 8.0. The resulting empirical plot of the pH inhibition function (figure 2.7) reflects the behaviour observed by Visser and Huser *et al.*

Table 2.8: Monod kinetic parameters for methanogenesis using acetate as substrate. Adapted from Harper and Pohland (1986), Gujer and Zehnder (1983) and Pavlostathis and Giraldo-Gomez (1991).

Reference	Culture	Conditions	T °C	Y mg biomass/ mg acetate	μ_{max} d ⁻¹	K_S mg acetate/l	K_{I,H_2S} mg H ₂ S/l	b d ⁻¹
Gupta <i>et al.</i> (1994a,b)	Mixed SRB and MPB	Continuous	35	0.0375 ¹		6.00		
Bhattacharya <i>et al.</i> (1996)	Mixed SRB and MPB	Batch	35	0.0346	0.112 ²	115.5		0
Visser <i>et al.</i> (1993)	Mixed SRB and MPB	UASB			0.055	0.07		
Maillacheruvu and Parkin (1996)	Enriched MPB	Batch		0.041 ³	0.14 ^{2,3}	27 ³	117 ³	0.013
Lawrence and McCarty (1969)	Enriched MPB	Continuous	25	0.054	0.250	869		0.011
	Enriched MPB	Continuous	30	0.058	0.270	333		0.037
	Enriched MPB	Continuous	35	0.04	0.357	154		0.015
	Enriched MPB	Continuous	37	0.04	0.39	168		0.02
Kugelman and Chin (1971)	Enriched MPB	Continuous	35		0.34	170		0.036
Moletta <i>et al.</i> (1986)	Mixed MPB	Batch	35	0.82	0.138	3		
van der Berg (1977)	Mixed MPB	Batch	20	0.05	0.13 ²			
		Batch	30	0.02	0.052–0.10 ²			
		Batch	35	0.02	0.052–0.10 ²			
Visser (1995)	Granular sludge	UASB	30		0.09–0.12	54		
Visser (1995)	Suspended sludge	UASB	30		0.03–0.09	29		
Wandrey and Aivasidis (1983)	<i>Methanosarcina barkeri</i>	Continuous	37	0.024	0.206	230		0.004
Schönheit <i>et al.</i> (1982)	<i>Methanosarcina barkeri</i>	Batch				180		
Weimer and Ziekus (1978a)	<i>Methanosarcina barkeri</i>		37	0.03				
Smith and Mah (1980)	<i>Methanosarcina barkeri</i>		36	0.05	0.44			
Smith and Mah (1978)	<i>Methanosarcina barkeri</i> 277		36	0.04	0.60	299		
Zinder and Mah (1979)	<i>Methanosarcina</i>		55	0.03	1.4	271		
Zehnder <i>et al.</i> (1980)	<i>Methanothrix soehngenii</i>		33	0.03	0.11	28		
Huser <i>et al.</i> (1982)	<i>Methanothrix soehngenii</i>		37	0.02	0.16	42		
Cappenburg (1975)	<i>Methanobacterium</i> species		30	0.01	0.26	10		

¹Based on biomass COD of 1.41 mg COD/mg biomass, ²Reported as k , ³Uncompetitive inhibition

Figure 2.7: Plot of pH inhibition of methanogenesis based on data from Huser *et al.* (1982). $\alpha_{LL} = 20$, $\alpha_{UL} = 20$, $pH_{LL} = 7.1$ and $pH_{UL} = 8.0$.

Absence of Hydrogen Inhibition

In contrast to the biological reactions discussed above, methanogenesis is not affected by elevated hydrogen concentrations (Mosey, 1983).

2.9 Sulphate Reduction

Sulphate reducing bacteria have been shown to be able to utilize a large number of substrates as electron donors. These include, hydrogen, volatile fatty acids up to C_{20} , alcohols, several amino acids, monomeric sugars and a large number of aromatic compounds (Hansen, 1993). However in the sort of digester under consideration, many of the above components may not be present in significant quantities (e.g. aromatic compounds). In addition the sulphate reducers may favour certain substrates, or may not be able to compete with other organisms utilizing the same substrates. Consequently, in the development of the model only hydrogen and some of the short chain fatty acids were be considered as substrates.

2.9.1 Basic Kinetics and Stoichiometry

Hydrogen Substrate

Along with acetate, hydrogen is probably the most important electron donor for sulphate reducing organisms. While both incompletely and completely oxidizing sulphate reducers are able to utilize hydrogen as an electron donor, only the complete oxidizers are capable of true autotrophic growth on H_2/CO_2 (Colleran *et al.*, 1995). The energy producing reaction is:

and the reaction for autotrophic cell synthesis utilizing CO_2 as the carbon source:

Growth kinetic constants obtained from literature are given in table 2.9.

Acetate Substrate

The reactions describing sulphate reduction and biomass production, using acetic acid as the electron donor and carbon source are:

Monod kinetic parameters as taken from the literature are given in table 2.9.

Propionate Substrate

Propionate can be utilized by both completely and incompletely oxidizing sulphate reducers. The reactions for complete and incomplete oxidation are respectively (Visser, 1995):

However the growth rates of complete oxidizers utilizing propionate is low (Colleran *et al.*, 1995); and from thermodynamic analysis incomplete propionate oxidation is expected to be the preferred pathway (Maillacheruvu and Parkin, 1996). Consequently only incomplete oxidation is expected to be of any importance.

The proposed biomass synthesis reaction is:

It should be noted that sulphate reduction has been incorporated in the cell synthesis reaction. If sulphate reduction is not included, it is impossible to write a synthesis reaction which does not result in the formation of hydrogen. This is unlikely to occur in reality.

Monod kinetic constants are shown in table 2.11.

Lactate Substrate

The reactions for incomplete oxidation and cell synthesis utilizing lactate as a substrate are:

Kinetic parameters taken from literature are given in table 2.12.

Table 2.9: Monod kinetic parameters for sulphate reduction using hydrogen as substrate. Table in part taken from Oude Elferink *et al.* (1994).

Reference	Culture	Conditions	T °C	Y mg biomass/ mg hydrogen	μ_{max} d ⁻¹	K_S μ g H ₂ /l	K_{I,H_2S} mg H ₂ S/l	b d ⁻¹
Robinson and Tiedje (1984)	<i>Desulfovibrio</i> G11	Batch	37	0.70–0.98 ¹	1.17–1.57	4.88–8.41		
Robinson and Tiedje (1984)	Mean of several species			0.84 ¹	1.4	6.7		
Maillacheruvu and Parkin (1996)	Enriched SRB	Batch		0.33	0.18 ²	25	149 ³	0.013
Kristjansson <i>et al.</i> (1982); Badziong and Thauer (1978); Badziong <i>et al.</i> (1978); Brandis and Thauer (1981)	<i>Desulfovibrio vulgaris</i> (Marburg)			0.55–1.3	3.6–5.5			
Badziong <i>et al.</i> (1978); Lupton and Zeikus (1984)	<i>Desulfovibrio vulgaris</i> (Madison)			0.3–0.55	0.7–5.5			
Brandis and Thauer (1981)	<i>Desulfovibrio desulfuricans</i>			0.94	2.6			
Nanninga and Gottschal (1987)	<i>Desulfovibrio desulfuricans</i>				2.8–4.3			
Widdel (1987)	<i>Desulfobacter hydrogenophilus</i>				0.98			
Brysch <i>et al.</i> (1987)	<i>Desulfobacterium autotrophicum</i>				0.83–1.04			
Widdel and Pfennig (1982)	<i>Desulfobulbus propionicus</i> lpr3				1.66			
Nanninga and Gottschal (1987)	<i>Desulfobulbus propionicus</i>				0.23–0.59			

¹Based on cell protein content of 0.5 g protein/g dry biomass, ²Reported as k , ³Uncompetitive inhibition,

Table 2.10: Monod kinetic parameters for sulphate reduction using acetate as substrate. Experimental conditions as follows: C Chemostat, B Batch, CL Carbon Limited, SL Sulphate Limited, SR Sulphide Removal. Table in part adapted from Oude Elferink *et al.* (1994)

Reference	Culture	Conditions	T °C	Y mg biomass/ mg AcH	μ_{max} d ⁻¹	$K_{S,AcH}$ mg AcH/l	$K_{S,SO_4^{2-}}$ mg SO_4^{2-} /l	K_{I,H_2S} mg H_2S /l	b d ⁻¹
Middleton and Lawrence (1977)	Enriched SRB	B, C,CL	20	0.065	0.33 ¹	250			0.00
			25	0.065	0.46 ¹	92			0.00
			31	0.065	0.57 ¹	5.7			0.00
Maillacheruvu and Parkin (1996)	Enriched SRB	B		0.025 ²	0.11 ^{1,2}	46.7 ²		8.5 ²	0.013
Bhattacharya <i>et al.</i> (1996)	Mixed SRB and MPB	B	35	0.0602	0.14 ¹	102.1			0
Gupta <i>et al.</i> (1994a,b)	Mixed SRB and MPB	C,SR	35	0.0469 ^{3,2}		0.84			
Visser <i>et al.</i> (1993)	Mixed SRB and MPB	UASB			0.07	5			
Visser (1995)	Granular sludge	UASB	30		0.11–0.12	55	33		
Visser (1995)	Suspended sludge	UASB	30		0.001–0.05	10	18		
Ingvorsen <i>et al.</i> (1984)	<i>Desulfobacter postgatei</i>	C,SL	30	0.072	0.32 ^{4,1}	3.84 ⁴	20.4 ⁴		
Ingvorsen <i>et al.</i> (1984)		C,CL	30	0.072	0.33 ^{4,1}	4.62 ⁴	21.1 ⁴		
Ingvorsen <i>et al.</i> (1984)		B	30				16.3 ⁴		
Schönheit <i>et al.</i> (1982)	<i>Desulfobacter postgatei</i>	B				12			
Brandis-Heep <i>et al.</i> (1983)	<i>Desulfobacter postgatei</i>		28	0.04	1.03	14			
Schauder <i>et al.</i> (1986)	<i>Desulfobacter postgatei</i>		30		0.6 ⁵				
Widdel (1987)	<i>Desulfobacter latus</i>				0.79				
Widdel (1987)	<i>Desulfobacter hydrogenophilus</i>		30		0.92 ⁵				
Widdel (1987)	<i>Desulfobacter curvatus</i>				0.79				
Widdel and Pfenning (1977)	<i>Desulfomaculum acetoxidans</i>		30	5.6	0.65–1.39				

¹Reported as k , ²Uncompetitive inhibition, ³Based on biomass COD of 1.41 mg COD/mg biomass, ⁴Reported as Michaelis-Menton parameter, ⁵Estimated from doubling time

Table 2.11: Monod kinetic parameters for sulphate reduction using propionate as substrate. Taken in part from Okabe and Characklis (1992) and Visser (1995)

Reference	Culture	Conditions	T °C	Y mg biomass/ mg propionate	μ_{max} d ⁻¹	$K_{S,PrH}$ mg PrH/l	K_{I,H_2S} mg H ₂ S/l	b d ⁻¹
Maillacheruvu and Parkin (1996)	Enriched SRB	Batch		0.048 ¹	0.11 ^{1,2}	27.2 ¹	206 ¹	0.021
Nanninga and Gottschal (1987)	<i>Desulfobulbus propionicus</i>	Batch	35		2.64			
Widdel and Pfennig (1982)	<i>Desulfobulbus propionicus</i>	Batch	39	0.071	1.66			
Widdel and Pfennig (1977)	<i>Desulfomaculum acetoxidans</i>	Batch	36		1.10			
Hunter (1989)	Mixed population	Continuous	35	0.022	1.68	90.0		
Samain <i>et al</i> (1984)	<i>Desulfobulbus elongatus</i>				1.39			
Stieb and Schink (1989)	<i>Desulfococcus multivorans</i>				0.17–0.23			

¹Uncompetitive inhibition, ²Reported as k

Table 2.12: Monod kinetic parameters for sulphate reduction using lactate as substrate. Adapted from Okabe and Characklis (1992).

Reference	Culture	Conditions	Y mg biomass/ mg LaH	μ_{max} d^{-1}	$K_{S,LaH}$ mg LaH/l	$K_{S,SO_4^{2-}}$ mg SO_4^{2-} /l
Cappenburg (1975)	<i>Desulfovibrio desulfuricans</i>	Continuous	0.343	8.64	4.4	
Traore <i>et al.</i> (1982)	<i>Desulfovibrio desulfuricans</i>	Batch	0.046	2.50		
Traore <i>et al.</i> (1982)	<i>Desulfovibrio africanus</i>	Batch	0.019	1.44		
Traore <i>et al.</i> (1982)	<i>Desulfovibrio gigas</i>	Batch	0.042	2.01		
Traore <i>et al.</i> (1981)	<i>Desulfovibrio vulgaris</i>	Batch	0.075			
Ingvorsen and Jørgensen (1984)	<i>Desulfovibrio vulgaris</i>	Batch		0.264		0.5
Ingvorsen and Jørgensen (1984)	<i>Desulfovibrio sapovarans</i>	Batch		0.168		0.7
Ingvorsen and Jørgensen (1984)	<i>Desulfovibrio salexigens</i>	Batch		0.504		7.4

Table 2.13: Monod kinetic parameters for sulphate reduction using butyrate as substrate. Taken in part from Visser (1995) and Okabe and Characklis (1992).

Reference	Culture	Conditions	μ_{max} d ⁻¹
Schauder <i>et al.</i> (1986)	<i>Desulfovibrio baarsi</i>	Batch	0.41
Nanninga and Gottschal (1987)	<i>Desulfovibrio sapovorans</i>	Batch	1.58
Stieb and Schink (1989)	<i>Desulfococcus multivorans</i>		0.17–0.23
Widdel and Pfennig (1981)	<i>Desulfotomaculum acetoxidans</i>		1.11
Kuever <i>et al.</i> (1993)	<i>Desulfotomaculum</i> strain Gro11		1.19–1.3
Brysch <i>et al.</i> (1987)	<i>Desulphobacterium autotrophicum</i>		0.67–1.11

Butyrate Substrate

The reactions for partial oxidation and cell synthesis, using butyrate as a substrate are:

As explained above, sulphate reduction has been included in the biomass synthesis reaction to avoid the production of hydrogen.

Kinetic parameters for butyrate sulphate reduction are given in table 2.13.

Sulphate Uptake

In order for the above sulphate reducing reactions to occur, sulphate must be present in sufficient quantities. Under sulphate limiting conditions a reduced rate results. This is usually accounted for by using dual substrate kinetics, i.e:

$$\mu = \mu_{max} \frac{S}{K_S + S} \frac{S_{\text{SO}_4^{2+}}}{K_{S,\text{SO}_4^{2-}} + S_{\text{SO}_4^{2-}}} \quad (2.73)$$

At present $K_{S,\text{SO}_4^{2-}}$ values have only been reported for acetate and lactate utilizing sulphate reducers. Ingvorsen *et al.* (1984) give a range of 11.5–31.7 mg/l with an average of around 20 mg/l for *Desulfobacter postgatei*. This is in accord with the results of Visser (1995), who reports values of 33 mg/l and 18 mg/l for mixed methanogenic and sulphate reducing, granular and suspended sludges in a UASB reactor.

2.9.2 Inhibition Kinetics

Sulphide Inhibition

In contrast to the other microbial reactions where only the undissociated form of hydrogen sulphide has been shown to be inhibitory, there is some debate as to whether total sulphide or only undissociated hydrogen sulphide is inhibitory to sulphate reducers. Hilton and Oleszkiewicz (1988) examined the effect of sulphide

Table 2.14: Summary of sulphide inhibition constants for sulphate reducers

Reference	Substrate	Noncompetitive		Uncompetitive	
		$K_{I,TS}$ mg S/l	K_{I,H_2S} mg H ₂ S/l	$K_{I,TS}$ mg S/l	K_{I,H_2S} mg H ₂ S/l
Maillacheruvu and Parkin (1996)	Hydrogen			422	149
Maillacheruvu and Parkin (1996)	Acetate			35	8.5
Maillacheruvu and Parkin (1996)	Propionate			681	206
Okabe <i>et al.</i> (1995)	Lactate	251	74 ¹		

¹Calculated from equilibrium at pH=7

on sulphate reduction over a pH range of 6.5 to 8.0. They found that even under alkaline conditions, where the concentration of undissociated sulphide is small, the sulphate reducers were inhibited. It was concluded that total sulphide was inhibitory. In their study of VFA degradation in a sulphidogenic reactor, Omil *et al.* (1996) found that a decrease in reactor pH from 8 to 7 resulted an increase in free sulphide levels. This was found to severely inhibit methanogenesis, while having little effect of sulphate reduction. This suggests that free sulphide and not total sulphide is inhibitory.

In contrast Reis *et al.* (1991) found that only the undissociated form was inhibitory over a pH range of 6.2–6.6. The same conclusion can be drawn from Visser (1995), whose results show that loss of activity has a better correlation with undissociated sulphide concentration, than with total sulphide. This is also in accord with the theory that only undissociated H₂S is able to penetrate the cell membrane Speece (1983).

Maillacheruvu and Parkin (1996) have shown that an uncompetitive model describes sulphide inhibition and have reported inhibition constants for partial propionate oxidizing, acetate oxidizing and hydrogen utilizing sulphate reducing bacteria. Values were reported for both total sulphide concentration and hydrogen sulphide concentration. Okabe *et al.* (1995) found that a noncompetitive model described sulphide inhibition of *Desulfovibrio desulfuricans* using lactate as a carbon source. A summary of sulphide inhibition constants for sulphate reducers is given in table 2.14. An important point worth mentioning at this stage, is the lower inhibition constants of the sulphate reducers as compared to methanogens. The implications of this are discussed below.

Acid Inhibition

As with the other microbial reactions occurring in an anaerobic digester, undissociated volatile fatty acids are inhibitory to sulphate reducers. Reis *et al.* (1990) examined the effect of undissociated acetic acid over a pH range of 5.8 to 7.0 on a sulphate reducing culture fed lactate. They concluded that a modified noncompetitive model describes the effect of acetic acid inhibition on sulphate reducers. Their equation is:

$$\frac{\mu}{\mu_{max}} = \frac{1}{1 + (S_{AcH}/K_{I,AcH})^{1.08}} \quad (2.74)$$

with an inhibition constant of $K_{I,AcH} = 54$ mg/l.

pH Inhibition

The optimal pH range for acetate utilizing sulphate reducers is about 7.0–7.8, with minimum and maximum values of around 5.5 and 9.0 (Visser, 1995; Fauque, 1995). However, Visser (1995) found that sulphate reducers in a sludge from a UASB reactor had an optimal range of 6.9 to 8.5, and were active up to a pH of 10. Presumably this is due to concentration gradients within the sludge particles. Based on these data the following values for a pH inhibition function are estimated: $\alpha_{LL} = 10$, $\alpha_{UL} = 10$, $\text{pH}_{LL} = 6.3$ and $\text{pH}_{UL} = 8.4$

2.10 Sulphate Reduction in Competition with Methanogenesis

The outcome of competition between the sulphate reducing bacteria and the methanogenic bacteria present in a sulphate reducing anaerobic reactor is important for several reasons. The most obvious point of consideration is that ‘useless’ methanogenic bacteria can be viewed as being in competition with ‘useful’ sulphate reducing bacteria for hydrogen and acetate substrates. As such, it would be desirable to operate a reactor in which sulphate reducing bacteria predominate. However, methanogenic bacteria have at least two ‘useful’ roles. In the first instance hydrogen utilizing methanogenic bacteria have a symbiotic relationship with fermenting bacteria. In order for effective fermentation to occur, a low hydrogen partial pressure is required (Harper and Pohland, 1986). The methanogenic bacteria are intimately associated with the fermenters and consequently the hydrogen produced by fermentation is rapidly consumed. The second useful function provided by methanogenic bacteria is their role in producing large biomass flocs. Sulphate reducing bacteria are poor extracellular polymeric substance (EPS) producers as compared to methanogens, and consequently do not do well in pure culture retained biomass systems such as UASB reactors. Methanogenic bacteria on the other hand secrete a larger amount of EPS and hence form good flocs in which both MPB and SRB are retained. Some methanogens would, therefore, be required in a retained biomass system or a system in which the biomass is thickened and returned (i.e. high rate system).

Several studies have been conducted into the outcome of competition between SRB and MPB (Kristjansson *et al.*, 1982; Schönheit *et al.*, 1982; Robinson and Tiedje, 1984; Isa *et al.*, 1986a,b; Oude Elferink *et al.*, 1994; Gupta *et al.*, 1994a; Harada *et al.*, 1994; Bhattacharya *et al.*, 1996; Shin *et al.*, 1996; Omil *et al.*, 1998). From a kinetic point of view the Monod constants μ_{max} and K_S can give insight as to which group will predominate. At high substrate levels the Monod equation approximates first order kinetics and consequently the group with the higher maximum specific growth rate can be expected to enjoy an advantage. Under low substrate conditions the group with the greater affinity for the substrate (i.e. lower K_S value) will have an advantage. From an inspection of tables 2.7 to 2.10, it is apparent that under first order conditions the two groups will have similar growth rates (the specific growth rates are of the same order of magnitude). However, under low substrate conditions, sulphate reducing bacteria will have an advantage as their half velocity constants are typically an order of magnitude less than those of the methanogens. Gupta *et al.* (1994a,b) have suggested that the ratios of the respective half velocity constants may be used as an indicator of the length of time it will take for one group to become extinct in a CSTR. The closer the ratio is to unity, the longer it will take for one group to become the sole user of substrate.

The above kinetic argument is only really valid for a continuous stirred type reactor.

In a retained biomass type system (i.e. where $SRT > HRT$) such as a UASB reactor, the better floc forming nature of methanogens can be expected to result in better retention. This is in accord with the results of Omil *et al.* (1998), who have reported that a shorter solids retention time results in a decrease in the time taken for SRB to predominate, and with the results of Isa *et al.* (1986b) who found that despite the lower growth rates of MPB they were able to dominate in a high-rate anaerobic reactor. Visser (1995) studied the biofilm and granulation forming abilities of pure methanogenic, mixed SRB/MP and pure sulphate reducing cultures in an UASB. He concluded that methanogens were necessary for the formation of sludge granules containing sulphate reducers. However, in a mixed culture over a period of months, sulphate reducers came to dominate—but not completely out compete—methanogens. In contrast to the results of Isa *et al.*, Visser observed that in a mixed culture the sludge immobilization capacity of the two groups are similar.

Another factor which will influence the outcome of competition is the relative sensitivities of the two groups to various inhibitory compounds. For example Mailacheruvu and Parkin (1996) have reported lower sulphide inhibition constants (i.e. greater inhibition) for both hydrogen and acetate utilizing SRB as compared to hydrogen and acetate utilizing methanogens. As has been observed by McCartney and Oleszkiewicz (1991) the greater sensitivity of the sulphate reducers to sulphide can be expected to negate some of the advantage gained from lower K_S values.

The optimal pH ranges for sulphate reducers and methanogens are roughly similar. However Visser (1995) has shown that in a mixed culture UASB, methanogenic species enjoy higher growth rates than sulphate reducers at pH values below 6.9. Between 6.9 and 8.5 the growth rates are roughly the same, and above pH 8.5 sulphate reducers have an advantage.

Other factors potentially affecting which group will predominate are sulphate concentration and the nature of the inoculum. Gupta *et al.* (1994a) have hypothesized that there is only a very narrow range of operating parameters in which SRB and MPB are able to coexist, and that a very slight change in one variable will result in the domination of one group over the other. It is hence apparent that in designing an anaerobic sulphate reducing reactor careful attention needs to be paid to ensure that reactor condition are optimal for maximum sulphate removal.

Chapter 3

Physical and Chemical Model Fundamentals

In addition to the biological reactions considered in the previous chapter, two further components are required to complete the development of an anaerobic sulphate reducing reactor model. In the first instance the physical chemistry of the system—comprising the solution chemistry and vapour liquid mass transfer effects—needs to be included. The second point to be included, is a mathematical description of the hydrodynamic characteristics of various biological reactor geometries. These two items are discussed below.

3.1 Physical and Chemical Processes Occurring

There are a number of reasons for including the effects of the chemical and physical reactions occurring, namely:

Biological reaction stoichiometry. It is apparent from the discussion in the previous chapter that depending on the dissociation state of the reactants, different stoichiometric relationships may be written for each biological reaction. For example, sulphate reduction using acetate as an electron donor has been represented by Isa *et al.* (1986a) as:

and by Middleton and Lawrence (1977) as:

Other forms in which sulphate is represented by HSO_4^- have also been used. However, the choice of stoichiometric reaction becomes arbitrary if the rate equation is coupled with algebraic equations describing the equilibrium dissociation of the components present.

Substrate concentrations. The substrates hydrogen and carbon dioxide are present in both the vapour and liquid phases. Since the kinetics are functions of substrate concentrations, the vapour/liquid mass transfer of at least these substrates needs to be included.

Inhibition effects. As discussed in the previous chapter, volatile fatty acids are only inhibitory when in their unionized states. Similarly hydrogen sulphide is often

only inhibitory in its unionized state. If the kinetics used incorporate inhibition terms, the exact ionization state of the inhibiting species must be calculated. Similarly pH inhibition models require the pH to be known.

H₂ regulatory effects. Hydrogen partial pressure has been shown to determine the relative production rates of acetate, propionate and lactate via anaerobic oxidation of higher order fatty acids and glucose. (Mosey, 1983; McCarty and Mosey, 1991). If the model incorporates kinetics describing this phenomenon, reactor hydrogen partial pressure must be calculated.

Heavy metal precipitation. An important feature of the biological treatment of AMD is the precipitation of heavy metals as sulphides, hydroxides and carbonates. In order to predict heavy metal levels in the effluent stream, metal precipitation combined with acid/base equilibria can be included.

Effluent quality prediction. It is desirable that the model be able to predict the quality of the final discharge. In order to calculate alkalinity, pH, heavy metal concentrations and sulphide content the chemistry of the system must be incorporated.

3.1.1 Gas/Liquid Mass Transfer

The rate of mass transfer between the vapour and liquid phases may be obtained from the familiar expression:

$$N = k_l a (c^* - c) \quad (3.1)$$

where N is the rate of mass transfer per unit volume ($\text{mmole} \cdot \text{l}^{-1} \text{d}^{-1}$); $k_l a$ is the product of the liquid side mass transfer coefficient and the specific gas-liquid interfacial area (d^{-1}); and $(c^* - c)$ is the difference between the equilibrium and actual molar concentrations of the soluble species in the liquid phase. The value of $k_l a$ is a function of several reactor conditions such as temperature, flow conditions, density and viscosity. It may either be determined experimentally or estimated from several correlations given in the literature. The equilibrium concentration c^* can be calculated from Henry's Law in which case equation 3.1 becomes:

$$N = k_l a \left(\frac{p}{\mathcal{H}} - c \right) \quad (3.2)$$

where p is the partial pressure of the component under consideration and $\mathcal{H}$ is its Henry's Law constant.

3.1.2 Solution Chemistry

Acid/Base Equilibria

Within the reactor a number of strong and weak acids and bases are expected to be present. These include:

- The strong acid H_2SO_4
- The weak acid H_2CO_3
- The weak acid H_2S
- The weak volatile fatty acids.

The simplest method for calculating the equilibrium of the acid dissociation reaction:

is from the equilibrium expression:

$$K_a = \frac{c_{\text{A}^-} \cdot c_{\text{H}^+}}{c_{\text{AH}}} \quad (3.4)$$

where K_a is a temperature dependent dissociation constant and c_i are molar concentrations. For a solution with a number of dissociating species, equation 3.4 is written for each species. This, combined with an ionic mass balance and a charge balance, results in a system of non-linear algebraic equations to be solved.

At high ionic concentrations significant deviation from equation 3.4 occurs and the concentration terms must be replaced with activities. (i.e. the activity coefficients are no longer close to unity). Equation 3.4 then becomes:

$$K_a = \frac{\gamma_{\text{A}^-} c_{\text{A}^-} \cdot \gamma_{\text{H}^+} c_{\text{H}^+}}{\gamma_{\text{AH}} c_{\text{AH}}} \quad (3.5)$$

The activity coefficients γ_i may be calculated using the Debye-Hückle limiting law (DHLL):

$$\log_{10}(\gamma_i) = -0.51 z_i^2 \sqrt{I} \quad (3.6)$$

where z_i is the ionic charge of the species and the ionic strength I is calculated from:

$$I = \frac{1}{2} \sum_i c_i z_i^2 \quad (3.7)$$

Empirical modifications to the DHLL to increase its accuracy at higher concentration include the Güntelberg equation (Sawyer and McCarty, 1989):

$$\log_{10}(\gamma_i) = -0.51 z_i^2 \frac{\sqrt{I}}{1 + \sqrt{I}} \quad (3.8)$$

and the Davies equation (van Haandel and Lettinga, 1994):

$$\log_{10}(\gamma_i) = -A z_i^2 \left[\frac{\sqrt{I}}{1 + \sqrt{I}} - 0.3I \right] \quad (3.9)$$

where the constant A is calculated from the temperature dependant relation (T in Kelvin):

$$A = 1.82 \times 10^6 (78.3T)^{-1.5} \quad (3.10)$$

An alternative method for calculating the equilibrium dissociation of a solution is by Gibbs free energy minimization. The total free energy is written as a function of the concentration of all of the species present. The minimum of this function is then determined subject to ionic mass balance and charge balance constraints. Conditions at this point represent the equilibrium. While this method is potentially very powerful, the fact that it involves an optimization algorithm makes it difficult to integrate with the differential equations describing the dynamics of the system.

A third alternative, for calculation of the solution chemistry, is to use a rate based model, such as that presented by Musvoto *et al.* (1997). Particular advantages of this approach are, that the assumption of equilibrium at every instance in time

can be relaxed, and that the model can be represented by a system of differential equations only. This is usually much simpler to solve than a mixed system of differential and algebraic equations. A disadvantage is that kinetic rate constants are required for both the forward and reverse reactions (although one rate constant can be written as a function of the other and the equilibrium constant). Another potential drawback particularly relevant to this project, is that the biological rates describe the production or consumption of the total (i.e. unionized and ionized) species. While an equilibrium based model can then be used to 'sort out' the concentrations of the two forms, this is not true of a rate based model, where the biological reactions would need to be written in terms of one or the other ionic species only. If the pH is near the pKa value for that component, this would result in the over production of one form and the underproduction of the other.

Metal Precipitation

As the pH of the reactor increases and as sulphide is generated the metal species present can be expected to precipitate as sulphides, carbonates and possibly, as hydroxides. The point at which precipitation of a divalent metal species M^{2+} occurs can be calculated from:

$$c_M \cdot c_{S^{2-}} \leq K_{sp,MS} \quad (3.11)$$

for a sulphide,

$$c_{M^{2+}} \cdot c_{CO_3^{2-}} \leq K_{sp,MCO_3} \quad (3.12)$$

for a carbonate and

$$2 c_{M^{2+}} \cdot c_{OH^-} \leq K_{sp,M(OH)_2} \quad (3.13)$$

for a hydroxide. Similar expressions may be written for trivalent ions.

3.2 Hydrodynamics of Reactors (Ideal and Real)

3.2.1 Ideal Continuous Stirred Tank Reactor (Liquid Phase Only)

Figure 3.1: Diagram of an ideal continuous stirred tank reactor (liquid phase only).

A diagram of a *liquid* phase ideal continuous stirred tank reactor (CSTR) is shown in figure 3.1. From the familiar mass balance expression:

$$\text{Rate of accumulation} = \text{Flow in} + \text{Rate of generation} - \text{Flow out}$$

and assuming constant reactor volume and constant stream density, the following equations describing the dynamic behaviour of an ideal CSTR may be derived:

Concentration of soluble components:

$$\frac{dS_2}{dt} = \frac{Q_1 S_1 - Q_2 S_2}{V_l} + r_S \quad (3.14)$$

$$= \frac{Q_1}{V_l} (S_1 - S_2) + r_S \quad (3.15)$$

and concentration of insoluble components:

$$\frac{dX_2}{dt} = \frac{Q_1 X_1 - Q_2 X_2}{V_l} + r_X \quad (3.16)$$

$$= \frac{Q_1}{V_l} (X_1 - X_2) + r_X \quad (3.17)$$

where S_1 , S_2 , X_1 and X_2 are vectors of the soluble and insoluble concentrations of the input and output streams as shown in the diagram. r_S and r_X are vectors of the overall rates of production (or consumption) of the soluble and insoluble components respectively. V_l is the reactor volume (l), and Q_1 and Q_2 are volumetric flow rates (l/day). The ratio Q_1/V_l is the inverse of the hydraulic residence time (HRT).

Equations 3.15 and 3.17, combined with equations representing the solution chemistry of the system, result in a system of differential and algebraic equations (DAEs). These may be solved to obtain the time dependent behaviour of the reactor.

The model may be reduced to a steady state version by setting the differentials to zero, the result of which is a system of non-linear algebraic equations.

3.2.2 Ideal Continuous Stirred Tank Reactor (Two Phase)

By including a term for the vapour liquid mass transfer the above model may be extended to include a vapour phase (Figure 3.2). Equations 3.14 and 3.15 then become:

$$\frac{dS_2}{dt} = \frac{Q_1 S_1 - Q_2 S_2}{V_l} + r_S - N \quad (3.18)$$

$$= \frac{Q_1}{V_l} (S_1 - S_2) + r_S - N \quad (3.19)$$

where the vector of mass transfer rates N , is determined using equation 3.2. Assuming that the ideal gas law holds, a mass balance over the vapour phase yields:

$$\frac{d\mathbf{p}}{dt} = \frac{-Q_G}{V_v} \mathbf{p} + \frac{V_l}{V_v} RTN \quad (3.20)$$

where $\mathbf{p}$ is a vector of the partial pressures, V_v is the vapour phase volume and Q_G is the gas flow rate leaving the reactor. A further condition required, to bring the number of equations to the number of unknowns, is that the sum of the partial pressures must equal the total pressure (usually one atmosphere).

If the reactor also has a gas input stream (for example a CO_2/H_2 feed stream or a stripping stream) equation 3.20 can be modified to include a term for the feed flowrate and feed partial pressures.

Figure 3.2: Diagram of an ideal continuous stirred tank reactor (two phase).

Figure 3.3: Diagram of a valley reactor.

3.2.3 Non-Ideal Plug Flow Reactor

The tanks-in-series model is often used to describe non-ideal plug flow (Levenspiel, 1972). As the name suggests, the model consists of a number of equal size theoretical CSTRs arranged in series. The number of reactors is usually determined from residence time distribution studies, as described by Levenspiel and others. The greater the number of reactors the more the model approximates ideal plug flow.

3.2.4 High Rate or Retained Biomass Systems (e.g. UASB)

Reactors with a solids retention time greater than the hydraulic residence time have an advantage in that they allow for a higher loading rate. Reactors of this type include upflow anaerobic sludge blanket (UASB) reactors, packed columns, 'valley' type reactors and the contact process. As shown in figure 3.3, a valley reactor (or falling sludge bed reactor) is, in effect, a settler with a solids recycle. This type of reactor is especially useful for fermenting insoluble solids to soluble substrates for use as sulphate reducing feed stock.

A retained biomass system is most easily modeled by using a CSTR combined with a solids recycle. The volume of the CSTR sets the hydraulic residence time, while the solids recycle may be adjusted to give the correct solids residence time. The solids recycle may have real meaning, as in the case of a valley reactor, or may be just a mathematical convenience, as in the case of a UASB reactor. A schematic

Figure 3.4: Mathematical representation of a retained biomass system.

diagram of this approach is shown in figure 3.4. A mass balance of the insoluble components over the CSTR yields:

$$V_1 \frac{dX_3}{dt} = X_1 Q_1 + X_4 Q_4 - X_3 Q_3 + r_X(X_3) V_1 \quad (3.21)$$

In addition a mass balance over the 'settler' results in:

$$X_2 Q_2 = X_3 Q_3 - X_4 Q_4 \quad (3.22)$$

$$= m X_3 Q_3 \quad (3.23)$$

where m is a parameter used to adjust the recycle rate, such that $0 < m \leq 1$. Substituting equation 3.22 into equation 3.21 gives:

$$V_1 \frac{dX_3}{dt} = X_1 Q_1 - X_2 Q_2 + r_X(X_3) V_1 \quad (3.24)$$

$$= Q_1 (X_1 - X_2) + r_X(X_3) V_1 \quad (3.25)$$

Using equation 3.23 to substitute $X_3 = Q_2 X_2 / (m Q_3)$:

$$\frac{dX_2}{dt} = \frac{m Q_3}{Q_2} \frac{Q_1}{V_1} (X_1 - X_2) + \frac{m Q_3}{Q_2} r_X(Q_2 X_2 / m Q_3) \quad (3.26)$$

The ratio $V_1 / m Q_3$ is the solids residence time (SRT) and is dependent on the solids retention capacity of the different components of the system (e.g. sludge settling, biomass floc formation, etc). Also since r_X is a linear function of solids concentration it is possible to simplify equation 3.26 finally giving:

$$\frac{dX_2}{dt} = \text{SRT}^{-1} (X_1 - X_2) + r_X(X_2) \quad (3.27)$$

Equation 3.27 is analogous to equation 3.17 with the solids residence time replacing the hydraulic residence time.

With respect to the soluble components in the system, there is no change in concentration across the settler, i.e. $S_2 = S_3 = S_4$. However, the rate of reaction, r_S , is a

function of the biomass concentration in the reactor X_3 . If r_S is to be expressed in terms of the exit biomass concentration, X_2 , algebraic manipulation similar to the above ultimately yields:

$$\frac{dS_2}{dt} = \text{HRT}^{-1}(S_1 - S_2) + \frac{\text{SRT}}{\text{HRT}}r_S(X_2) - N \quad (3.28)$$

Hence, the dynamic behaviour of a single stage high rate reactor is described by equations 3.27 and 3.28, combined with algebraic equations describing the solution chemistry.

3.2.5 Anaerobic Baffled Reactor (ABR)

Figure 3.5: Diagram of a five compartment baffle plate reactor.

A diagram of an anaerobic baffled reactor (ABR) is shown in figure 3.5. The reactor is a high rate reactor in the sense that the series of baffles allows for biomass retention and, hence, an increased loading rate. The baffled reactor can be regarded as a number of UASB reactors in series. Bachmann *et al.* (1985) used this approach combined with a fixed-film model to simulate an ABR. Grobicki and Stuckey (1992) performed residence time distribution studies on both clean and working ABRs and concluded that the hydraulic flow may be characterized by a number of theoretical perfectly mixed reactors in series. The number of ideal reactors required being equal to the number of reactor compartments.

An ABR reactor may, therefore, be modeled by a number of ideal two phase CSTRs with internal solids recycle in series. The solids retention within each compartment is accounted for by separating the solids and hydraulic residence times.

Chapter 4

Model Implementation and Results

In the previous two chapters the fundamental components of an anaerobic digester model were reviewed. This chapter deals with the synthesis of these components into a working model. This process involves several steps, namely:

- The selection of a computer package in which the model will be implemented.
- The selection of specific kinetic constants from the sometimes vast range presented in Chapter 2.
- Developing and testing submodules describing the biological reactions, equilibrium chemistry and vapour/liquid mass transfer.
- Combining these submodules into the final model.
- Calibration and testing of the final model against pilot plant and literature data.

4.1 Selection of Modelling Packages

In choosing a modelling package there are essentially two options, both with associated advantages and disadvantages. The first approach is to use a purpose written process simulation package such as Aspen or Hysis. These packages generally include a graphical user interface, built in chemistry data and physical property methods for a large number of components, modules describing a number of unit operations, numerical algorithms for the solution of the model, and facilities for displaying and plotting results. Using these packages it is generally very easy to develop a simulation using *existing* built in components and unit operations. However, the situation is somewhat different for the development of a new unit operation (in this case an anaerobic digester). While these packages generally provide facilities for incorporating user written code, there are some shortcomings:

- In general there is a trade off between ease of use of and flexibility. The package may impose strict limitations on how the problem must be formulated, regardless of whether this is the best or most convenient method. For example, the entire model may need to be constructed as a single file, which can be unwieldy, rather than dividing it into simpler more logical units.

- Much of the process of solving the model is hidden from the user. This can be a significant disadvantage during the development stage when it is desirable to monitor the behaviour of the model as it is solved.
- Perhaps the most serious problem is the limitation of the numerical routines used by the package. Although these can be quite sophisticated, convergence difficulties can still result. These can often be overcome by changing parameters relating to the numerical method, however on occasion it may be impossible to obtain a solution. In these cases it may be that all that is required is for the user to supply a good guess or for a different numerical method to be used, however this is prevented by the inflexibility of the package. Also, the program usually only reports that the model did not converge, but gives little insight into where the problem lies.
- Most packages are limited to steady state simulations. This is unfortunate as very often the dynamics of the system are also of interest.

The alternative option is to write the model ‘from the ground up’, using a high level programming or modelling language. Suitable programming languages include C and Fortran, for which a large number of prewritten numerical routines are freely available. Slightly more high level, are modelling languages such as Matlab and Octave. Features of these include strong support for matrix and vector operations and a number of built in numerical methods.

Particular advantages of this approach are: that the model may be constructed in modules each of which can be tested separately; flexibility in the choice of numerical method; the ability to aid convergence by providing a good guess and by monitoring the solution of the problem; and the ability to perform both dynamic and steady state simulations. The primary disadvantage of developing the model in this way, is that all of the system chemistry must be programmed in. Another disadvantage is that even if the resulting code is well documented and commented it can be difficult for a new user to pick up the model.

During the course of this project it was decided to attempt to have the best of both worlds. The model development, calibration and testing was done using the mathematical modelling languages Matlab and Octave, with the final result ported to Aspen as a Fortran 77 function. The features of these two tools as they relate to this project are discussed below.

Matlab and Octave

Apart from some minor differences in syntax Matlab and Octave are nearly identical¹. Their strong support for matrix and vector calculations makes it possible to represent the model’s mathematical equations in very compact code. Another important consideration is the large number of numerical routines available. For example a great deal of the code from the Numerical Algorithms Group (NAG) has been ported to Matlab. Similarly Octave utilizes the public domain routines, MINPACK, LSODE and DASSL respectively, for solving nonlinear algebraic, ordinary differential, and mixed systems of ordinary differential and algebraic equations.

¹The most important difference is that Octave is free while Matlab is a commercial product. Octave is named after the author of ‘Chemical Reaction Engineering’ (Levenspiel, 1972).

Aspen with Fortran 77 User Kinetics

Aspen is a flowsheeting package developed for use in the chemical process industries. A steady state simulation is built up from a number of mathematical unit operations provided with the package. The package is highly customizable in that the user may add in custom written unit operations, or may add specific kinetics to the reactor blocks. The programming language Fortran 77 is used for this purpose. Aspen also includes an extensive database of chemical components along with their physical and chemical properties. The package incorporates a number of equation of state (EOS) models for the calculation of solution chemistry and vapour liquid equilibria.

4.2 Evaluation and Selection of Kinetic Data

Figure 2.1 provides a map for the biological reactions occurring. However, before constructing a function describing the kinetics of the entire system, it is useful to examine each reaction group individually to select specific constants for inclusion in the final model. Three scenarios exist:

- In a number of cases there is a large selection of kinetic constants available. (This is particularly true of methanogenesis and sulphate reduction). The values reported may cover a large range and, depending on which are selected, can have a large impact on the model behaviour. Consequently in these cases a formal method of selecting suitable kinetics up front (or at least of whittling the selection down) is required.
- In those cases where only one or a few sets of constants have been reported it is necessary to make do with what is available. Fortunately this is usually only the case for reactions of lesser importance (e.g., acetogenesis).
- No constants are available. For example the unionized fatty acid and sulphide inhibition constants are unknown for many of the reactions. Similarly $K_{S,SO_4^{2-}}$ values for many of the sulphate reducing reactions are unknown (an important consideration under sulphate limited conditions). In these instances it is necessary to assume a reasonable value, usually based on one from a similar reaction.

With regard to the first point, the following procedure was used to select suitable constants for inclusion in the model:

- As far as possible the kinetic constants for a specific reaction should be those determined from the same group of experiments. While this is not usually possible for constants which are rarely reported (such as $K_{S,SO_4^{2-}}$ and inhibition constants), it is the case for the three Monod constants typically reported (i.e. Y , μ_{max} and K_S).
- The next step is to plot the behaviour of the reaction under batch, dynamic CSTR and steady state CSTR conditions. Plots of biomass concentration and substrate concentration are made. The idea here is that a general trend should emerge. The kinetics are then chosen, based on the assumption that greater confidence can be placed on those kinetics which conform to the general trend.

Figure 4.1: Biomass growth of acetate MPB under batch conditions near 35°C. Initial conditions are: Acetate 600 mg/l, biomass 5 mg/l. See table 2.8 for values of the kinetic parameters.

- Another point to be considered when selecting kinetics is the temperature of the reactor. Since very few correlations of kinetic constants with temperature have been made, it is important that the kinetic constants are selected from those determined at or near the temperature of the simulation.
- The expected substrate concentration within the reactor should also be taken into account. The reason being that at high substrate concentrations the value of the maximum specific growth rate rather than the half velocity coefficient determines which species will dominate. Conversely under low substrate conditions the value of K_S can be the deciding factor.

While this procedure is useful, it is not entirely satisfactory as it requires some subjective judgment. By way of example, consider methanogenesis using acetate as a substrate. From table 2.8 in Chapter 2 it is apparent that the reported kinetic constants span several orders of magnitude, i.e. Y : 0.01–0.82, μ_{max} : 0.03–1.4 and K_S : 0.07–869. Even if the extreme outlying points are eliminated, the values span a large range.

From table 2.8, we select those kinetics for which all three Monod parameters are given, and which are valid for a constant temperature. (In this case $35 \pm 2^\circ\text{C}$). Using these values a plot of the cell concentration as function of time for a batch reactor is made (See figure 4.1). A similar plot for a dynamic CSTR is given in figure 4.2.

Figure 4.2: Dynamics of biomass production of MPB in a CSTR near 35°C. Initial conditions are: Acetate 600 mg/l, biomass 5 mg/l. Feed is sterile 600mg/l acetate. Residence time of ten days. See table 2.8 for values of the kinetic parameters. See figure 4.1 for key.

On the figures two groupings appear: A group which has a rapid initial growth, and a high yield; and a group with a lower initial growth and yield. One option would then be to use kinetics which are representative of one of these trends. However, this does not take into account reactor conditions, specifically feed substrate concentration and residence time.

To illustrate this point, consider a single CSTR reactor seeded with the two species *Methanosarcina barkeri* and *Methanoxthrix soehngenii*. The respective Monod constants for these two species are: $Y = 0.024$, $\mu_{max} = 0.206$, $K_S = 230$ (Wandrey and Aivasidis, 1983); and $Y = 0.02$, $\mu_{max} = 0.16$, $K_S = 42$ (Huser *et al.*, 1982). Figure 4.3 shows the competitive behaviour of these two species at different residence times (10 days and 6 days). When the dilution rate (i.e. the inverse of residence time) is below the μ_{max} value of both species, it is the species with the better substrate uptake ability (i.e. lowest K_S) which ultimately predominates. If the dilution rate lies between the two specific growth rates, washout of the species with the lower specific growth rate occurs, regardless of its K_S value. At dilution rates above the highest specific growth rate washout of both species occurs.

Hence, which species will be favoured by selection pressure can easily be determined from a quick comparison of the dilution rate and Monod constants. However, to determine the length of time required for steady state to be established, a solution of the dynamic model is required. (Note the long time periods in figure 4.3).

The above argument can be extended to include the effect of inhibition. For example, if inhibition has been shown to be noncompetitive the dilution rate should be compared to $\mu_{max}(K_I/(K_I + I))$ rather than μ_{max} . Similarly the species with the better substrate uptake ability under competitive conditions is that with the lowest $K_S(1 + 1/K_I)$ value. However, the paucity of experimentally determined inhibition constants makes this (at this stage) only of theoretical interest.

(a) Residence time = 10 days. Low substrate levels in reactor. K_S determines outcome of competition, hence *Methanotherix soehngenii* dominates.

(b) Residence time = 6 days. High substrate levels in reactor. μ_{max} determines outcome of competition, hence *Methanosarcina barkeri* dominates.

Figure 4.3: Effect of residence time on long term competition between acetate utilizing methanogens in a CSTR. (Note difference in time scales). Feed is sterile 1500 mg/l acetate, initial biomass concentrations of 5 mg/l.

4.3 Synthesis and Implementation of the Model

4.3.1 Model Components

The following components were taken as being representative of those found in an anaerobic high sulphate system:

Insoluble components The insoluble components present are: carbohydrate represented by the formula $C_6H_{10}O_5$; a generic protein represented by $C_5H_7O_2N$; and a generic lipid (the triglyceride of palmitic acid). These components are assumed to be present in both degradable and nondegradable forms. In addition dead biomass ($C_5H_7O_2N$) was also included as an insoluble component.

Non-dissociating soluble components A generic amino acid $C_5H_9O_3N$, glucose, glycerol and the long chain fatty acid palmitic acid are the non-dissociating products of hydrolysis.

Dissociating soluble components These include: four short chain fatty acids, acetate, propionate, butyrate and lactate; hydrogen sulphide and its ions; carbon dioxide, carbonic acid and carbonates; ammonia; sulphuric acid, sodium sulphate and their ions.

Vapour phase components Hydrogen sulphide, carbon dioxide, methane and hydrogen are present in both the vapour and liquid phases. When the model incorporates gas stripping nitrogen is included in the vapour phase only.

Microbial groups Based on the substrates utilized, twelve different microbial groups (not species) are assumed to be present. Apart from the fermenters which utilize amino acids, glucose and glycerol. All other species were assumed to be specialized, i.e. only one compound is utilized as a substrate. In all cases biomass is represented by the empirical formula $C_5H_7O_2N$. The groups are:

- Fermenters (FER) utilizing glucose, amino acids and glycerol
- Lactate fermenters (*l*FER)
- Beta oxidizing bacteria (BOB) utilizing long chain fatty acids
- Butyrate utilizing acetogens (*b*ACE)
- Propionate utilizing acetogens (*p*ACE)
- Hydrogen utilizing methanogens (*h*MPB)
- Acetate utilizing methanogens (*a*MPB)
- Hydrogen utilizing sulphate reducers (*h*SRB)
- Acetate utilizing sulphate reducers (*a*SRB)
- Lactate utilizing sulphate reducers (*l*SRB)
- Propionate utilizing sulphate reducers (*p*SRB)
- Butyrate utilizing sulphate reducers (*b*SRB)

Physical properties required for these components include, molecular weights, COD values, K_a values for the weak acids, k_{la} and Henry's constants for those components present in the vapour phase, and kinetic constants for the microbial groups. Tables A.1–A.6 in Appendix A give a complete list all components along with their physical, chemical and kinetic properties.

Table 4.1: Stoichiometry of sulphate reducing microbial reactions included in the model. (Water and hydrogen ions not shown).

Hydrogen substrate:

Acetate substrate:

Propionate substrate (partial oxidation):

Lacate substrate (partial oxidation):

Butyrate substrate (partial oxidation):

4.3.2 Microbial Reaction Rates and Stoichiometry

Tables 4.1 and 4.2 show the stoichiometry of the microbial reactions used in the model. To reiterate, the reactions should be interpreted as the production or consumption of the total amount of each species present. The exact degree of dissociation of each species is determined by the equilibrium chemistry as described below. Note that water has not been included in the reactions as the concentration of water has no meaning. Protons are also not included, as the pH is either fixed or determined from a charge balance.

The microbial module is implemented as a function which returns the rate of change of the concentration all species as function of reactor conditions, i.e:

$$\mathbf{r} = f(\mathbf{X}_b, \mathbf{X}_s, \mathbf{S}, VFA, pH, FS) \quad (4.1)$$

where $\mathbf{r}$ is a column vector of the rate of change in concentration of the microbial, solid and dissolved species concentrations; $\mathbf{X}_b$ is the concentration of the twelve microbial groups; $\mathbf{X}_s$ is the concentration of the degradable solid components and $\mathbf{S}$ is the concentration of dissolved species. The total undissociated volatile fatty acid concentration (VFA), the pH and free sulphide concentration (FS) are also included because of their inhibitory effects.

The Matlab/Octave code for equation 4.1 (`biorates.m`) is given in Appendix B along with a description of its internal workings. The Fortran equivalent, for use with Aspen, (`USRSAN.F`) is given in Appendix C.

Table 4.2: Stoichiometry of non-sulphate reducing microbial reactions included in the model. (Water not shown).

Hydrolysis:

Glucose fermentation:

Amino acid fermentation:

Glycerol fermentation:

Lactate fermentation:

Beta oxidation:

Acetogenesis (propionate substrate):

Acetogenesis (butyrate substrate):

Methanogenesis (hydrogen and carbon dioxide substrates):

Methanogenesis (acetate substrate):

The following framework was used to calculate the biological rates:

- Hydrolysis of the insoluble components was calculated using first order kinetics (equation 2.17).
- Glucose and lactate fermentation are according to the hydrogen regulated kinetics of Costello *et al.* (1991a,b).
- Amino acid fermentation uses Monod kinetics, producing a fixed ratio of acetate and propionate, as outlined in section 2.4.3.
- Glycerol fermentation is by Monod kinetics but uses rate data determined for glucose.
- The remaining reaction rates are calculated using Monod kinetics.
- The sulphate reduction reactions incorporate terms for both sulphate and the electron donor uptake.

Except for hydrolysis, all of the above reaction rates include terms for sulphide, fatty acid and pH inhibition. However, the modular nature of the model makes it easy to disable any of these if so required.

The following is a description of the kinetic constants included in the model along with the reasons for their selection:

- The beta oxidation of palmitic acid uses the constants reported by Gujer and Zehnder (1983) as given in table 2.4. Since a limited amount of data describing the beta oxidation of LCFAs are available these values were selected by default.
- As the fermentation of glucose and lactate is described by the hydrogen regulated scheme of Costello *et al.*, the kinetic constants given by these workers are used here.
- As no data for the fermentation of amino acids are available, the kinetic constants were estimated using the approach described in section 2.4.3.
- The acetogenesis of propionate is described using the kinetics of Maillacheruvu and Parkin (1996). Data from these workers are used as all the Monod constants are reported (including sulphide inhibition) and because the values given lie near the middle of the ranges of values reported by other workers (See table 2.5).
- The acetogenesis of butyrate is described using the kinetics of Lawrence and McCarty (1969). Data from these workers are used as it is the most complete set. (Only the sulphide inhibition constant is unknown). In addition the specific growth rate lies near the middle of the range of values as reported in table 2.6. No values for the sulphide inhibition constant have been reported. Consequently the value reported for propionate acetogenesis is used instead.
- The kinetics of Maillacheruvu and Parkin (1996) are used for hydrogen methanogenesis. Data from these workers are used as all the Monod constants are reported (including sulphide inhibition) and because the values given are representative of values reported by other workers. (See table 2.7). Similarly the data of these workers are also used to describe acetate methanogenesis, hydrogen sulphate reduction, acetate sulphate reduction and propionate sulphate reduction.

- In the case of lactate sulphate reduction only one complete set of the three Monod constants is available (Cappenburg, 1975) however, the yield and maximum specific growth rate are about an order of magnitude larger than those reported by other workers. (See table 2.12). Consequently the more representative values of Traore *et al.* (1982) were used for the yield and maximum specific growth rate. The half velocity constant used is still that of Cappenburg as this is the only one available. In addition the sulphide inhibition constant reported by Okabe *et al.* (1995) is used.
- μ_{max} values are the only Monod constants reported for butyrate utilizing sulphate reducers (see table 2.13). Consequently the values used for the remaining kinetic constants are assumed to be the same as those used for propionate sulphate reduction. The μ_{max} value used is that given by Schauder *et al.* (1986) which lies roughly near the mean of the values reported.
- $K_{S,SO_4^{2-}}$ values have been reported only for lactate and acetate sulphate reduction. (See tables 2.10 and 2.12). The values reported by Ingvorsen and Jørgensen (1984) and Ingvorsen *et al.* (1984) are used here. For the remaining sulphate reducing reactions, the sulphate uptake constants were assumed to be the same as that for acetate sulphate reducers.
- Very little data is available for fatty acid inhibition. The $K_{I,VFA}$ values reported by Costello *et al.* (1991a,b) are used for all of the non-sulphate reducing reactions. As no value is reported for LCFA beta oxidizing bacteria, it is assumed to be the same as for the fermenters. The only fatty acid inhibition constant reported for sulphate reducers is that for lactate utilizing bacteria (Reis *et al.*, 1990). In the absence of any data the same value is used for the remaining sulphate reducing reactions.

A summary of the kinetic constants used is given in tables A.5 and A.6 in Appendix A.

It should be noted at this point that the kinetic constants used in the model should be regarded as model constants and not as adjustable parameters. In the same way that a model of a distillation column would not be calibrated by adjusting the universal gas constant or Antoine equation constants, neither should the kinetic constants be used to calibrate this model. Admittedly the values of the constants used here are known less precisely than the value of the gas constant however, their exact values are best determined from purpose designed experiments rather than by tweaking them to make the model fit. (To continue the analogy, fitting a model of a distillation column to experimental data would be a poor way of calculating vapour pressure data).

4.3.3 Solution Chemistry and Vapour/Liquid Mass Transfer

The following approach was used to determine the equilibrium dissociation of the liquid phase of the system. Activity based equilibrium expressions (equation 3.5) for the following acid/base reactions are written:

K_a constants for the above reactions are given in table A.4 in Appendix A. Any other sulphate species present (represented by Na_2SO_4) are assumed to dissociate completely. Carbon dioxide is present in the liquid phase in four forms, $\text{CO}_{2,l}$, H_2CO_3 , HCO_3^- and CO_3^{2-} . The equilibrium between dissolved carbon dioxide and carbonic acid is given by the reaction:

Bailey and Ollis (1986) give the reaction constants of the forward and reverse reactions as $k_1 = 20 \text{ s}^{-1}$ and $k_{-1} = 0.03 \text{ s}^{-1}$ (25°C) from which the equilibrium constant can be calculated.

In addition, mass balance equations are written for each species present. For example, for sulphide:

$$[\text{H}_2\text{S}]_l + [\text{HS}^-] + [\text{S}^{2-}] = [\text{H}_2\text{S}]_{total}$$

Finally the proton concentration (and hence the pH) is calculated from a charge balance, i.e:

$$\sum_i c_i z_i = 0 \quad (4.2)$$

where c_i and z_i are the molar concentration and charge of species i . In those cases where the pH is directly controlled, equation 4.2 is not used—the hydrogen ion concentration being directly calculated from the pH. It should also be mentioned at this point, that at high pH values, where the concentration of H^+ ions is low, it is often more accurate to calculate pH from the OH^- concentration.

The above can be represented as a system of nonlinear algebraic equations of the form:

$$f(\mathbf{c}) = 0 \quad (4.3)$$

which may then be solved using a suitable numerical procedure, such as the Newton-Raphson method. Equation 4.3 is implemented as the Octave/Matlab function `equilib.m` (See Appendix B).

Mass transfer rates between the liquid and vapour phases for the four vapour phase components, CO_2 , H_2S , H_2 and CH_4 , are calculated using equation 3.2, i.e.

$$N = k_l a \left(\frac{p}{\mathcal{H}} - c \right)$$

The volumetric mass transfer coefficient, $k_l a$, is principally dependant on the gas diffusivity and the gas liquid interfacial area. Diffusivity in turn is mainly a function

of temperature. The interfacial area can be dependent on a large number of factors, including the liquid viscosity and density, flow velocity and type (i.e. laminar or turbulent), degree of mixing and reactor geometry. Unfortunately data for anaerobic systems is scarce, and correlations for calculating k_{la} are usually only applicable to the mass transfer of oxygen in aerobic systems.

In their model of an anaerobic digester Graef and Andrews (1974) used a value of 100 day^{-1} for carbon dioxide. Janssen (1996) gives a value of 4320 day^{-1} for hydrogen sulphide. van Houten *et al.* (1994) calculated k_{la} values for hydrogen in a gas lift reactor at different gas flow rates (53–457 l/h). For a two phase system k_{la} values were between 2082 day^{-1} and 3205 day^{-1} . In a three phase system, in which pumice stone was added, values were somewhat lower, ranging from 687 day^{-1} to 1840 day^{-1} . No values have been reported for methane. However, since methane is only very sparingly soluble in water it can be expected to rapidly reach equilibrium concentrations in the vapour and liquid phases. Consequently in the model a large value (10^6 day^{-1}) was assumed. This has the effect of allowing the liquid and vapour concentrations to come to equilibrium rapidly. In any case, since methane does not have an effect on microbial rates (as H_2 , CO_2 and H_2S do) not knowing the exact rate of its mass transfer is a very minor inaccuracy in the model.

Vapour/liquid mass transfer is calculated by the Octave function `vap_liq.m` (See Appendix B). It has the format:

$$N = f(P, c)$$

i.e., given the partial pressures and liquid phase concentration it returns the volumetric rates of mass transfer.

4.4 Example Simulations

The model was tested against the results of a number of different experimental data sets presented in the literature. Where transient data is presented the model was run as a dynamic simulation and the results compared on a graph. In those cases where only steady state values are reported the model was run as a steady state simulation only.

In all of the experiments used for testing the model the hydraulic residence time was reported (or could be calculated). However, the solids residence time was always unknown and could not be easily calculated. This posed a problem as the model requires the SRT as an input parameter. A solution to this was to run the model for a number of different solids residence times to see which best predicted the reactor biomass concentration (or some other parameter). When the experiment covered a number of time periods with different experimental conditions, the first period was used to predict the SRT and the others used for comparison.

4.4.1 Test Simulation No. 1—Reactor Composition as a Function of pH

The first test simulation deals only with the solution chemistry and vapour/liquid mass transfer. For a system with fixed initial concentrations, the model was used to determine the ionization and partial pressure of each species as a function of pH. Log concentration plots versus pH were prepared, some of which are shown in figures 4.4 and 4.5. Figure 4.4(a) shows the concentration of the four forms of carbon

dioxide, and is in good agreement with similar plots given in the literature (see for example Bailey and Ollis, 1986; Loewenthal *et al.*, 1989). Similarly figure 4.4(b) shows the concentration of the three forms of sulphide present. These two figures illustrate, how in the pH range of anaerobic digestion, more than one form of carbon dioxide and hydrogen sulphide may be present in significant amounts. Propionate and butyrate have been omitted from the plot of VFA concentrations (figure 4.5), as their K_a values are close to those of acetate.

Figure 4.6 shows the partial pressures of CO_2 and H_2S as functions of pH. As the pH is increased to above the pK_a value the partial pressure decreases as the solution is able to hold more of the species in its ionized forms.

4.4.2 Test Simulation No. 2—Packed Bed Reactor fed Molasses

Maree *et al.* (1987) operated an upflow packed bed reactor for 200 days on a feed of molasses and acid mine drainage. Two thirds of the 1 liter reactor was packed with dolomite pebbles with a void ratio of 50%. The relevant operating parameters for the duration of the experiment are given in table 4.3.

Since the physical reactor had a high recycle rate relative to the feed flow (20 l/day against 1.8 l/day), the reactor hydraulic phase was modelled as an ideal CSTR. The molasses feed was approximated as glucose giving a simulated feed COD of 3210 mg/l (as compared to 3360 mg/l in reality). The initial concentrations of each microbial species were unknown and arbitrary values of 1 mg/l were assumed; the long duration of the experiment allowing the simulated species to grow (or washout) to steady state concentrations. As the solids residence time was not reported, this was estimated by running the model for a number of different sludge ages between 2 and 7 days to see which gave the best estimate of COD and sulphate concentrations. This was only done for the first period (days 1-90), the second and third periods being used for comparison.

The error was quantified by summing the absolute difference between the experimental and model predicted values. A plot of the error as function of solids residence time for both COD and sulphate is shown in figure 4.7. A coarse plot (figure 4.7a), taken at intervals of 0.5 days SRT, indicates that the SRT giving the best fit to the data is around 3.5 days. A more detailed plot around this point (figure 4.7b) indicates that a SRT of 3.4 days gives the best prediction of COD, while a SRT of 3.6 gives a better prediction for sulphate. Consequently in the final simulation a SRT of 3.5 days was used. This value is well within the range that might be expected of a reactor of this type.

Plots of actual and simulated sulphate and COD concentrations in the effluent are shown in figure 4.8. The predicted sulphate level is in good agreement with

Table 4.3: Experimental conditions for molasses fed packed bed reactor (Maree *et al.*, 1987).

Period	1	2	3
Days	0-90	91-122	123-200
Retention time (h)	20	15	10
Molasses (mg/l)	2000	2000	3000
Sulphate (mg/l)	2480	2480	2480

(a) Initial carbon dioxide concentration is 100 mmol/l.

(b) Initial hydrogen sulphide concentration is 10 mmol/l.

Figure 4.4: Equilibrium concentrations of carbon dioxide ions and sulphide ions as a function of pH

Figure 4.5: Equilibrium concentrations of VFAs and their ions as a function of pH. Initial concentrations of acetic and lactic acid are 10 mmole/l. Propionate and Butyrate are not shown as their pKa values (and hence the corresponding curves) are close to that of acetate.

Figure 4.6: Equilibrium partial pressures of hydrogen sulphide and carbon dioxide in a closed system as a function of pH.

(a) Coarse scale.

(b) Fine scale.

Figure 4.7: Sum of absolute error of sulphate and COD as a function of the model SRT for a molasses fed packed bed reactor

Figure 4.8: Actual and simulated COD and sulphate concentrations in a molasses fed packed bed reactor.

the experimental values. The COD is slightly underestimated, although it is still within the range of experimental values. It is worth noting that the predicted COD is in better agreement in the second and third time periods of the experiment, even though the SRT time was only determined from data from the first time period. Presumably this is due to the fact that the initial biomass concentration (as input into the model) has less of an effect later in time.

Hence, in spite of the arbitrarily chosen initial biomass concentration, the model gives a good prediction of the behaviour of packed bed reactor.

4.4.3 Test Simulation No. 3—UASB fed Molasses

In conjunction with the above experiment, Maree *et al.* (1987) operated a UASB fed the same combination of sulphate water and molasses. The retention time and molasses dosages for the duration of the experiment are shown in table 4.4. In addition, on days 162 and 208 half of the biomass was removed to prevent the reactor becoming overgrown.

The model was setup in a similar manner to the above case. The liquid phase of the reactor was assumed to be perfectly mixed and was modelled as a CSTR. The solids retention time was not reported, and consequently was determined from a number of trial runs (data from the first period only was used for this). As only sulphate concentration was reported, COD was not used to calculate the error. Figure 4.9 gives a plot of the error in sulphate concentration (as defined above) as a function of SRT over coarse and fine time scales. As can be seen the SRT giving the best fit to the data is around 3.6 days. This value is within the range that might be expected of a UASB type reactor.

A plot of the experimental and model predicted sulphate concentrations using this solids retention time is shown in figure 4.10. There is good agreement between the predicted and experimental values, particularly in the second half of the experiment when the physical reactor had settled down. The increase in sulphate levels, due to sludge wasting on days 162 and 208, is also predicted.

4.4.4 Test Simulation No. 4—Packed Bed fed Sewage Sludge

Maree and Strydom (1983) operated a series of packed bed reactors on mixture of mine water and sugar, sulphite pulp mill effluent (lignosulphonate), sewage sludge and sewage sludge with sugar. Simulations of only the last two experiments were performed. (The results of a simulation of the sugar fed reactor are expected to be similar to those of the molasses fed packed bed reactor described above; and no kinetic data is available for the anaerobic degradation of lignosulphonate).

The reactor used had a high recirculation rate relative to the feed flow (50:1), hence the liquid phase could be approximated as an ideal CSTR. The reactor

Table 4.4: Experimental conditions for molasses fed UASB (Maree *et al.*, 1987).

Period	2	3
Days	65–120	121–215
Retention time (h)	22	15
Molasses (mg/l)	2000	3000
Sulphate (mg/l)	2400	2400

(a) Coarse scale.

(b) Fine scale.

Figure 4.9: Sum of absolute error of sulphate as a function of the model SRT for a molasses fed UASB

Figure 4.10: Actual and simulated sulphate concentrations in a molasses fed UASB.

Figure 4.11: Absolute error in steady state sulphate concentration as a function of SRT in sewage fed packed bed reactor.

had a volume of 1 liter and was packed with a stone media (50% voidage). The recirculation line passed through a photosynthetic reactor in which sulphide was partially oxidized to elemental sulphur. This side stream reactor ensured that the hydrogen sulphide level seldom exceeded 100 mg/l as SO_4^{2-} (35 mg/l as H_2S).

The feed sulphate level was 1350 mg/l at a pH of 7.4. The high sulphate water was mixed with sludge to give a feed COD of 5000 mg/l. Where sugar was added the COD increased to 5564 mg/l (554 mg/l being contributed by the sugar). The reactor was operated at a hydraulic retention time of 2 days.

As no dynamic data was presented for the cases of the reactor running on sewage sludge, only steady state simulations were performed. The steady state case does not require the initial biomass concentration to be known. The sludge composition used is given in Appendix A. As the SRT was not reported, the first experiment was used as a calibration case. Figure 4.11 shows the absolute error in predicted sulphate concentration as a function of solids residence time. The diagram indicates that a SRT of around 7 days gives the best fit.

Table 4.5 shows the actual and predicted amounts of sulphate and COD removed, for both of the experimental periods calculated at the 7 day SRT. A good agreement between predicted and actual COD removal is found for the first experiment (COD was not used in the calibration), and between the actual and predicted sulphate and COD removal in the second experiment.

4.4.5 Test Simulation No. 5—Gas Lift Reactor fed H_2/CO_2

van Houten *et al.* (1994) operated a sulphate reducing gas lift reactor, fed carbon dioxide and hydrogen. Pumice particles were used as the carrier material giving the reactor an active volume of 4.5 litres. The reactor was operated at 30°C and a pH of 7.0. Gas feed varied between 2.4 and 6.0 l/h and was recycled at a rate of 350 l/h. Table 4.6 shows the other relevant operating parameters for the experiment.

The initial biomass concentrations of $h\text{MPB}$ and $h\text{SRB}$ were arbitrarily set to 1 mg/l. As the actual gas recycle rate was high (350 l/h), it was assumed that the reactor was well mixed, and that the hydraulic phase could therefore be modelled as a CSTR. As before the solids residence time was unknown and it was necessary to try a number of values to see which gave the best fit.

This was done using data from only the last two periods (i.e. from day 11 on) when the reactor was at steady state. This removes the effect of the arbitrary initial biomass concentration. Figure 4.12 shows a plot of the sum of the absolute error for these two periods, as a function of the SRT. The predicted SRT giving the least error is 1.3 days. However, the slope of the error curve for an SRT greater than 1.3 is very gentle and consequently a SRT time considerably greater than 1.3 results in very little increase in the error. It is therefore possible that the actual SRT may be somewhat higher than 1.3.

Table 4.5: Sulphate and COD removal in a packed bed fed sewage sludge and sugar. Actual and simulated results.

Parameter	Sewage sludge			Sewage sludge + sugar		
	Actual	Simulated	Error	Actual	Simulated	Error
COD removed (mg/l)	3800	3500	8%	4364	4064	7%
SO_4^{2-} removed (mg/l)	1050	1065	1%	1300	1265	3%

Table 4.6: Experimental conditions for hydrogen/carbon dioxide fed gas lift reactor (van Houten *et al.*, 1994).

Period	1	2	3	4
Days	1-4	5-6	7-29	30-40
Hydraulic retention time (h)	27	13.5	4.5	4.5
Gas flow (normal l/h)	2.43	2.55	3.30	6.60
H ₂ in gas (% v/v)	99	94	80	80
CO ₂ in gas (% v/v)	1	6	20	20
Sulphate (mg/l)	3361	3361	3361	3361

Figure 4.12: Sum of absolute error of the sulphate conversion rate as a function of the model SRT

Figure 4.13: Actual and simulated sulphate loading and conversion rates in a hydrogen fed gas lift reactor.

Figure 4.13 shows the actual and experimental sulphate conversion rates for the forty days of the experiment as calculated using the above SRT. The sulphate loading rate is also shown for comparison. During the growth period, as the reactor is brought to steady state, the predicted and actual conversion rates are in good agreement. The sharp peaks on days 5 and 7 are a result of an increase in the loading rate; when the conversion rate is momentarily artificially high until the reactor effluent concentration reacts to the change in feed. The reactor reaches steady state around day 11 (both simulated and experimental), however the predicted removal rate is some $2 \text{ g l}^{-1} \text{ d}^{-1}$ higher than the actual rate. This is an error of about 15%.

On day 30 the gas feed flow rate was doubled to 6.6 l/h . The experimental conversion rate increases after about two days to the maximum rate possible. The predicted rate also increases, however the change is immediate and the predicted rate is a little less than the maximum achievable. van Houten *et al.* ascribe the increase in sulphate reduction to increased stripping of hydrogen sulphide (and hence lower sulphide inhibition), rather than being due to an increase in hydrogen concentration. However the model predicts that the change is due to better hydrogen mass transfer to the liquid phase, due to a higher hydrogen partial pressure in the gas phase.

The increase in the model conversion rate after the increase in gas flow rate on day 30 is relatively small. In addition while the experimental conversion rate equals the loading rate after day 30, the model result falls a little short. Since hydrogen was found to be limiting in the reactor, this is probably due to too low a mass transfer rate from the vapour to the liquid phase.

4.4.6 Test Simulation No. 6—Valley Reactor fed Sewage S-ludge

The 'Rhodes Biodesalination Process' developed by the Department of Biochemistry and Microbiology at Rhodes University (Corbett, 1998) is a three stage process for

Figure 4.14: The Rhodes biological AMD treatment pilot plant flow diagram.

the treatment of AMD using sewage sludge as a carbon source. The first stage is a 'valley' type reactor, with a very high solids retention time, in which the sludge is solubilised and fermented. Some sulphate reduction occurs in this stage. The second stage is a high rate baffle reactor, in which the remainder of the sulphate reduction occurs. This is followed by an algal pond for removal of the residual COD. The process as it was implemented at the Grootvlei pilot plant is shown in figure 4.10.

The mine water treated in the pilot plant had a sulphate content of around 1700 mg/l at a pH of about 6.5. The reactor was operated at COD to sulphate ratios of 3:1, 2:1 and 1.5:1. The last mix ratio was only used for four days—perhaps not long enough for a steady state to develop.

The model was used to predict the VFA, sulphate and soluble COD concentrations exiting the first reactor. Although the solids distribution within the valley reactor has been shown to vary both as a function of height and horizontal distance from the inlet, it was assumed that the liquid phase could be modelled by an ideal CSTR. The solids residence time is not known, however the solids concentrations and flow rates of the three underflow lines are known. From this the solids recycle flow was estimated and the SRT of the model adjusted to give the same value. The SRT calculated using this procedure was 19 days. This value is not unreasonable given that the valley reactor is designed to have a high solids retention time.

The composition of the sludge as input into the model is given in table A.3 in Appendix A. The high solids residence time allows for near complete solubilisation of the sludge. During the setup of the model it was found that in order to achieve the observed COD and sulphate removals, the fractions of the sludge which should be regarded as degradable were at the upper end of those values listed in table 2.3. This is in accord with the experimental observation that the high solids residence time allows for near complete solubilisation of the sludge.

In addition the sludge has a VFA content, resulting from fermentation which had occurred in the sludge holding tank and earlier. This was reported as being in the range of 130–390 mg/l (as measured in the mixed feed). The form of the VFA was not noted. Consequently in the model an acetate content of 200 mg/l in the feed was used.

The actual and predicted steady state sulphate and COD concentrations exiting the valley reactor are shown in table 4.7. In the case of the experimental data, the

Table 4.7: Actual and predicted effluent quality in a primary sludge fed valley reactor. Actual figures are the nominal concentrations taken from Corbett (1998).

COD:Sulphate	Sulphate (mg/l)			COD (mg/l)		
	Actual	Predicted	Error	Actual	Predicted	Error
3:1	~ 1200	1360	~ 13%	~ 1800	2253	~ 25%
2:1	~ 1200	1459	~ 22%	~ 1600	1514	~ 5%
1.5:1	~ 1400	1527	~ 9%	~ 1600	1145	~ 28%

values covered a fairly large range and only the nominal concentrations are shown. The model predicted results are somewhat higher than the experimental results (5-28%), however they are still well within the range of values reported by Corbett (1998).

Chapter 5

Case Study—Design of a Full Scale Plant

The aim of this chapter is to demonstrate some of the model's capabilities not shown in the previous chapter. This is done by way of a case study design and evaluation of a hypothetical reactor. The stream to be treated is a 20 Ml/day acid mine drainage stream with a pH of 2.5 and a sulphate content of 2000 mg/l. Primary sewage sludge is available as a carbon source. The sulphate content of the water is to be lowered to below 200 mg/l and the pH must be between 6.5 and 8.5. The following questions were considered:

- What reactor configurations—i.e. UASB, CSTR, baffle plate, etc—would be suitable?
- What solid and liquid residence times are required to achieve the required level of sulphate removal?
- What effect would the simultaneous removal of sulphide (e.g., by stripping or side stream partial oxidation) have on reaction rates and hence on the reactor size?
- What is the effect of a shock increase in COD load on reactor performance?

Reactor Type

With respect to choosing the reactor configuration, the model is only of limited use. This is because the model only takes into account the different hydraulic and solids retention times, rather than using more complicated expressions to account for the reactor geometry. For example, no distinction is made between a UASB or a packed bed reactor, with respect to their different flow and mixing characteristics. The selection of a reactor can, therefore, only be partially considered (and then only qualitatively).

The low pH of the mine water means that a reactor with near ideal plug flow characteristics would not be suitable. The reactor should either be well mixed or have a high recycle rate to neutralize the incoming feed. In addition, to reduce the reactor volume and hence its cost, the reactor should be capable of good solids retention. In this particular case, this is important both to ensure effective acidogenesis of the sludge and to maintain a high biomass concentration. In addition, a granular or

Table 5.1: Demonstration simulation. Sulphate and sulphide concentrations at different SRT and HRT. Sulphate in mg/l, sulphide is total sulphide (TS) in mg/l as H_2S . Sulphide levels controlled by stripping.

HRT days	SRT							
	2.0		5.0		10.0		20.0	
	SO_4^{2-} mg/l	TS mg/l	SO_4^{2-} mg/l	TS mg/l	SO_4^{2-} mg/l	TS mg/l	SO_4^{2-} mg/l	TS mg/l
2.0	1723	2.24	736	10	251	14	147	15
1.0	1750	2.00	738	10	254	14	158	14
0.5	1780	1.74	745	10	262	13	165	13

sludge composed of flocs is better able to withstand pH extremes or high sulphide levels. As discussed above, this is presumably due to concentration gradients within the particles.

A high rate baffle plate type reactor was discarded as an option as it would be unable to raise the pH across the length of the digester unless a recycle was provided. However a high recycle increases the through flow rate and negates many of the advantages of this type of reactor. This reactor is hence only suitable for treating a stream with a pH near to that required for optimal anaerobic digestion.

Suitable reactors therefore include any high rate, well mixed reactor such as a UASB with recycle, a valley reactor or a contact process reactor.

Effluent Quality as a Function of Retention Times

In general, the higher the solids residence time the better the solubilisation of the sludge and the higher the biomass concentration. However, the law of diminishing marginal returns operates, and at some point the advantages of a higher SRT may be outweighed by the higher costs needed to achieve it. In addition, as the SRT increases the portion of inactive biomass in the reactor increases (This is accounted for by the 'dead biomass' fraction in the model).

A lower hydraulic residence time implies a smaller reactor and therefore lower cost. However, if the HRT is too low, the soluble products of hydrolysis are removed before they can undergo acidogenesis. Consequently too low a HRT can limit sulphate reduction. As before, there is a point at which the benefits of a higher HRT are outweighed by the associated costs.

In addition the solid and hydraulic residence times are limited by what is practically achievable.

Engineering design is often an iterative process involving trial and error and the design of an anaerobic sulphate reducing reactor is no exception. A number of different solid and liquid residence times were simulated. The sulphate concentrations in the effluent (for a fixed COD) feed are shown as functions of SRT and HRT in table 5.1. As can be seen, the effluent quality is mainly a function of the SRT and only weakly dependent on HRT. This, however, does not mean that the HRT can be set arbitrarily low, as the SRT is still dependent on the reactor volume. From the table it is apparent that, to achieve the required sulphate removal a solids retention time of between 10 and 20 days is required.

Effect of Sulphide Removal

Sulphide stripping of a recirculating side stream will result in lower sulphide levels and, hence, is expected to increase reaction rates. Against the advantage of a smaller reactor, there is the added cost of a stripping column. Alternatively, as in the scheme proposed by Maree and Strydom (1983), a recirculating side stream can be passed through a photosynthetic reactor where the sulphide is partially oxidized to elemental sulphur. Again the benefits of lower sulphide levels are offset by the cost of an additional reactor.

In any event, if hydrogen sulphide is not to be discharged to the atmosphere and receiving waters, some sulphide removal and sulphur recovery system is required. Hence, it might be desirable to intergrate this with the reduction stage, where the benefits of lower sulphide levels will be felt, rather than positioning it as a second stage in series.

The effect of sulphide stripping was examined running the reactor at a different stripping gas flow rates. (A stripping gas flow was modelled by including a term for an inert gas feed in the model). The gas flow rate was reduced from 2×10^8 to 2×10^7 on day 100 and then further reduced to 2×10^6 on day 200. (All flow rates in normal l/day). Figure 5.1 and 5.2 show the effect of the changes on reactor sulphide and sulphate concentrations. The change in acetate concentration is shown in figure 5.3. There is a rapid increase in acetate concentration on day 200 indicating the failure of sulphate reduction. The concentration then gradually decreases, as methanogens start to establish themselves. This is shown by the increase in methane partial pressure in figure 5.4. (Note that the initial rapid increase in methane concentration on day 200 is not due to a sudden growth in methanogens, but is due to the gas stripping flow rate dropping by a factor of 10, and hence the partial pressure increasing by the same factor).

Therefore, in order to maintain sulphate reduction a gas stripping flow rate of around 2×10^7 nl/d is required.

Figure 5.1: Effect of gas stripping on reactor sulphide concentration. Stripping gas flow rates are 2×10^8 l/d (days 1–100), 2×10^7 l/d (days 101–200) and 2×10^6 l/d (days 201–300).

Figure 5.2: Effect of gas stripping on reactor sulphate concentration. Stripping gas flow rates are 2×10^8 l/d (days 1–100), 2×10^7 l/d (days 101–200) and 2×10^6 l/d (days 201–300).

Figure 5.3: Effect of gas stripping on reactor acetate concentration. Stripping gas flow rates are 2×10^8 l/d (days 1–100), 2×10^7 l/d (days 101–200) and 2×10^6 l/d (days 201–300).

Figure 5.4: Effect of gas stripping on methane partial pressure. Stripping gas flow rates are 2×10^8 l/d (days 1–100), 2×10^7 l/d (days 101–200) and 2×10^6 l/d (days 201–300).

Effect of an Increased Load on Reactor Operation

Conventional (i.e. low sulphate) anaerobic digesters have been shown to be sensitive to sudden increases in COD loading rates. A rapid increase in load can lead to a build up of volatile fatty acids with a consequent drop in pH and possible digester failure. A mechanism to account for this, suggested by McCarty and Mosey (1991), is as follows: A sudden increase in an easily degraded compound, such as glucose, results in a rapid increase in production of hydrogen. As the hydrogen partial pressure increases, acidogenic bacteria switch production away from acetic acid to higher order fatty acids such as propionate. Propionate utilizing acetogens are highly specialized, and are only able to use propionate as a substrate. Since the normal levels of propionate in the digester are low these bacteria have to grow up from a very small population. In addition their specific growth rates are low, $\mu_{max} \sim 0.3$ (See table 2.5). As a result propionate may persist for quite some time, depressing the reactor pH. The low pH (or high unionized propionate levels) inhibit the hydrogen scavenging methanogens, which in turn results in an increase in hydrogen levels, further compounding the problem.

It is suggested that a high sulphate reactor should be more resilient to shock loads in COD. The reason for this is that sulphate reducing bacteria are able to utilize propionate and higher order fatty acids. Their maximum specific growth rates are also higher than those for acetogens (e.g. $\mu_{max} \sim 1.3$ for *Desulfobulbus propionicus*). In addition, hydrogen utilizing sulphate reducing bacteria are better scavengers than methanogens. (Compare the K_S and μ_{max} values in tables 2.7 and 2.9).

This hypothesis was tested using the model. The response of a reactor to a step increase in feed COD was simulated under both high and low sulphate conditions. The reactor was operated with sulphide stripping to remove the effect of sulphide inhibition. The HRT was 2 days and the SRT 5 days. The reactor was operated with a sludge feed at a COD of 6000 mg/l. The sulphate concentration in the high sulphate case was 2000 mg/l. The step increase in COD was effected by increasing the glucose in the feed from 0 to 500 mg/l on day 150.

Plots of VFA concentrations are shown in figures 5.5–5.6. In the case of the high sulphate reactor, VFA concentrations are low both before and after the increase in glucose feed. The exception to this is acetate, the concentration of which increases dramatically but returns to the original level after about 50 days. The fact that acetate is the preferred product of acidogenesis suggests that hydrogen levels in the reactor are low. The hydrogen partial pressure is shown in figure 5.7, which shown this to be the case.

In contrast, in the low sulphate reactor (with otherwise identical conditions), the higher order fatty acids lactate, propionate and butyrate are present in greater amounts. In addition, lactate and propionate levels do not recover after the increase in feed COD. This suggests a high reactor hydrogen partial pressure, which is confirmed in figure 5.7. (The slight drop in hydrogen partial pressure on day 150, is due to lactate to propionate fermentation which is a hydrogen consuming reaction).

It is, therefore, suggested that: All else remaining equal, a high sulphate anaerobic digester is likely to be more stable and less prone to failure than a conventional digester, provided that low sulphide levels can be maintained.

Figure 5.5: Change in acetate and lactate levels in response to an increase in digester feed COD on day 150, in high and low sulphate digesters.

Figure 5.6: Change in propionate and butyrate levels in response to an increase in digester feed COD on day 150, in high and low sulphate digesters.

Figure 5.7: Change in hydrogen partial pressure in response to an increase in digester feed COD on day 150, in high and low sulphate digesters.

Chapter 6

Conclusions

A mathematical model describing the dynamic and steady state behaviour of an anaerobic digester, treating high sulphate waste waters has been developed. The model has been shown to be capable of predicting a number of different scenarios, including:

- The ionization states of the weak acids and bases, occurring in an anaerobic system, as function of pH.
- The time dependant concentration of sulphate and COD in molasses fed packed bed and UASB reactors.
- The steady state COD and sulphate concentrations in a packed bed reactor fed sewage and sugar.
- The dynamic sulphate conversion rate in a gas lift reactor fed hydrogen and carbon dioxide.
- The steady state COD and sulphate concentrations of valley reactor fed sewage sludge.

In addition, the model was used to demonstrate its usefulness as a design tool. The following scenarios were considered:

- The effect of HRT and SRT on sulphate removal. Sulphate removal was found to be strongly dependent on the solids residence time, and only weakly dependent on the hydraulic residence time.
- The effect of sulphide removal on COD and sulphate removal efficiencies. High sulphide levels resulted in the failure of sulphate reduction and an increase in methanogenesis.
- Whether high sulphate levels result in better reactor stability as compared to a conventional anaerobic digester. A high sulphate reactor was found to have lower hydrogen and VFA levels as compared to a low sulphate digester. In addition, a high sulphate reactor exhibits better recovery from a shock change in COD loading rate.

Recommendations for Further Research

The removal of heavy metals from low pH, high sulphate streams has not been extensively covered in this thesis. The usefulness of the model can be extended by including terms describing the precipitation and bio-adsorption of heavy metals.

From an experimental point of view, there are a number of areas where no or very little data is available. In particular, Monod half velocity constants for sulphate uptake ($K_{S,SO_4^{2-}}$), and VFA and sulphide inhibition constants for a number of reactions are unknown.

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Appendix A

Model Parameters and Constants

Table A.1: Properties of insoluble components included in model.

Component	Formula	MW	COD
		g/gmole	mg COD/mg
All biomass	$C_5H_7O_2N$	114.12	1.42
Protein	$C_5H_7O_2N$	114.12	1.42
Carbohydrate	$C_6H_{10}O_5$	162.14	1.18
Lipids	$C_{51}H_{98}O_3$	807.34	2.87

Table A.2: Properties of vapour phase components used in the model.

Component	MW	COD	$\mathcal{H}$ atm/(mmole/l)		$k_L a$ day ⁻¹
	g/gmole	mg COD/mg	25°C	35°C	
CO ₂	44.01	0		3.76×10^{-2}	>100
H ₂ S	34.08	1.88	9.714×10^{-3} †	1.218×10^{-2} †	4320
H ₂	2.02	7.94	1.273*	1.336*	687–3205
CH ₄	16.04	3.99	0.7440*	0.8755*	$\sim 1 \times 10^6$

*Perry and Green (1984)

†Carroll (1990)

Table A.3: Primary sludge data used in the model. Based on data from tables 2.2 and 2.3

Component	Dry weight %	Degradable fraction	k_{hyd} d ⁻¹
Protein	0.385	0.7	0.69
Carbohydrate	0.267	0.7	0.58
Lipid	0.281	0.5	0.30
VFA (acetate)	0.067	-	-

Table A.4: Properties of soluble components included in model. K_a values from Ebbing (1990).

Component	Formula	MW g/gmole	COD mg COD/mg	Charge	K_a (25°C) gram mole basis
Amino acids	$C_5H_9O_3N$	131.13	1.22	0	-
Glucose	$C_6H_{12}O_6$	180.26	1.07	0	-
Glycerol	$CH_2OHCHOHCH_2OH$	92.09	1.22	0	-
Palmitic acid	$CH_3(CH_2)_{14}COOH$	256.43	2.87	0	-
Acetic acid	CH_3COOH	60.05	1.07	0	1.7×10^{-5}
Acetate ion	CH_3COO^-	59.05	-	-1	-
Propionic acid	CH_3CH_2COOH	74.08	1.51	0	1.3×10^{-5}
Propionate ion	$CH_3CH_2COO^-$	73.07	-	-1	-
Butyric acid	$CH_3(CH_2)_2COOH$	88.11	1.82	0	1.4×10^{-5}
Butyrate ion	$CH_3(CH_2)_2COO^-$	87.10	-	-1	-
Lactic acid	$CH_3CHOHCOOH$	90.08	1.60	0	1.3×10^{-4}
Lactate ion	$CH_3CHOHCOO^-$	89.07	-	-1	-
Hydrogen sulphide	H_2S	34.08	1.88	0	8.9×10^{-8}
Hydrogen sulphide ion	HS^-	33.07	-	-1	1.2×10^{-13}
Sulphide ion	S^{2-}	32.07	-	-2	-
Carbon dioxide	CO_2	44.01	0	0	1.5×10^{-4} *
Carbonic acid	H_2CO_3	62.03	0	0	4.3×10^{-7}
Hydrogen carbonate ion	HCO_3^-	61.01	-	-1	4.8×10^{-11}
Carbonate ion	CO_3^{2-}	60.01	-	-2	-
Sulphuric acid	H_2SO_4	98.08	0	0	∞
Hydrogen sulphate ion	HSO_4^-	97.06	-	-1	1.1×10^{-2}
Sulphate ion	SO_4^{2-}	96.06	-	-2	-
Ammonia	NH_3	17.03	0	0	-
Ammonium ion	NH_4^+	18.04	-	+1	5.6×10^{-10}
Sodium sulphate	Na_2SO_4	142.04	0	0	-
Sodium ion	Na	22.99	-	+1	-
Hydrogen ion (proton)	H^+	1.01	-	+1	-
Hydroxide ion	OH^-	17.01	-	-1	-

* K value for the reaction: $CO_2 + H_2O \rightleftharpoons H_2CO_3$

Table A.5: Summary of Monod parameters used in the model.

Group	Substrate	Abbreviation	μ_{max} d ⁻¹	Y mg/mg	K_S mg/l	$K_{S,SO_4^{2-}}$ mg/l	b d ⁻¹
Fermenters	Amino acids	FER	10 ¹	0.55 ¹	20 ¹	-	0.02 ¹
	Glucose	FER	varies	varies	23 ²	-	0.02 ²
	Glycerol	FER	1.5 ¹	0.82 ¹	20 ¹	-	0.02 ¹
Lactate fermenters	Lactate	lFER	varies	varies	34 ²	-	0.02 ²
	Palmitic acid	BOB	0.12 ³	0.3 ³	49.8 ³	-	0.01 ³
LCFA beta oxidizers	Butyrate	bACE	0.37 ⁴	0.085 ⁴	7.2 ⁴	-	0.027 ⁴
Butyrate utilizing acetogens	Propionate	pACE	0.15 ⁵	0.063 ⁵	17.9 ⁵	-	0.021 ⁵
Propionate acetogens	Hydrogen	hMPB	0.18 ⁵	0.39 ⁵	30 × 10 ⁻³ (6)	-	0.013 ⁵
Hydrogen utilizing methanogens	Acetate	aMPB	0.14 ⁵	0.041 ⁵	27 ⁵	-	0.013 ⁵
Acetate utilizing methanogens	Hydrogen	hSRB	0.18 ⁵	0.33 ⁵	25 × 10 ⁻³ (5)	20 ¹	0.013 ⁵
Hydrogen utilizing sulphate reducers	Acetate	aSRB	0.11 ⁵	0.025 ⁵	46.7 ⁵	20 ⁶	0.013 ⁵
Acetate utilizing sulphate reducers	Lactate	lSRB	2.50 ⁷	0.046 ⁷	4.4 ⁸	1.0 ⁹	0.02 ¹
Lactate utilizing sulphate reducers	Propionate	pSRB	0.11 ⁵	0.048 ⁵	27.2 ⁵	20 ¹	0.021 ⁵
Propionate utilizing sulphate reducers	Butyrate	bSRB	0.41 ¹⁰	0.04 ¹	10 ¹	20 ¹	0.02 ¹
Butyrate utilizing sulphate reducers							

¹No datum available. Value is assumed.²Costello *et al.* (1991a,b)³Gujer and Zehnder (1983)⁴Lawrence and McCarty (1969)⁵Maillacheruvu and Parkin (1996)⁶Ingvorsen *et al.* (1984)⁷Traore *et al.* (1982)⁸Cappenburg (1975)⁹Ingvorsen and Jørgensen (1984)¹⁰Schauder *et al.* (1986)

Table A.6: Summary of inhibition parameters used in the model.

Group	K_{I,H_2} atm	$K_{I,VFA}$ mmole/l	K_{I,H_2S} mg/l	$K_{I,TS}$ mg/l as S	α_{LL}	α_{UL}	pH_{LL}	pH_{UL}
FER	Eqn 2.34 ^{*,2}	10.0 ^{*,2}	100 ^{*,1}	-	4	0.5	5	10.5
lFER	Eqn 2.34 ^{*,2}	10.0 ^{*,2}	100 ^{*,1}	-	4	0.5	5	10.5
BOB	Eqn 2.34 ^{*,2}	10.0 ^{*,1}	100 ^{*,1}	-	4	0.5	5	10.5
bACE	Eqn 2.34 ^{*,2}	3.0 ^{†,2}	26.6 ^{†,1}	-	20	20	7.1	8.0
pACE	Eqn 2.34 ^{*,2}	30.0 ^{†,2}	26.6 ^{†,3}	-	20	20	7.1	8.0
hMBP	-	0.7 ^{*,2}	664 ^{†,3}	-	20	20	7.1	8.0
aMPB	-	0.7 ^{*,2}	117 ^{†,3}	-	20	20	7.1	8.0
hSRB	-	1 ^{*,1}	-	422 ^{†,3}	10	10	6.3	8.4
aSRB	-	1 ^{*,1}	-	35 ^{†,3}	10	10	6.3	8.4
lSRB	-	1 ^{*,4}	-	251 ^{*,5}	10	10	6.3	8.4
pSRB	-	1 ^{*,1}	-	681 ^{†,3}	10	10	6.3	8.4
bSRB	-	1 ^{*,1}	-	681 ^{†,1}	10	10	6.3	8.4

*Noncompetitive inhibition, [†]Uncompetitive inhibition, [†]Competitive inhibition¹No datum available, value is assumed²Costello *et al.* (1991b)³Maillacheruvu and Parkin (1996)⁴Reis *et al.* (1990)⁵Okabe *et al.* (1995)

Appendix B

Matlab/Octave Code

The following code describes the model as it has been implemented using Octave. Some minor modification would be required for the code to be used in Matlab. In particular the comment symbol, # should be replaced by the Matlab equivalent %. Also the differential algebraic solver used, `dassl` would need to be replaced by a Matlab version.

Subroutine of Equilibrium Algebraic Equations—`equilib.m`

```
1 #####
2 #
3 #           High Sulphate Anaerobic Digester Simulation
4 #
5 # Author:      Anthony Knobel
6 # Date:        15 August 1999
7 # File name:   equilib.m
8 # Description: This function returns the residual of the
9 #             algebraic equations describing the acid/base
10 #             equilibrium of the anaerobic system.
11 #             At equilibrium the residuals are equal to zero.
12 #
13 #####
14
15 # Define Nomenclature local to this function
16
17 # St is a vector of the total molar concentrations of each component
18 # present before equilibrium is calculated: i.e. all components
19 # in undissociated liquid phase.
20
21 # St:    1 AcH
22 #        2 PrH
23 #        3 BuH
24 #        4 LaH
25 #        5 H2
26 #        6 CO2
27 #        7 S04 (Na2S04+H2S04)
28 #        8 NH3
29 #        9 Na2S04
30
31 # Se: Equilibrium concentration (1-26)
```

```

32
33 # Se:    1 H+
34 #        2 AcH
35 #        3 Ac-
36 #        4 PrH
37 #        5 Pr-
38 #        6 BuH
39 #        7 Bu-
40 #        8 LaH
41 #        9 La-
42 #       10 H2S(1)
43 #       11 HS-
44 #       12 S--
45 #       13 CO2(1)
46 #       14 H2CO3
47 #       15 HCO3-
48 #       16 CO3--
49 #       17 H2SO4
50 #       18 HSO4-
51 #       19 SO4--
52 #       20 NH3
53 #       21 NH4+
54 #       22 OH-
55 #       23 Na2SO4
56 #       24 Na+
57
58 function res = equilib(Se,St)
59
60 global SeCH
61
62 ### Misc. constants
63 T = 25+273.15;
64 R = 82.06;      # Gas constant (atm.l/(K.mmole))
65 NoSe = 30;
66
67 res = zeros(NoSe,1);
68
69 ### Equilibrium constants (all concentrations in mmole/l)
70 # Equilibrium constants for loss of proton
71 K_AcH = 1.76E-5/1E3;      # mmole/l (mole/l*10^-3)
72 K_PrH = 1.34E-5/1E3;      # mmole/l (mole/l*10^-3)
73 K_BuH = 1.54E-5/1E3;      # mmole/l (mole/l*10^-3)
74 K_LaH = 1.4E-4/1E3;       # mmole/l (mole/l*10^-3)
75 K_H2S = 8.9E-8/1E3;       # mmole/l (mole/l*10^-3)
76 K_HS = 1.2E-13/1E3;       # mmole/l (mole/l*10^-3)
77 K_CO2 = 1E12/1E3;         # unitless (CO2(1) <-----> H2CO3)
78 K_H2CO3 = 4.3E-7/1E3;     # mmole/l (mole/l*10^-3)
79 K_HCO3 = 4.8E-11/1E3;     # mmole/l (mole/l*10^-3)
80 K_H2SO4 = 1E12/1E3;       # mmole/l (mole/l*10^-3)
81 K_HSO4 = 1E-2/1E3;        # mmole/l (mole/l*10^-3)
82 K_NH4 = 5.6E-19/1E3;      # mmole/l (mole/l*10^-3)
83 K_H2O = 1E-14/1E6;        # (mmole/l)^2 ((mole/l)^2*10^-6)
84
85 ### Alternatively use:
86 # K_H2CO3 = (10^-(17052/T+215.21*log10(T)-0.12675*T-545.56))/1E3;
87 # K_HCO3 = (10^-(2902.39/T+0.0239*T-6.498))/1E3
88 # K_H2O = (10^-(4787.3/T+7.1321*log10(T)+0.01037*T-22.801))/1E6
89

```

```

90  ### Charge balance
91  res(1) = Se(1:24)'*SeCH(1:24);
92
93  ### Mass balances and equilibria
94  ## Calculate activity coefficients
95  I = 0.5*((Se(1:24)./1000)'*(SeCH(1:24).^2));
96  A = 1.82E6*(78.3*T)^-1.5;
97  a = 10.^(-A*(SeCH(1:24).^2)*(sqrt(I)/(sqrt(I)+1)-0.3*I));
98  ASe = Se(1:24).*a;
99
100 ## AcH
101 res(2) = Se(2)+Se(3)-St(1);
102 res(3) = K_AcH*ASe(2)-ASe(3)*ASe(1);
103
104 ## PrH
105 res(4) = Se(4)+Se(5)-St(2);
106 res(5) = K_PrH*ASe(4)-ASe(5)*ASe(1);
107
108 ## BuH
109 res(6) = Se(6)+Se(7)-St(3);
110 res(7) = K_BuH*ASe(6)-ASe(7)*ASe(1);
111
112 ## LaH
113 res(8) = Se(8)+Se(9)-St(4);
114 res(9) = K_LaH*ASe(8)-ASe(9)*ASe(1);
115
116 ## H2S(l), HS-, S--, H2S(v)
117 res(10) = Se(10)+Se(11)+Se(12)-St(5);
118 res(11) = K_H2S*ASe(10)-ASe(11)*ASe(1);
119 res(12) = K_HS*ASe(11)-ASe(12)*ASe(1);
120
121 ## CO2, H2CO3, HCO3-, CO3--
122 res(13) = Se(13)+Se(14)+Se(15)+Se(16)-St(6);
123 res(14) = K_CO2*ASe(13)-ASe(14);
124 res(15) = K_H2CO3*ASe(14)-ASe(15)*ASe(1);
125 res(16) = K_HCO3*ASe(15)-ASe(16)*ASe(1);
126
127 ## H2SO4, HSO4-, SO4-- Na2SO4
128 res(17) = Se(17)+Se(18)+Se(19)+Se(23)-St(7);
129 #res(18) = K_H2SO4*ASe(17)-ASe(18)*ASe(1);
130 res(18) = Se(17);
131 res(19) = K_HSO4*ASe(18)-ASe(19)*ASe(1);
132
133 ## NH3 NH4+
134 res(20) = Se(20)+Se(21)-St(8);
135 res(21) = K_NH4*ASe(21)-ASe(20)*ASe(1);
136
137 ## H+,OH-
138 # Use one of the following equations:
139 # (No 2 gives best results in pH range 1-8)
140 #res(22) = 14 + log10(Se(22)/1000)+log10(Se(1)/1000);
141 #res(22) = Se(22)/1000-(10^-(14+log10(Se(1)/1000)));
142 res(22) = Se(1)*Se(22)-K_H2O;
143
144 ## Na2SO4, Na
145 K_Na2SO4 = 1E9; # (Complete dissociation)
146 res(23) = (2*Se(23)+Se(24))-2*St(9);
147 #res(24) = K_Na2SO4*ASe(23)-ASe(24)^2*ASe(19);

```



```

148 res(24) = Se(23);
149
150 endfunction

```

Subroutine of Biological Kinetics—biorates.m

```

1 #####
2 #
3 #           High Sulphate Anaerobic Digester Simulation
4 #
5 # Author:      Anthony Knobel
6 # Date:        15 August 1999
7 # File name:    biorates.m
8 # Description:  This file calculates the rates of production and
9 #               consumption of components participating in the
10 #               biological reaction. For dissociating species
11 #               the overall sum of all ions is calculated.
12 #
13 #####
14
15 # Define local nomenclature
16
17 # Xb, rateXb, etc
18 # 1 Fermenters
19 # 2 LaH Ferementers
20 # 3 LCFA Oxidizers
21 # 4 BuH Acetogens
22 # 5 PrH Acetogens
23 # 6 H2 MPB
24 # 7 AcH MPB
25 # 8 H2 SRB
26 # 9 AcH SRB
27 # 10 LaH SRB
28 # 11 PrH SRB
29 # 12 BuH SRB
30
31 # Xs, rateXs, etc
32 # 1 Degradable sludge protein
33 # 2 Degradable sludge carboydrate
34 # 3 Degradable sludge lipid
35 # 4 Dead Biomass
36
37 # S, SMW, etc (all MW are for undissociated form, except S04--)
38 # 1 Total AcH: AcH + Ac-
39 # 2 Total PrH: PrH + Pr-
40 # 3 Total BuH: BuH + Bu-
41 # 4 Total LaH: LaH + La-
42 # 5 Total sulphide : H2S(1) + HS- + S--
43 # 6 Total carbonate: CO2(1) + H2CO3 + HCO3- + CO3--
44 # 7 Total sulphate: H2SO4 + HS04- + S04-- + Na2S04
45 # 8 Total ammonia: NH3 + NH4+
46 # 9 Amino acids
47 # 10 Glucose
48 # 11 Glycerol
49 # 12 Palmitic
50 # 13 CH4(1)
51 # 14 H2(1)

```

```

52
53 # M, MMW, etc (Used for calculating inhibition parameters only)
54 # 1 Undissociated VFA: i.e. AcH + PrH + BuH + LaH
55 # 2 Undissociated H2S: H2S(1)
56 # 3 H+ (Used to calculate pH)
57
58 function [rateXb, rateXs, rateS] = biorates(Xb,Xs,S,M,XbMW,XsMW,SMW,MMW)
59
60 NoXb = 12;
61 NoXs = 4;
62 NoS = 14;
63 NoM = 3;
64
65 ### Set parameters shared by some reactions
66 ## pH Inhibition parameters
67 # Fermenters and beta oxidizers:
68 a_LL_F = 4;
69 a_UL_F = 0.5;
70 pH_LL_F = 5;
71 pH_UL_F = 10.5;
72
73 # Acetogens and Methane Producers:
74 a_LL_MP = 20;
75 a_UL_MP = 20;
76 pH_LL_MP = 7.1;
77 pH_UL_MP = 8.0;
78
79 # Sulphate Reducers:
80 a_LL_SR = 10;
81 a_UL_SR = 10;
82 pH_LL_SR = 6.3;
83 pH_UL_SR = 8.4;
84
85 # Common hydrogen inhibition parameter
86 T = 273.15+35;
87 KI_H2_pp = 10-(1139/T-7); # atm
88 H = 1.356; # Hydrogen Henry's constant atm/(mmole/l)
89 KI_H2 = (KI_H2_pp/H); # mmole/l
90
91 # Calculate pH
92 pH = -log10(M(3)/1000);
93
94 ### Alternatively uncomment the following lines to disable inhibition
95 pH = 7.5; # Optimal pH range
96 M(1) = 0; # No VFA inhibition
97 M(2) = 0; # No undissociated H2S inhibition
98 #S(14) = 1E-10; # No H2 inhibition (NB: affects product regulation & H2 SR,MP)
99 S(5) = 0; # No total sulphide inhibition
100
101 ### Biological reactions
102 ## Set size of rate matrices.
103 ## Rows contain component, Columns bioreaction number (NR)
104 NoRxn = 19;
105 RXb = zeros (NoXb,NoRxn);
106 RXs = zeros (NoXs,NoRxn);
107 RS = zeros (NoS,NoRxn);
108 RM = zeros (NoM,NoRxn);
109

```

```

110 ## Calculate rates due to each bioreaction
111 NR = 0;
112
113 # Use TEST to toggle reactions on(1) and off(0)
114 TEST = [1,1,1,1,1,1,1,1,1,1,1,1,1,1,1,1]';
115
116 # 1. Protein Hydrolysis
117 # C5H7O2N + H2O -----> C5H9O3N
118 NR = NR + 1;
119 if ((TEST(1)==1))
120     k = 0.69;                                # day-1
121     RXs(1,NR) = -k*Xs(1);                    # Protein consumption
122     RS(9,NR) = -RXs(1,NR);                   # Amino acid production
123 endif
124
125 # 2. Carbohydrate Hydrolysis
126 # C6H10O5 + H2O -----> C6H12O6
127 NR = NR + 1;
128 if ((TEST(2)==1))
129     k = 0.58;                                # day-1
130     RXs(2,NR) = -k*Xs(2);                    # Carbohydrate consumption
131     RS(10,NR) = -RXs(2,NR);                  # Glucose production
132 endif
133
134 # 3. Lipid Hydrolysis
135 # C51H98O6 + 3 H2O -----> CH2OHCHOHCH2OH + 3 CH3(CH2)14COOH
136 NR = NR + 1;
137 if ((TEST(3)==1))
138     k = 0.3;                                # day-1
139     RXs(3,NR) = -k*Xs(3);                    # Lipid consumption
140     RS(11,NR) = -RXs(3,NR);                  # Glycerol production
141     RS(12,NR) = -3*RXs(3,NR);                # Palmitic acid production
142 endif
143
144 # 4. Amino Acid Fermentation
145 # C5H9O3N + 3 H2O -----> 2 CH3COOH + CO2 + H2 + NH3
146 # C5H9O3N + 3 H2O -----> CH3CH2COOH + 2 CO2 + 3 H2 + NH3
147 # C5H9O3N -----> FER + H2O
148 NR = NR + 1;
149 if ((TEST(4)==1))
150     mu_max = 1.5;                            # day-1
151     Y = 0.55*(SMW(9)/XBMW(1));               # mmole/mmole
152     Ks = 20/SMW(9);                          # mmole/l
153     Ac_Pr_ratio = 0.64;                      # Split between PrH and AcH prod. on a molar basis
154                                           # 1 = 100% AcH production, 0 = 100% PrH production
155     KI_H2S = 100 / MMW(2);                   # Undissociated H2S inhibition constant (noncomp)
156     KI_VFA = 10;                             # Undissociated VFA inhibition constant (noncomp)
157
158     RXb(1,NR) = mu_max*Xb(1)*ESU(S(9),Ks)*ENI(M(2),KI_H2S)*ENI(M(1),KI_VFA)...
159         *EPh(pH,a_LL_F,a_UL_F,pH_LL_F,pH_UL_F);
160                                           # Biomass production with H2S, VFA, pH inhib.
161     RS(9,NR) = -RXb(1,NR)/Y;                 # Total amino acid consumption
162     AA_cell = -RXb(1,NR);                    # Amino acids used for cell production only
163     AA_Ac_Pr = RS(9,NR) - AA_cell;           # Amino acids used to produce AcH and PrH
164     RS(1,NR) = -Ac_Pr_ratio*AA_Ac_Pr;        # AcH + Ac- production
165     RS(2,NR) = -(1-Ac_Pr_ratio)*AA_Ac_Pr;    # PrH + Pr- production
166     RS(6,NR) = RS(1,NR)/2 + RS(2,NR)*2;      # CO2 production
167     RS(14,NR) = RS(1,NR)/2 + RS(2,NR)*3;     # H2 production

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168     RS(8,NR) = -AA_Ac_Pr;          # NH3 production
169 endif
170
171 # 5. Glucose Fermentation
172 # C6H12O6 + 2 H2O -----> 2 CH3COOH + 2 CO2 + 4H2
173 # C6H12O6 -----> CH3CH2CH2COOH + 2 CO2 + 2 H2
174 # C6H12O6 -----> 2 CH3CHOHCOOH
175 # 5 C6H12O6 + 6 NH3 -----> 6 FER + 18 H2O
176 NR = NR + 1;
177 if ((TEST(5)==1))
178     k = 268 * (XbMW(1)/SMW(10)); # (Uptake only) mmole glucose/mmole biomass/day
179     Ks = 23/SMW(10);             # mmole glucose/l
180     KI_VFA = 10;                 # Undissociated VFA inhibition constant (noncomp)
181     KI_H2S = 100 / MMW(2);       # Undissociated H2S inhibition constant (noncomp)
182
183     Y_F_AcH = 0.33*(SMW(1)/XbMW(1)); # Prod. based yields: mmole biomass/mmole prod.
184     Y_F_BuH = 0.34*(SMW(3)/XbMW(1));
185     Y_F_LaH = 0.11*(SMW(4)/XbMW(1));
186
187     RGl = -k*Xb(1)*ESU(S(10),Ks)*ENI(S(14),KI_H2)*ENI(M(1),KI_VFA)*ENI(M(2),KI_H2S)...
188     *EpH(pH,a_LL_F,a_UL_F,pH_LL_F,pH_UL_F);
189     # Rate of gl uptake for energy prod. only
190     RS(1,NR) = -2*RGl/(1+S(14)/KI_H2)^2; # Rate of AcH + Ac- production
191     RS(3,NR) = -RGl*(S(14)/KI_H2)/(1+S(14)/KI_H2)^2; # Rate of BuH + Bu- production
192     RS(4,NR) = -2*RGl*(S(14)/KI_H2)/(1+S(14)/KI_H2); # Rate of LaH + La- production
193     RS(6,NR) = RS(1,NR)+2*RS(3,NR); # Rate of CO2 production
194     RS(14,NR) = 2*RS(1,NR)+2*RS(3,NR); # Rate of H2 production
195
196     RXb(1,NR) = Y_F_AcH*RS(1,NR)+Y_F_BuH*RS(3,NR)+Y_F_LaH*RS(4,NR);
197     # Biomass production based on variable yield
198     Gl_cell = -RXb(1,NR)*5/6; # Glucose consumed for cell production only
199     RS(10,NR) = RGl+Gl_cell; # Total glucose consumption
200     RS(8,NR) = Gl_cell; # NH3 + NH4+ consumption
201 endif
202
203 # 6. Lactate Fermentation
204 # CH3CHOHCOOH + H2 -----> CH3CH2COOH + H2O
205 # CH3CHOHCOOH + H2O -----> CH3COOH + CO2 + 2 H2
206 # 5 CH3CHOHCOOH + 3 NH3 -----> 3 laFER + 9 H2O
207 NR = NR + 1;
208 if ((TEST(6)==1))
209     k = 46 * (XbMW(2)/SMW(4)); # Uptake rate constant:
210     # (Energy prod only) mmole LaH/mmole biomass/day
211     Ks = 34/SMW(4); # mmole LaH/l
212     KI_VFA = 10; # Undissociated VFA inhibition constant (noncomp)
213     KI_H2S = 100/MMW(2); # Undissociated H2S inhibition constant (noncomp)
214     Y_lF_PrH = 0.067*(SMW(2)/XbMW(2)); # Prod. based yields: mmole biomass/mmole product
215     Y_lF_AcH = 0.17*(SMW(1)/XbMW(2));
216
217     RLaH = -k*Xb(2)*ESU(S(4),Ks)*ENI(S(14),KI_H2)*ENI(M(1),KI_VFA)*ENI(M(2),KI_H2S)...
218     *EpH(pH,a_LL_F,a_UL_F,pH_LL_F,pH_UL_F);
219     # Rate of LaH uptake for energy prod. only
220     RS(2,NR) = -RLaH*(S(14)/KI_H2)/(1+S(14)/KI_H2); # Rate of PrH + Pr- production
221     RS(1,NR) = -RLaH/(1+S(14)/KI_H2); # Rate of AcH + Ac- production
222     RS(14,NR) = -RS(2,NR)+RS(1,NR)*2; # Rate of H2 production
223     RS(6,NR) = RS(1,NR); # Rate of CO2 production
224     RXb(2,NR) = Y_lF_AcH*RS(1,NR)+Y_lF_PrH*RS(2,NR); # Biomass prod. based on variable yield
225     LaH_cell = -RXb(2,NR)*5/3; # LaH consumed for cell production only

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226     RS(4,NR) = RLah + Lah_cell;           # Total Lah consumption
227     RS(8,NR) = Lah_cell;                   # NH3 + NH4+ consumption
228 endif
229
230 # 7. Glycerol Fermentation
231 # CH2OHCHOHCH2OH + H2O -----> CH3COOH + 3 H2
232 # 2 CH2OHCHOHCH2OH + NH3 -----> FER + CO2 + H2O + 4H2
233 NR = NR + 1;
234 if ((TEST(7)==1))
235     mu_max = 10;                          # day-1
236     Ks = 23/SMW(11);                      # mmole/l
237     Y = 0.55*(SMW(11)/XbMW(1));           # mmole biomass/mmole BuH
238     KI_VFA = 10;                          # Undissociated VFA inhibition constant (comp)
239     KI_H2S = 100/MMW(2);                  # Undissociated H2S inhibition constant (uncomp)
240     RXb(1,NR) = mu_max*Xb(1)*ESU(S(11),Ks)*ENI(M(2),KI_H2S)*ENI(M(1),KI_VFA)...
241                                     *EpH(pH,a_LL_F,a_UL_F,pH_LL_F,pH_UL_F);
242                                     # Biomass production
243
244     RS(11,NR) = -RXb(1,NR)/Y;              # Total glycerol consumption
245     Gly_cell = -RXb(1,NR)/2;               # Glycerol used for cell production
246     Gly_AcH = RS(11,NR) - Gly_cell;        # Glycerol used for AcH production
247     RS(1,NR) = -Gly_AcH;                  # AcH + Ac- production
248     RS(14,NR) = -Gly_cell*2-Gly_AcH*3;    # Hydrogen production
249     RS(6,NR) = RXb(1,NR);                 # Rate of CO2 production
250     RS(8,NR) = -RXb(4,NR);                # NH3 + NH4+ consumption
251 endif
252
253 # 8. Long Chain Fatty Acid Beta Oxidation
254 # CH3(CH2)14COOH +14 H2O -----> 8 CH3COOH + 14 H2
255 # 5 CH3(CH2)14COOH + 16 NH3 + 30 H2O -----> 16 BOB + 14 H2
256 NR = NR + 1;
257 if ((TEST(8)==1))
258     mu_max = 0.12;                        # day-1
259     Ks = 49.8 / SMW(12);                  # mmole/l
260     Y = 0.3*(SMW(4)/XbMW(3));            # mmole biomass/mmole palmitic acid
261     KI_VFA = 10;                          # Undissociated VFA inhibition constant (noncomp)
262     KI_H2S = 100/MMW(2);                  # Undissociated H2S inhibition constant (noncomp)
263
264     RXb(3,NR) = mu_max*Xb(3)*ESU(S(4),Ks)*ENI(S(14),KI_H2)*ENI(M(1),KI_VFA)...
265     *ENI(M(2),KI_H2S)*EpH(pH,a_LL_F,a_UL_F,pH_LL_F,pH_UL_F);
266                                     # Biomass production
267     RS(12,NR) = -RXb(3,NR)/Y;              # Total palmitic consumption
268     Pal_cell = -RXb(3,NR)*5/16;            # Palmitic used for cell production
269     Pal_AcH = RS(12,NR) - Pal_cell;        # Palmitic used for AcH production
270     RS(1,NR) = -Pal_AcH*8;                 # AcH + Ac- production
271     RS(8,NR) = -RXb(3,NR);                # NH3 + NH4+ consumption
272     RS(14,NR) = RS(1,NR)*14/8+RXb(3,NR)*14/16; # H2 production
273 endif
274
275 # 9. BuH Acetogenesis
276 # CH3CH2CH2COOH + 2 H2O -----> 2 CH3COOH + 2H2
277 # 5 CH3CH2CH2COOH + 4 NH3 -----> 4 bACE + 2 H2O + 10 H2
278 NR = NR + 1;
279 if ((TEST(9)==1))
280     mu_max = 0.37;                        # day-1
281     Ks = 7.2/SMW(3);                      # mmole/l
282     Y = 0.085*(SMW(3)/XbMW(4));          # mmole biomass/mmole BuH
283     KI_VFA = 30;                          # Undissociated VFA inhibition constant (comp)

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284     KI_H2S = 26/MMW(2);          # Undissociated H2S inhibition constant (uncomp)
285
286     RXb(4,NR) = mu_max*Xb(4)*S(3)/(Ks*(1+M(1)/KI_VFA)+S(3)*(1+M(2)/KI_H2S))...
287         *ENI(S(14),KI_H2)*EpH(pH,a_LL_MP,a_UL_MP,pH_LL_MP,pH_UL_MP);
288                                     # Biomass production
289     RS(3,NR) = -RXb(4,NR)/Y;      # Total BuH consumption
290     BuH_cell = -RXb(4,NR)*5/4;    # BuH used for cell production
291     BuH_AcH = RS(3,NR) - BuH_cell; # BuH used for AcH production
292     RS(1,NR) = -BuH_AcH*2;        # AcH + Ac- production
293     RS(14,NR) = -BuH_cell*2-BuH_AcH*2; # Hydrogen production
294     RS(8,NR) = -RXb(4,NR);        # NH3 + NH4+ consumption
295 endif
296
297 # 10. PrH Acetogenesis
298 # CH3CH2COOH + 2 H2O -----> CH3COOH + CO2 + 3 H2
299 # 5 CH3CH2COOH + 3 NH3 -----> 3 pACE + 4 H2O + 5 H2
300 NR = NR + 1;
301 if ((TEST(10)==1))
302     mu_max = 0.31;                # day^-1
303     Ks = 758/SMW(2);              # mmole/l
304     Y = 0.064*(SMW(2)/XbMW(5));  # mmole biomass/mmole PrH
305     KI_VFA = 3;                  # Undissociated VFA inhibition constant (comp)
306     KI_H2S = 26.6/MMW(2);        # Undissociated H2S inhibition constant (uncomp)
307
308     RXb(5,NR) = mu_max*Xb(5)*S(2)/(Ks*(1+M(1)/KI_VFA)+S(2)*(1+M(2)/KI_H2S))...
309         *ENI(S(14),KI_H2)*EpH(pH,a_LL_MP,a_UL_MP,pH_LL_MP,pH_UL_MP);
310                                     # Biomass production
311     RS(2,NR) = -RXb(5,NR)/Y;      # Total PrH consumption
312     PrH_cell = -RXb(5,NR)*5/3;    # PrH used for cell production
313     PrH_AcH = RS(2,NR) - PrH_cell; # PrH used for AcH production
314     RS(1,NR) = -PrH_AcH;          # AcH + Ac- production
315     RS(14,NR) = -PrH_cell-PrH_AcH*3; # Hydrogen production
316     RS(6,NR) = -PrH_AcH;          # CO2 production
317     RS(8,NR) = -RXb(5,NR);        # NH3 + NH4+ consumption
318 endif
319
320 # 11. Hydrogen Methanogenesis
321 # 4 H2 + CO2 -----> CH4 + 2 H2O
322 # 5 CO2 + 10 H2 + NH3 -----> hMPB + 8 H2O
323 NR = NR + 1;
324 if ((TEST(11)==1))
325     mu_max = 0.18;                # day^-1
326     Ks = 30E-3/SMW(14);           # mmole/l
327     Y = 0.39*(SMW(14)/XbMW(6));  # mmole biomass/mmole H2
328     KI_VFA = 0.7;                # Undissociated VFA inhibition constant (noncomp)
329     KI_H2S = 664/MMW(2);         # Undissociated H2S inhibition constant (uncomp)
330
331     RXb(6,NR) = mu_max*Xb(6)*EUI(S(14),Ks,M(2),KI_H2S)*ENI(M(1),KI_VFA)...
332         *EpH(pH,a_LL_MP,a_UL_MP,pH_LL_MP,pH_UL_MP);
333                                     # Biomass production
334     RS(14,NR) = -RXb(6,NR)/Y;     # Total H2 consumption
335     H2_cell = -RXb(6,NR)*10;      # H2 used for cell production
336     H2_CH4 = RS(14,NR) - H2_cell; # H2 used for CH4 production
337     RS(13,NR) = -H2_CH4/4;        # CH4 production
338     RS(6,NR) = H2_CH4/4+H2_cell*2; # CO2 consumption
339     RS(8,NR) = -RXb(6,NR);        # NH3 + NH4+ consumption
340 endif
341

```

```

342 # 12. AcH Methanogenesis
343 # CH3COOH -----> CH4 + CO2
344 # 5 CH3COOH + 2 NH3 -----> 2 C5H7O2N + 6 H2O
345 NR = NR + 1;
346 if ((TEST(12)==1))
347     mu_max = 0.357;                # day-1
348     Ks = 154/SMW(1);              # mmole/l
349     Y = 0.04*(SMW(1)/XbMW(7));    # mmole biomass/mmole AcH
350     KI_VFA = 0.7;                 # Undissociated VFA inhibition constant (noncomp)
351     KI_H2S = 117/MMW(2);          # Undissociated H2S inhibition constant (uncomp)
352
353     RXb(7,NR) = mu_max*Xb(7)*EUI(S(1),Ks,M(2),KI_H2S)*ENI(M(1),KI_VFA)...
354         *EPh(pH,a_LL_MP,a_UL_MP,pH_LL_MP,pH_UL_MP);
355     # Biomass production
356     RS(1,NR) = -RXb(7,NR)/Y;      # Total AcH consumption
357     AcH_cell = -RXb(7,NR)*5/2;    # AcH used for cell production
358     AcH_CH4 = RS(1,NR)-AcH_cell;  # AcH used for CH4 production
359     RS(13,NR) = -AcH_CH4;         # CH4 production
360     RS(6,NR) = -AcH_CH4;         # CO2 production
361     RS(8,NR) = -RXb(7,NR);        # NH3 + NH4+ consumption
362 endif
363
364 # 13. H2 Sulphate Reduction
365 # 4 H2 + H2SO4 -----> H2S + 4 H2O
366 # 5 CO2 + 10 H2 + NH3 -----> hMPB + 8 H2O
367 NR = NR + 1;
368 if ((TEST(13)==1))
369     mu_max = 1.17;                # day-1
370     Ks = 4.88E-3/SMW(14);         # mmole/l
371     Y = 0.7*(SMW(14)/XbMW(8));    # mmole biomass/mmole H2
372     KI_VFA = 1;                   # Undissociated VFA inhibition constant (noncomp)
373     KI_H2S = 422/32.06;           # Total sulphide (as S) inhibition constant (uncomp)
374     Ks_SO4 = 20/SMW(7);           # mmole/l
375
376     RXb(8,NR) = mu_max*Xb(8)*EUI(S(14),Ks,S(5),KI_H2S)*ESU(S(7),Ks_SO4)...
377         *ENI(M(1),KI_VFA)*EPh(pH,a_LL_SR,a_UL_SR,pH_LL_SR,pH_UL_SR);
378     # Biomass production
379     RS(14,NR) = -RXb(8,NR)/Y;     # Total H2 consumption
380     H2_cell = -RXb(8,NR)*10;      # H2 used for cell production
381     H2_H2S = RS(14,NR) - H2_cell; # H2 used for H2S production
382     RS(7,NR) = H2_H2S/4;          # SO4 consumption
383     RS(5,NR) = -H2_H2S/4;         # H2S production
384     RS(6,NR) = -RXb(8,NR)*5;      # CO2 consumption
385     RS(8,NR) = -RXb(8,NR);        # NH3 + NH4+ consumption
386 endif
387
388 # 14. AcH Sulphate Reduction
389 # CH3COOH + H2SO4 -----> H2S + 2 CO2 + 2 H2O
390 # 5 CH3COOH + 2 NH3 -----> 2 aSRB + 6 H2O
391 NR = NR + 1;
392 if ((TEST(14)==1))
393     mu_max = 0.36;                # day-1
394     Ks = 46.7/SMW(1);             # mmole/l
395     Y = 0.072*(SMW(1)/XbMW(9));   # mmole biomass/mmole AcH
396     KI_VFA = 1;                   # Undissociated VFA inhibition constant (noncomp)
397     KI_H2S = 35/32.06;            # Total sulphide (as S) inhibition constant (uncomp)
398     Ks_SO4 = 20/SMW(7);           # mmole/l
399

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400   RXb(9,NR) = mu_max*Xb(9)*EUI(S(1),Ks,S(5),KI_H2S)*ESU(S(7),Ks_S04)...
401       *ENI(M(1),KI_VFA)*Eph(pH,a_LL_SR,a_UL_SR,pH_LL_SR,pH_UL_SR);
402                                     # Biomass production
403   RS(1,NR) = -RXb(9,NR)/Y;          # Total AcH consumption
404   AcH_cell = -RXb(9,NR)*5/2;        # AcH used for cell production
405   AcH_H2S = RS(1,NR)-AcH_cell;      # AcH used for H2S production
406   RS(7,NR) = AcH_H2S;               # S04 consumption
407   RS(5,NR) = -AcH_H2S;              # H2S production
408   RS(6,NR) = -AcH_H2S*2;            # CO2 production
409   RS(8,NR) = -RXb(9,NR);            # NH3 + NH4+ consumption
410   endif
411
412   # 15. LaH Sulphate Reduction
413   # 2 CH3CHOHCOOH + H2SO4 -----> 2 CH3COOH + H2S + 2 CO2 + 2 H2O
414   # 5 CH3CHOHCOOH + 3 NH3 -----> 3 lSRB + 9 H2O
415   NR = NR + 1;
416   if ((TEST(15)==1))
417       mu_max = 2.50;                 # day-1
418       Ks = 4.4/SMW(4);               # mmole/l
419       Y = 0.046*(SMW(4)/XbMW(10)); # mmole biomass/mmole LaH
420       KI_VFA = 1;                    # Undissociated VFA inhibition constant (noncomp)
421       KI_H2S = 251/32.06;            # Total sulphide (as S) inhibition constant (noncomp)
422       Ks_S04 = 1/SMW(7);             # mmole/l
423
424   RXb(10,NR) = mu_max*Xb(10)*ESU(S(4),Ks)*ENI(S(5),KI_H2S)*ESU(S(7),Ks_S04)...
425       *ENI(M(1),KI_VFA)*Eph(pH,a_LL_SR,a_UL_SR,pH_LL_SR,pH_UL_SR);
426                                     # Biomass production
427   RS(4,NR) = -RXb(10,NR)/Y;          # Total LaH consumption
428   LaH_cell = -RXb(10,NR)*2/3;        # LaH used for cell production
429   LaH_H2S = RS(4,NR)-LaH_cell;       # LaH used for H2S production
430   RS(7,NR) = LaH_H2S/2;              # S04 consumption
431   RS(5,NR) = -LaH_H2S/2;             # H2S production
432   RS(1,NR) = -LaH_H2S;               # AcH + Ac- production
433   RS(6,NR) = -LaH_H2S;               # CO2 production
434   RS(8,NR) = -RXb(10,NR);            # NH3 + NH4+ consumption
435   endif
436
437   # 16. PrH Sulphate Reduction
438   # CH3CH2COOH + 0.75 H2SO4 -----> 0.75 H2S + CH3COOH + CO2 + H2O
439   # 5 CH3CH2COOH + 3 NH3 -----> 3 pSRB + 4 H2O + 5 H2
440   NR = NR + 1;
441   if ((TEST(16)==1))
442       mu_max = 1.68;                 # day-1
443       Ks = 90.0/SMW(2);               # mmole/l
444       Y = 0.022*(SMW(2)/XbMW(11)); # mmole biomass/mmole PrH
445       KI_VFA = 1;                    # Undissociated VFA inhibition constant (noncomp)
446       KI_H2S = 681/32.06;            # Total sulphide inhibition constant (uncomp)
447       Ks_S04 = 20/SMW(7);            # mmole/l
448
449   RXb(11,NR) = mu_max*Xb(11)*EUI(S(2),Ks,S(5),KI_H2S)*ESU(S(7),Ks_S04)...
450       *ENI(M(1),KI_VFA)*Eph(pH,a_LL_SR,a_UL_SR,pH_LL_SR,pH_UL_SR);
451                                     # Biomass production
452   RS(2,NR) = -RXb(11,NR)/Y;          # Total PrH consumption
453   PrH_cell = -RXb(11,NR)*5/3;        # PrH used for cell production
454   PrH_H2S = RS(2,NR)-PrH_cell;       # PrH used for H2S production
455   RS(7,NR) = PrH_H2S*0.75;           # S04 consumption
456   RS(5,NR) = -PrH_H2S*0.75;          # H2S production
457   RS(1,NR) = -PrH_H2S;               # AcH + Ac- production

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458     RS(6,NR) = -PrH_H2S;          # CO2 production
459     RS(14,NR) = -PrH_cell;         # H2 production
460     RS(8,NR) = -RXb(11,NR);        # NH3 + NH4+ consumption
461 endif
462
463 # 17. BuH Sulphate Reduction
464 # 2 CH3CH2CH2COOH + H2SO4 -----> 4 CH3COOH + H2S
465 # 5 CH3CH2CH2COOH + 4 NH3 -----> 4 bSRB + 2 H2O + 10 H2
466 NR = NR + 1;
467 if ((TEST(17)==1))
468     mu_max = 1.11;                # day-1
469     Ks = 10/SMW(3);                # mmole/l
470     Y = 0.04*(SMW(3)/XbMW(12));    # mmole biomass/mmole BuH
471     KI_VFA = 1;                    # Undissociated VFA inhibition constant (noncomp)
472     KI_H2S = 500/32.06;            # Total sulphide inhibition constant (uncomp)
473     Ks_SO4 = 20/SMW(7);            # mmole/l
474
475     RXb(12,NR) = mu_max*Xb(12)*EUI(S(3),Ks,S(5),KI_H2S)*ESU(S(7),Ks_SO4)...
476         *ENI(M(1),KI_VFA)*Eph(pH,a_LL_SR,a_UL_SR,pH_LL_SR,pH_UL_SR);
477                                     # Biomass production
478     RS(3,NR) = -RXb(12,NR)/Y;       # Total BuH consumption
479     BuH_cell = -RXb(12,NR)*5/4;     # BuH used for cell production
480     BuH_H2S = RS(3,NR)-BuH_cell;    # BuH used for H2S production
481     RS(7,NR) = BuH_H2S/2;           # SO4 consumption
482     RS(5,NR) = -BuH_H2S/2;          # H2S production
483     RS(1,NR) = -BuH_H2S*2;          # AcH + Ac- production
484     RS(14,NR) = -BuH_cell*2;        # H2 production
485     RS(8,NR) = -RXb(12,NR);         # NH3 + NH4+ consumption
486 endif
487
488 # 18. Cell Death
489 NR = NR + 1;
490 if (TEST(18)==1)
491     b = [0.02,0.02,0.01,0.027,0.01,0.013,0.013,0.013,0.013,0.02,0.021,0.02];
492     RXb(:,NR) = -Xb.*(b');
493     RXs(4,NR) = b*Xb;
494 endif
495
496 # 19. Cell Lysis
497 NR = NR + 1;
498 if (TEST(19)==1)
499     k = 1;                          # day-1
500     RXs(4,NR) = -k*Xs(4);           # Dead cell breakdown
501     RXs(1,NR) = -RXs(4,NR);         # Protein production
502 endif
503
504 ### Sum rates
505 j = 19;
506 rateXb = (sum(RXb(:,1:j)))';
507 rateXs = (sum(RXs(:,1:j)))';
508 rateS = (sum(RS(:,1:j)))';
509
510 endfunction
511
512
513
514
515

```

Vapour Liquid Mass Transfer Function—vap_liq.m

```

1  #####
2  #
3  #           High Sulphate Anaerobic Digester Simulation
4  #
5  # Author:      Anthony Knobel
6  # Date:        15 August 1999
7  # File name:   vap_liq.m
8  # Description: This function calculates the mass transfer rates
9  #              of the gas species from the liquid to the vapour
10 #              phase.
11 #
12 #####
13
14 # Local nomenclature
15 # X,P concentration (mmole) and partial pressure (atm) of
16 # 1 H2S
17 # 2 CO2 (Concentration should be CO2(l) + H2CO3)
18 # 3 CH4
19 # 4 H2
20
21 function N = vap_liq(XL,P)
22
23 ## Henry's law constants (atm/(mmole/l)) at 35'C
24 H_H2S = 1.2176E-2;
25 H_CO2 = 0.0376;
26 H_CH4 = 0.744;
27 H_H2 = 1.2726;
28
29 ## Vapour/liquid mass transfer constants (d^-1)
30 kla_H2S = 4320;
31 kla_CO2 = 100;
32 kla_CH4 = 1E6;
33 kla_H2 = 2000;
34
35 ## Calculate vapour/liquid mass transfer rates (mmole/(l.d))
36 N(1) = -kla_H2S*(P(1)/H_H2S-XL(1));
37 N(2) = -kla_CO2*(P(2)/H_CO2-XL(2));
38 N(3) = -kla_CH4*(P(3)/H_CH4-XL(3));
39 N(4) = -kla_H2*(P(4)/H_H2-XL(4));
40
41 endfunction

```

Monod Substrate Uptake and Inhibition Functions

As outlined in section 2.2.2, the overall specific growth rate can be expressed as a product of several factors. This approach was used in the biological rates function `biorates.m`, for which the functions listed below are used. (See table 2.1).

Substrate Uptake—ESU.m

```

1  # Monod substrate utilization function
2
3  function E=ESU(S,Ks)
4      E=S/(Ks+S);

```

```

5  endfunction
6

```

Noncompetitive Inhibition—ENI.m

```

1  # Noncompetitive inhibition function
2
3  function E=ENI(I,KI)
4      E=KI/(KI+I);
5  endfunction
6

```

Noncompetitive Inhibition (Variation)—ENIn.m

```

1  # Noncompetitive inhibition function (modified)
2
3  function E=ENIn(I,KI,n)
4      E=1/(1+(I/KI)^-1);
5  endfunction
6

```

Uncompetitive Inhibition—EUI.m

```

1  # Substrate utilization with uncompetitive inhibition
2
3  function E=EUI(S,Ks,I,KI)
4      E=S/(Ks+S*(1+I/KI));
5  endfunction
6

```

Competitive Inhibition—ECI.m

```

1  # Substrate utilization with competitive inhibition
2
3  function E=ECI(S,Ks,I,KI)
4      E=S/(S+Ks*(1+I/KI));
5  endfunction
6

```

pH Inhibition—EpH.m

```

1  # pH Inhibition function
2
3  function E=EpH(pH,a_LL,a_UL,pH_LL,pH_UL)
4      E=1/((1+exp(-a_LL*(pH-pH_LL)))*(1+exp(a_UL*(pH-pH_UL))));
5  endfunction
6

```

Switching Function—ESF.m

```

1  # Switching function
2
3  function E=ESF(a,C,B)

```

```

4      E=1/(1+exp(a*(C-B)));
5  endfunction
6

```

Rate ‘Packaging’ Function—rate.m

The function `rate.m` is used to combine the biorates, equilibrium and mass transfer functions, and reactor equations into a single function which can be called by the differential or algebraic/differential equation solvers. The format required by Octave’s differential algebraic intergrater, `dassl`, is:

$$\mathbf{res} = f(\mathbf{x}, \frac{d\mathbf{x}}{dt}, t)$$

where $\mathbf{x}$ is a vector describing the state of the system (in this case the reactor concentrations), t is the time and $\mathbf{res}$ is a vector of the residuals (or error) which the solver attempts to set to zero.

Alternatively, with some slight modification, `rate.m` can be used to solve the steady state case. This is done by making `rate.m` only a function of concentration and by setting all the differentials to zero. The solution is then obtained using a non-linear solver.

```

1  # 1 Fermenters
2  # 2 LaH Ferementers
3  # 3 LCFA Oxidizers
4  # 4 BuH Acetogens
5  # 5 PrH Acetogens
6  # 6 H2 MPB
7  # 7 AcH MPB
8  # 8 H2 SRB
9  # 9 AcH SRB
10 # 10 LaH SRB
11 # 11 PrH SRB
12 # 12 BuH SRB
13 # 13 Degradable sludge protein
14 # 14 Degradable sludge carboydrate
15 # 15 Degradable sludge lipid
16 # 16 Dead Biomass
17 # 17 Degradable sludge protein
18 # 18 Degradable sludge carboydrate
19 # 19 Degradable sludge lipid
20 # 20 AcH
21 # 21 PrH
22 # 22 BuH
23 # 23 LaH
24 # 24 H2S
25 # 25 CO2
26 # 26 SO4
27 # 27 NH3
28 # 28 Amino acids
29 # 29 Glucose
30 # 30 Glycerol
31 # 31 Palmitic
32 # 32 CH4
33 # 33 H2
34 # 34 H2S

```

```

35 # 35 CO2
36 # 36 CH4
37 # 37 H2
38
39 function Xdot = rate_mysim(X,t)
40
41 global MW V1 Vv R T X1 Q beta pH SRT HRT QG
42 H = (10^-pH)*1000;
43
44 Ka_H2CO3 = 4.3E-7*1E3;
45 Ka_H2S = 8.9E-8*133;
46 Ka_AcH = 1.7E-5*1E3;
47 Ka_BuH = 1.3E-5*1E3;
48 Ka_PrH = 1.3E-5*1E3;
49 Ka_LaH = 1.3E-4*1E3;
50
51 H2CO3 = X(25)-(Ka_H2CO3/H)/(1+Ka_H2CO3/H)*X(25);
52 H2S1 = X(24)-(Ka_H2S/H)/(1+Ka_H2S/H)*X(24);
53 AcH = X(20)-(Ka_AcH/H)/(1+Ka_AcH/H)*X(20);
54 PrH = X(21)-(Ka_PrH/H)/(1+Ka_PrH/H)*X(21);
55 BuH = X(22)-(Ka_BuH/H)/(1+Ka_BuH/H)*X(22);
56 LaH = X(23)-(Ka_LaH/H)/(1+Ka_LaH/H)*X(23);
57
58 VFA = AcH+PrH+BuH+LaH;
59 FS = H2S1;
60 #TS = X(24);
61
62 KI_H2_pp = 10^(1139/T-7); # atm
63 HH = 1.356; # Hydrogen Henry's constant atm/(mmole/l)
64 KI_H2 = (KI_H2_pp/HH); # mmole/l
65
66 Xb=X(1:12);
67 Xs=X(13:16);
68 S=X(20:33);
69 M=[VFA,FS,H];
70 XbMW=MW(1:12);
71 XsMW=MW(13:16);
72 SMW=MW(20:33);
73 MMW=[-123,MW(24),1.01];
74
75 [rateXb, rateXs, rateS] = biorate_mysim(Xb,Xs,S,M,XbMW,XsMW,SMW,MMW);
76
77 Xdot = zeros(37,1);
78 Xdot(1:12) = SRT^-1*(X1(1:12)-X(1:12))+rateXb;
79 Xdot(13:16) = SRT^-1*(X1(13:16)-X(13:16))+rateXs;
80 Xdot(17:19) = SRT^-1*(X1(17:19)-X(17:19));
81
82 Xdot(20:33) = HRT^-1*(X1(20:33)-X(20:33))+rateS*SRT/HRT;
83
84 XL(1) = H2S1;
85 XL(2) = H2CO3;
86 XL(3) = X(32);
87 XL(4) = X(33);
88 PP(1:4) = X(34:37);
89 N = vap_liq (XL,PP);
90
91 Xdot(24:25) = Xdot(24:25) - N(1:2);
92 Xdot(32:33) = Xdot(32:33) - N(3:4);

```

```

93
94   #QG = 20E7;
95   Xdot(34:37) = (-QG/Vv*X(34:37) + V1/Vv*R*T*N(1:4));
96
97   endfunction

```

Main Calling Routine—septicsim.m

The main routine `septicsim.m`¹, is called from the Octave or Matlab command line. The routine is responsible for some administrative detail such as setting up vectors of the component molecular weights, COD values and ionic charges. The program calls the integrators `dassl` or `lsode` which return a matrix of the reactor concentrations at discrete time intervals. The results of the simulation are then saved.

```

1  #####
2  #
3  #           High Sulphate Anaerobic Digestor Simulation
4  #
5  # Author:      Anthony Knobel
6  # Date:        15 August 1999
7  # File name:   septicsim.m
8  # Description: This is the main file of the dynamic version of
9  #              the model. The file loads the component data,
10 #              sets various parameters and intergrates the model
11 #              dynamics. Solution chemistry and vapour/liquid
12 #              mass transfer is included. Batch, CSTR and high
13 #              rate simulations are possible.
14 #
15 #####
16
17 # 1 Fermenters
18 # 2 LaH Ferementers
19 # 3 LCFA Oxidizers
20 # 4 BuH Acetogens
21 # 5 PrH Acetogens
22 # 6 H2 MPB
23 # 7 AcH MPB
24 # 8 H2 SRB
25 # 9 AcH SRB
26 # 10 LaH SRB
27 # 11 PrH SRB
28 # 12 BuH SRB
29 # 13 Degradable sludge protein
30 # 14 Degradable sludge carboydrate
31 # 15 Degradable sludge lipid
32 # 16 Dead Biomass
33 # 17 Non Degradable sludge protein
34 # 18 Non Degradable sludge carboydrate
35 # 19 Non Degradable sludge lipid
36 # 20 AcH
37 # 21 PrH
38 # 22 BuH
39 # 23 LaH

```

¹'septicsim' stands for septic simulator. It is also a near homonym of scepticism; with, a healthy dose of which, the results of any mathematical model should be veiwed.

```

40 # 24 H2S
41 # 25 CO2
42 # 26 SO4
43 # 27 NH3
44 # 28 Amino acids
45 # 29 Glucose
46 # 30 Glycerol
47 # 31 Palmitic
48 # 32 CH4
49 # 33 H2
50 # 34 H2S
51 # 35 CO2
52 # 36 CH4
53 # 37 H2
54
55 clear
56
57 global X1 MW Q V1 Vv R T beta pH SRT HRT QG
58
59 Q = 20E6;
60 #Vv = 4500;
61 R = 82.06/10E6;
62 T = 273.15+30;
63 pH = 7.0;
64 SRT = 20;
65 HRT = 2;
66 V1 = Q*HRT;
67 Vv = 3E6;
68
69
70 MW = zeros(37,1);
71 COD = zeros(37,1);
72 # Define MWs & CODs
73 for i=1:12
74     MW(i) = 113.12;
75     COD(i) = 1.42;
76 endfor
77
78 MW(13:19) = [114.12,162.14,807.34,114.12,114.12,162.12,807.34]';
79 COD(13:19) = [1.42,1.18,2.87,1.42,1.42,1.18,2.87];
80 MW(20:33) = [60.05,74.08,88.11,90.08,34.08,44.01,96.06,17.03,131.13,180.26,92.09,256.43,16.04,2.0];
81 COD(20:33) = [1.07,1.51,1.82,1.60,1.88,0,0,0,1.22,1.07,1.22,2.87,3.99,7.94];
82 MW(34:37) = [34.08,44.01,16.04,2.02]';
83
84 X1 = zeros(37,1);
85 SO4 = 2000;
86 COD_SO4 = 3;
87 X1(26) = SO4/MW(26);
88
89 ## Sludge composition (dry basis) mg/mg total
90 protein = 0.3847;# 0.2857; #
91 carbohydrate = 0.267;# 0.2857;
92 lipid = 0.281;#0.429; #
93 VFA = 0.067;
94 # Fraction of each sludge component which is degradable
95 f_p = 0.9;
96 f_c = 0.9;
97 f_l = 0.9;

```

```

98
99 # COD of sludge (mgCOD/mg)
100 COD_sludge = protein*COD(13)+carbohydrate*COD(14)+lipid*COD(15)+VFA*COD(20);
101
102 X1(13) = (protein*COD_S04*S04*f_p/COD_sludge)/MW(13);
103 X1(14) = (carbohydrate*COD_S04*S04*f_c/COD_sludge)/MW(14);
104 X1(15) = (lipid*COD_S04*S04*f_l/COD_sludge)/MW(15);
105 X1(16) = 0;
106
107 X1(17) = (protein*COD_S04*S04*(1-f_p)/COD_sludge)/MW(17);
108 X1(18) = (carbohydrate*COD_S04*S04*(1-f_c)/COD_sludge)/MW(18);
109 X1(19) = (lipid*COD_S04*S04*(1-f_l)/COD_sludge)/MW(19);
110
111 X1(20) = (VFA*COD_S04*S04/COD_sludge)/MW(20);
112
113 #X2o = X1;
114 X2o = zeros(37,1);
115 for i=1:12
116     X2o(i) = 0.1;
117 endfor
118 X2o(1) = 3;
119 #X2o(9) = 1;
120
121 QG = 20E7;
122 t_1 = linspace (0,100,100);
123 X2_1 = lsode("rate_mysim",X2o,t_1);
124
125 QG = 2E7;
126 t_2 = linspace (100,200,100);
127 X2o = X2_1(100,:);
128 X2_2 = lsode("rate_mysim",X2o,t_2);
129
130 QG = 2E6;
131 t_3 = linspace (200,300,100);
132 X2o = X2_2(100,:);
133 X2_3 = lsode("rate_mysim",X2o,t_3);
134
135 t = [t_1,t_2,t_3];
136 X = [X2_1;X2_2;X2_3];
137
138 save('shock1.res','X','t');
139
140

```


Appendix C

Aspen Model

The following is a description of the user kinetics Fortran subroutine describing the kinetics of a high sulphate anaerobic system.

Within the Fortran subroutine the following notation is used: XB, XS and S are vectors (type `REAL*8`) of the concentrations of the microbial groups, insoluble components and soluble components respectively. The concentrations are expressed in units of mg/l. Similarly XBMW, XSMW and SMW are vectors of the component molecular weights. The numbering of each component within these vectors is defined in lines 79–141.

A component numbering system specific to the subroutine was used rather than the global Aspen component numbering. This allows components to be added or removed from the Aspen model without the need to alter the numbering within the routine. The two numbering systems are linked using the `KCCIDC()` function as shown in lines 185–225. The Aspen component numbers corresponding to the components names are stored in the integer vectors NXB, NXS and NS. The only restriction imposed by this approach is that the component IDs as input to `KCCIDC()` must correspond exactly to those specified in the Aspen Components.Main form.

All of the component properties as taken from the Aspen databases or supplied by the user are stored in the Aspen PLEX area. In this subroutine only the component molecular weights are required. These are obtained as follows: Lines 57–60 set the array `/PLEX/IB` as common and set B as equivalent to IB. The offset of the molecular weights within IB (or B) is determined using the `IFCMNC()` function in line 228. The molecular weights are then loaded from B and stored in the appropriate arrays (Lines 230–238).

The input vector SOUT contains the component flow rates (kgmole/s) of the feed stream to the reactor in positions 0 to NC. Other variables required from SOUT are the total feed stream flow rate (kgmole/s) at position NC+1; the temperature (K) at position NC+2; the overall stream density (kg/m³) at position NC+8; and the overall molecular weight at position NC+9. Lines 176–179 store these values in the variables F, T, RHO and TOTMW. From these variables the overall volumetric flow rate (l/min) VOLF is calculated (line 180).

Component concentrations of the feed stream are calculated in lines 240–251 from SOUT, the molecular weights and VOLF, and stored in the arrays XB, XS and S.

Rates of production and consumption are calculated as per the stoichiometry and rate equations outlined in section X. The calculated rates are stored in the matrices RXB, RXS and RS (units of mmol.l⁻¹d⁻¹). Each row represents a different component

and each column a different biological reaction. For example RXS(4,3) refers to the rate of production of LCFA (component 4) due to lipid hydrolysis (reaction 3). Summing across the rows will give the overall rates.

A more detailed explanation of the mechanics of the rate calculations is provided using acetate substrate methanogenesis as an example. (Refer to lines 444–475). The IF / END IF block is used to turn the reaction on or off from within the Aspen user interface without the need to modify the code. This is achieved by setting a position in the integer parameter array INT to 0 (off) or 1 (on). For example setting INT(9) to 1 enables acetate methanogenesis. Within the IF block first the relevant parameters (MUMAX,KS, etc) are set. Next MU is calculated as a product of substrate uptake and inhibition factors (see section 2.2.2). A facility is provided for enabling or disabling pH inhibition via the flag PHINHIB. The rate of biomass production and overall acetate consumption are then calculated. From these two rates the rates of acetate being used only for cell production AC_CELL and only for methane production AC_MP are calculated. These two rates are then used as the basis for determining the rates of the remaining components.

The overall reaction rate vector as passed back to Aspen RATES (kgmoles/s) is calculated by summing the matrices RXB, RXS and RS across the rows and multiplying by the reactor volume VOLRC (along with the appropriate unit conversion).

```

1  C      ***** UCT Biological Acid Mine Drainage Treatment Model *****
2  C      *
3  C      *      Name:      USRSAN
4  C      *      Written by: Anthony N. Knobel
5  C      *      Date:      March 1999
6  C      *      Purpose:   Fortran/Aspen function describing biokinetics
7  C      *                  of a high sulphate anaerobic system.
8  C      *                  Function takes stream composition, flowrate etc
9  C      *                  as input and returns rate of production or
10 C      *                  consumption of model components.
11 C      *
12 C      *****
13
14 C-----See Aspen v9.3 reference manuals:
15 C      - Volume 1, Chapter 7 for use of RCSTR with user kinetics
16 C      - Volume 6, Chapter 4 for use of KFORMC and related utilities
17 C      - Volume 6, Chapter 4 and Appendix A for details on
18 C      extracting component properties from the PLEX (IFCMNC & B)
19 C      - Volume 6, Appendix C for stream structure (i.e. SOUT)
20 C      - Volume 6, Chapter 11 for details on user kinetics
21 C      subroutines
22 C      - User Guide volume 2, Chapter 8 for details of accessing
23 C      stream and block variables
24
25
26 C-----Function preliminaries: Parameters to be passed, common variables
27 C      shared with Aspen main, dimension arrays, set data types etc
28
29      SUBROUTINE USRSAN (SOUT,  NSUBS,  IDXSUB,  ITYPE,  NINT,
30      2                  INT,    NREAL,  REAL,    IDS,    NPO,
31      3                  NBOPST, NIWORK, IWORK,   NWORK,  WORK,
32      4                  NC,     NR,     STOIC,   RATES,  FLUXM,
33      5                  FLUXS,  XCURR,  NTCAT,   RATCAT, NTSSAT,
34      6                  RATSSA, KCALL,  KFAIL,   KFLASH, NCOMP,
35      7                  IDX,    Y,      X,       X1,    X2,
```

```

36      8          NRALL,  RATALL,  NUSERV,  USERV,  NINTR,
37      9          INTR,   NREALR,  REALR,   NIWR,   IWR,
38      *          NWR,    WR,      NRL,     RATEL,  NRV,
39      1          RATEV)
40
41  C      Set data types of variables
42      IMPLICIT REAL*8 (A-H,O-Z)
43      REAL K,KS,KSS04,KIH2S,N,M,MU,MUMAX,LA
44
45      DIMENSION SOUT(1), IDXSUB(NSUBS), ITYPE(NSUBS), INT(NINT),
46      2          REAL(NREAL), IDS(2), NBOPST(6,NPO), IWORK(NIWORK),
47      3          WORK(NWORK), STOIC(NC,NSUBS,NR), RATES(1),
48      4          FLUXM(1), FLUXS(1), RATCAT(NTCAT), RATSSA(NTSSAT),
49      5          IDX(NCOMP), Y(NCOMP), X(NCOMP), X1(NCOMP), X2(NCOMP),
50      6          RATALL(NRALL), USERV(NUSERV), INTR(NINTR),
51      7          REALR(NREALR), IWR(NIWR), WR(NWR), RATEL(1), RATEV(1)
52
53  C      Aspen CSTR common variables.
54  C      VOLRC = CSTR volume
55      COMMON /RCSTRR/ VOLRC
56
57  C      Need common PLEX for MW, etc
58      COMMON/PLEX/IB(1)
59      DIMENSION B(1)
60      EQUIVALENCE (IB(1), B(1))
61
62  C      Dimension matrices of concentrations(XB,XS,S), reaction
63  C      rates(RXB,RXS,RS) and Aspen component numbers(NXB,NXS,NS)
64
65      REAL*8 XB(7), XS(3), S(26), RRXB(7), RRXS(3), RRS(26), RTEMP(36),
66      2      XBMW(7), XSMW(3), SMW(26)
67
68      REAL*8 RXB(7,12), RXS(3,12), RS(26,12)
69      INTEGER NXB(7), NXS(3), NS(26)
70
71  C      No. of reaction (NRXN) as numbered below
72  C      & No. of components (NOXB,NOXS,NOS)
73  C      (Values must correspond to matrix sizes above)
74      NRXN = 12
75      NOXB = 7
76      NOXS = 3
77      NOS = 26
78
79  C-----Define nomenclature local to this function
80
81  C      XB = Vector of concentration of microbial groups [mg/l]
82  C      Note: 'Molecular formula' of biomass is C5H7O2N
83
84  C      1 = Hydrolysing and acid fermenting bacteria (HFB)
85  C      2 = LCFA beta oxidizing bacteria (BOB)
86  C      3 = Methane producing bacteria - hydrogen substrate (hMPB)
87  C      4 = Methane producing bacteria - acetate substrate (acMPB)
88  C      5 = Sulphate reducing bacteria - hydrogen substrate (hSRB)
89  C      6 = Sulphate reducing bacteria - acetate substrate (acSRB)
90  C      7 = Sulphate reducing bacteria - propionate substrate (ppRSRB)
91  C      What about Pr Bu La oxidizers (lump with BOB ?)
92
93  C      XS = Vector of insoluble substate concentrations (i.e. sludge

```

```

94  C      components) [mg/l]
95
96  C      1 = Protein fraction of sludge - [C16H28O7N4]n
97  C      2 = Insoluble carbohydrate fraction of sludge - [C6H10O5]n
98  C      3 = Lipid fraction of sludge - generic lipid used i.e.
99  C
100 C      CH2OCO(CH2)14CH3
101 C      |
102 C      CHOCO(CH2)14CH3
103 C      |
104 C      CH2OCO(CH2)14CH3
105
106 C      S = Vector of soluble components concentrations (substrates and
107 C      inhibitors) [mg/l]
108
109 C      1 = Amino acids - C16H30O8N4
110 C      2 = Sugars - C6H12O6
111 C      3 = Glycerol - i.e.
112 C
113 C      CH2OH
114 C      |
115 C      CHOH
116 C      |
117 C      CH2OH
118 C
119 C      4 = Generic LCFA (Palmetic acid) - CH3(CH2)14COOH
120 C      5 = AcH - CH3COOH
121 C      6 = Ac - CH3COO-
122 C      7 = PrH - CH3CH2COOH
123 C      8 = Pr - CH3CH2COO-
124 C      9 = BuH - CH3(CH2)2COOH
125 C      10 = Bu - CH3(CH2)2COO-
126 C      11 = LaH - CH3CHOHCOOH
127 C      12 = La - CH3CHOHCOO-
128 C      13 = H2S
129 C      14 = HS-
130 C      15 = CO2 (Dissolved)
131 C      16 = HCO3-
132 C      17 = H2
133 C      18 = CH4
134 C      19 = NH3
135 C      20 = NH4+
136 C      21 = H2O
137 C      22 = H3O+
138 C      23 = OH-
139 C      24 = SO4--
140 C      25 = HSO4-
141 C      26 = H2SO4
142
143 C-----Matrices (RXB, RXS & RS) of reaction rates of each
144 C      component [mmole/(l.s)], I = component, J = reaction number
145 C      Summing across the rows will give a column vector of total
146 C      reaction rates
147
148 C      Set reaction rate matrices to 0
149 C      DO 20 J=1, NRXN
150 C          DO 10 I = 1, NOXB
151 C              RXB(I,J) = 0

```

```

152 10      CONTINUE
153 20      CONTINUE
154
155      DO 40 J=1, NRXN
156          DO 30 I = 1, NOXS
157              RXS(I,J) = 0
158 30      CONTINUE
159 40      CONTINUE
160
161      DO 60 J=1, NRXN
162          DO 50 I = 1, NOS
163              RS(I,J) = 0
164 50      CONTINUE
165 60      CONTINUE
166
167 C-----Convert Aspen stream input vector (SOUT) to local vectors
168 C      and variables
169 C      Note: All Aspen SOUT flows have units of kgmole/s
170 C      F = total flow (kgmole/s)
171 C      T = Temp(K)
172 C      RHO = overall density (kg/m3)
173 C      TOTMW = overall molecular weight (kg/kgmole)
174 C      VOLF = volumetric flow (l/s)
175
176      F = SOUT(NC+1)
177      T = SOUT(NC+2)
178      RHO = SOUT(NC+8)
179      TOTMW = SOUT(NC+9)
180      VOLF = F*TOTMW/RHO*1000
181
182 C-----Get Aspen numbering corresponding to local numbering and
183 C      store in NXB, NXS & NS
184
185 C      Microbial groups must have these Comp IDs in Components.Main
186      NXB(1) = KCCIDC('HFB')
187      NXB(2) = KCCIDC('BOB')
188      NXB(3) = KCCIDC('HMPB')
189      NXB(4) = KCCIDC('ACMPB')
190      NXB(5) = KCCIDC('HSRB')
191      NXB(6) = KCCIDC('ACSRB')
192      NXB(7) = KCCIDC('PPRSRB')
193
194 C      Insoluble components must have these Comp IDs in Components.Main
195      NXS(1) = KCCIDC('PROTEIN')
196      NXS(2) = KCCIDC('CARB')
197      NXS(3) = KCCIDC('LIPID')
198
199 C      Soluble components must have these Comp IDs in Components.Main
200      NS(1) = KCCIDC('AMINO')
201      NS(2) = KCCIDC('SUGAR')
202      NS(3) = KCCIDC('GLYCEROL')
203      NS(4) = KCCIDC('LCFA')
204      NS(5) = KCCIDC('ACH')
205      NS(6) = KCCIDC('AC-')
206      NS(7) = KCCIDC('PRH')
207      NS(8) = KCCIDC('PR-')
208      NS(9) = KCCIDC('BUH')
209      NS(10) = KCCIDC('BU-')

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210      NS(11) = KCCIDC('LAH')
211      NS(12) = KCCIDC('LA-')
212      NS(13) = KCCIDC('H2S')
213      NS(14) = KCCIDC('HS-')
214      NS(15) = KCCIDC('CO2')
215      NS(16) = KCCIDC('HCO3-')
216      NS(17) = KCCIDC('H2')
217      NS(18) = KCCIDC('CH4')
218      NS(19) = KCCIDC('NH3')
219      NS(20) = KCCIDC('NH4+')
220      NS(21) = KCCIDC('H2O')
221      NS(22) = KCCIDC('H3O+')
222      NS(23) = KCCIDC('OH-')
223      NS(24) = KCCIDC('SO4--')
224      NS(25) = KCCIDC('HSO4-')
225      NS(26) = KCCIDC('H2SO4')
226
227      C-----Load molecular weights from PLEX and store in XBMW, XSMW & SMW
228      LMW = IFCMNC('MW')
229
230      DO 70 I = 1, NOXB
231          XBMW(I) = B(LMW+NXB(I))
232      70  CONTINUE
233      DO 80 I = 1, NOXS
234          XSMW(I) = B(LMW+NXS(I))
235      80  CONTINUE
236      DO 90 I = 1, NOS
237          SMW(I) = B(LMW+NS(I))
238      90  CONTINUE
239
240      C-----Calculate feed stream concentration in mg/l and store in XB, XS & S
241      DO 100 I = 1, NOXB
242          XB(I) = SOUT(NXB(I))*XBMW(I)*1E6/VOLF
243      100 CONTINUE
244
245      DO 110 I = 1, NOXS
246          XS(I) = SOUT(NXS(I))*XSMW(I)*1E6/VOLF
247      110 CONTINUE
248
249      DO 120 I = 1, NOS
250          S(I) = SOUT(NS(I))*SMW(I)*1E6/VOLF
251      120 CONTINUE
252
253      C      Calculate pH
254      PH = -LOG10(S(22)/SMW(22)/1000)
255
256      C      Assuming all rates are based on total substrate concentration
257      C      (i.e. ionized and un-ionized states) convert both to a common basis
258
259      AC = S(5)+S(6)*SMW(5)/SMW(6)
260      PR = S(7)+S(8)*SMW(7)/SMW(8)
261      BU = S(9)+S(10)*SMW(9)/SMW(10)
262      LA = S(11)+S(12)*SMW(11)/SMW(12)
263
264      C      Calculate various rates [mmole/(l.d)]
265      C      NR = Reaction number (initialize to 0)
266      NR = 0
267

```

```

268
269 C-----Primary sludge protein hydrolysis
270 C      [C16H28O7N4]n + nH2O -----> nC16H30O8N4
271
272     IF (INT(1) .EQ. 1) THEN
273         NR = NR + 1
274         K = 0.123
275         XINF = 1.23
276         RXS(1,NR) = -K*(XS(1)-XINF)/XSMW(1)
277         RS(21,NR) = RXS(1,NR)
278         RS(1,NR) = -RXS(1,NR)
279     END IF
280
281 C-----Primary sludge carbohydrate hydrolysis
282 C      [C6H10O5]n + nH2O -----> nC6H12O6
283
284     IF (INT(2) .EQ. 1) THEN
285         NR = NR + 1
286         K = 0.123
287         XINF = 1.23
288         RXS(2,NR) = -K*(XS(2)-XINF)/XSMW(2)
289         RS(21,NR) = RXS(2,NR)
290         RS(2,NR) = -RXS(2,NR)
291     END IF
292
293 C-----Primary sludge lipid hydrolysis
294
295 C      CH2OCO(CH2)14CH3                      CH2OH
296 C      |                                         |
297 C      CHOCO(CH2)14CH3 + 3H2O -----> CHOH + 3CH3(CH2)14COOH
298 C      |                                         |
299 C      CH2OCO(CH2)14CH3                      CH2OH
300
301     IF (INT(3) .EQ. 1) THEN
302         NR = NR + 1
303         K = 0.123
304         XINF = 1.23
305         RXS(3,NR) = -K*(XS(3)-XINF)/XSMW(3)
306         RS(21,NR) = 3*RXS(3,NR)
307         RS(3,NR) = -RXS(3,NR)
308         RS(4,NR) = -RXS(3,NR)*3
309     END IF
310
311 C-----Amino acid fermentation
312 C      C16H30O8N4 + 8H2O -----> 8CH3COOH + H2 + 4NH3
313 C      C16H30O8N4 + 13/3H2 + 8/3H2O -----> 16/3CH3CH2COOH + 4NH3
314 C      C16H30O8N4 -----> 3HFB + CO2 + NH3 + 3H2
315
316     IF (INT(4) .EQ. 1) THEN
317         NR = NR + 1
318         MUMAX = 0.123
319         KS = 0.123
320         YLD = 0.123
321         MU=MUMAX*S(1)/(KS+S(1))
322         RXB(1,NR) = MU*XB(1)/XBMW(1)
323         RS(1,NR) = -MU*XB(1)/(YLD*SMW(1))
324         AA_CELL = -RXB(1,NR)/3
325         AA_AC_PR = RS(1,NR) - AA_CELL

```

```

326          AC_TOT = 0.5
327          AA_AC = AC_TOT*AA_AC_PR
328          AA_PR = (1-AC_TOT)*AA_AC_PR
329          RS(21,NR) = 8*AA_AC+8/3*AA_PR
330          RS(5,NR) = -8*AA_AC
331          RS(7,NR) = -16/3*AA_PR
332          RS(17,NR) = -AA_AC + 13/3*AA_PR - 3*AA_CELL
333          RS(19,NR) = -4*AA_AC - 4*AA_PR - AA_CELL
334          RS(15,NR) = -AA_CELL
335      END IF
336
337  C-----Sugar fermentation
338  C      C6H12O6 + 2H2O -----> 2CH3COOH + 2CO2 + 4H2
339  C      C6H12O6 + 2H2 -----> 2CH3CH2COOH + 2H2O
340  C      C6H12O6 -----> 2CH3CHOHCOOH
341
342  *      IF (INT(5) .EQ. 1) THEN
343  *          NR = NR + 1
344  *          MUMAX = 1.23
345  *          KS = 1.23
346  *          YLD = 0.123
347  *      END IF
348
349  C-----Glycerol fermentation
350
351  C      CH2OH
352  C      |
353  C      CHOH -----> CH3CH2COOH + H2O
354  C      |
355  C      CH2OH
356
357  C      CH2OH
358  C      |
359  C      CHOH + H2O -----> CH3COOH + CO2 + 3H2
360  C      |
361  C      CH2OH
362
363  C      CH2OH
364  C      |
365  C      2 CHOH + NH3 -----> HFB + CO2 + 2H2O + 4H2
366  C      |
367  C      CH2OH
368
369  IF (INT(6) .EQ. 1) THEN
370      NR = NR + 1
371      MUMAX = 0.123
372      KS = 0.123
373      YLD = 0.123
374      MU=MUMAX*S(3)/(KS+S(3))
375      RXB(1,NR) = MU*XB(1)/XBMW(1)
376      RS(3,NR) = -MU*XB(1)/(YLD*SMW(3))
377      GL_CELL = -RXB(1,NR)*2
378      GL_AC_PR = RS(3,NR) - GL_CELL
379      AC_TOT = 0.5
380      GL_AC = AC_TOT*GL_AC_PR
381      GL_PR = (1-AC_TOT)*GL_AC_PR
382      RS(5,NR) = -GL_AC
383      RS(7,NR) = -GL_PR

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```

384          RS(21,NR) = -GL_PR + GL_AC - GL_CELL
385          RS(15,NR) = -GL_AC - GL_CELL/2
386          RS(17,NR) = -GL_AC*3 -GL_CELL*4/2
387          RS(19,NR) = GL_CELL/2
388      END IF
389
390  C-----LCFA beta oxidation
391  C      CH3(CH2)14COOH + 14H2O -----> 8CH3COOH + 14H2
392  C      CH3(CH2)14COOH + 6H2O + 3NH3 -----> 3BOB + CO2 + 16H2
393
394      IF (INT(7) .EQ. 1) THEN
395          NR = NR + 1
396          MUMAX = 0.123
397          KS = 0.123
398          YLD = 0.123
399          MU=MUMAX*S(4)/(KS+S(4))
400          RXB(2,NR) = MU*XB(2)/XBMW(2)
401          RS(4,NR) = -MU*XB(2)/(YLD*SMW(4))
402          LCFA_CELL = -RXB(2,NR)/3
403          LCFA_BO = RS(4,NR) - LCFA_CELL
404          RS(5,NR) = -LCFA_BO*8
405          RS(21,NR) = LCFA_BO*14 + LCFA_CELL*6
406          RS(17,NR) = -LCFA_BO*14 -LCFA_CELL*16
407          RS(19,NR) = LCFA_CELL*3
408          RS(15,NR) = -LCFA_CELL
409      END IF
410
411  C-----Methanogenesis hydrogen substrate
412  C      4H2 + CO2 -----> CH4 + 2H2O
413  C      5CO2 + NH4+ + 10H2 -----> hMBP + 7H2O + H3O+
414
415      IF (INT(8) .EQ. 1) THEN
416          NR = NR + 1
417          MUMAX = 0.1813
418          KS = 0.1338
419          YLD = 0.38906
420  C      B = 0.013
421          KIH2S = 664.301
422
423          PHINHIB = 0
424          IF (PHINHIB .EQ. 1) THEN
425              PH_LL = 1.23
426              PH_UL = 1.23
427              M = 1.23
428              ZPHHMP = ((PH-PH_LL)/(PH_UL-PH_LL))**M
429          ELSE
430              ZPHHMP = 1
431          END IF
432          MU = MUMAX*S(17)/(KS+S(17)*(1+S(13)/KIH2S))*ZPHHMP
433          RXB(3,NR) = MU*XB(3)/XBMW(3)
434          RS(17,NR) = -MU*XB(3)/(YLD*SMW(17))
435          H2_CELL = -RXB(3,NR)*10
436          H2_MP = RS(17,NR) - H2_CELL
437          RS(15,NR) = H2_MP/4 + H2_CELL*5/10
438          RS(18,NR) = -H2_MP/4
439          RS(21,NR) = -H2_MP*2/4 - H2_CELL*7/10
440          RS(20,NR) = H2_CELL/10
441          RS(22,NR) = -H2_CELL/10

```

```

442         END IF
443
444 C-----Methanogenesis acetate substrate
445 C   CH3COOH -----> CH4 + CO2
446 C   5CH3COOH + 2NH4+ -----> 2acMPB + 2H3O+ + 4H2O
447
448     IF (INT(9) .EQ. 1) THEN
449         NR = NR + 1
450         MUMAX = 0.1368
451         KS = 27.1028
452         YLD = 0.04066
453 C     B = 0.013
454         KIH2S = 116.917
455
456         PHINHIB = 0
457         IF (PHINHIB .EQ. 1) THEN
458             PH_LL = 1.23
459             PH_UL = 1.23
460             M = 1.23
461             ZPHACMP = ((PH-PH_LL)/(PH_UL-PH_LL))**M
462         ELSE
463             ZPHACMP = 1
464         END IF
465         MU = MUMAX*AC/(KS+AC*(1+S(13)/KIH2S))*ZPHACMP
466         RXB(4,NR) = MU*XB(4)/XBMW(4)
467         RS(5,NR) = -MU*XB(4)/(YLD*SMW(5))
468         AC_CELL = -RXB(4,NR)*5/2
469         AC_MP = RS(5,NR) - AC_CELL
470         RS(21,NR) = - AC_CELL*4/5
471         RS(18,NR) = -AC_MP
472         RS(15,NR) = -AC_MP
473         RS(20,NR) = AC_CELL*2/5
474         RS(22,NR) = -AC_CELL*2/5
475     END IF
476
477 C-----Sulphate reduction hydrogen substrate
478 C   H2 + 0.25SO4-- +0.25H3O+ -----> 0.25HS- + 1.25H2O
479 C   5CO2 + NH4+ + 10H2 -----> hSRB + 7H2O + H3O+
480
481     IF (INT(10) .EQ. 1) THEN
482         NR = NR + 1
483         MUMAX = 0.1804
484         KS = 0.11268
485         KSSO4 = 0.123
486         YLD = 0.32554
487 C     B = 0.013
488         KIH2S = 148.8035
489
490         PHINHIB = 0
491         IF (PHINHIB .EQ. 1) THEN
492             PH_LL = 1.23
493             PH_UL = 1.23
494             M = 1.23
495             ZPHHSR = ((PH-PH_LL)/(PH_UL-PH_LL))**M
496         ELSE
497             ZPHHSR = 1
498         END IF
499         MU = MUMAX*S(17)/(KS+S(17)*(1+S(13)/KIH2S))*S(24)/(KSSO4+S(24))*ZPHHSR

```

```

500      RXB(5,NR) = MU*XB(5)/XBMW(5)
501      RS(17,NR) = -MU*XB(5)/(YLD*SMW(17))
502      H2_CELL = -RXB(5,NR)*10
503      H2_SR = RS(17,NR) - H2_CELL
504      RS(24,NR) = 0.25*H2_SR
505      RS(22,NR) = 0.25*H2_SR
506      RS(14,NR) = -0.25*H2_SR
507      RS(21,NR) = -1.25*H2_SR - H2_CELL*7/10
508      RS(15,NR) = H2_CELL*5/10
509      RS(20,NR) = H2_CELL/10
510      RS(22,NR) = -H2_CELL/10
511  END IF
512
513  C-----Sulphate reduction acetate substrate
514  C      CH3COOH + SO4-- ----> H2S + 2HCO3-
515  C      5CH3COOH + 2NH4+ ----> 2acSRB + 2H3O+ + 4H2O
516
517      IF (INT(11) .EQ. 1) THEN
518          NR = NR + 1
519          MUMAX = 0.1058
520          KS = 46.729
521          KSSO4 = 0.123
522          YLD = 0.02461
523  C      B = 0.013
524          KIH2S = 8.503
525
526          PHINHIB = 0
527          IF (PHINHIB .EQ. 1) THEN
528              PH_LL = 1.23
529              PH_UL = 1.23
530              M = 1.23
531              ZPHACSR = ((PH-PH_LL)/(PH_UL-PH_LL))**M
532          ELSE
533              ZPHACSR = 1
534          END IF
535          MU = MUMAX*AC/(KS+AC*(1+S(13)/KIH2S))*S(24)/(KSSO4+S(24))*ZPHACSR
536          RXB(6,NR) = MU*XB(6)/XBMW(6)
537          RS(5,NR) = -MU*XB(6)/(YLD*SMW(5))
538          AC_CELL = -RXB(6,NR)*5/2
539          AC_SR = RS(5,NR) - AC_CELL
540          RS(24,NR) = AC_SR
541          RS(13,NR) = -AC_SR
542          RS(16,NR) = -2*AC_SR
543          RS(20,NR) = 2*AC_CELL/5
544          RS(22,NR) = -2*AC_CELL/5
545          RS(21,NR) = -4*AC_CELL/5
546      END IF
547
548  C-----Sulphate reduction propionate substrate (partial oxidation)
549  C      CH3CH2COOH + 0.75SO4-- +0.25H2O ---->
550  C      CH3COOH + HCO3- +0.75HS- + 0.25H3O+
551  C      5CH3CH2COOH + 3NH4+ ----> 3pprSRB + H2O + 5H2 + 3H3O+
552
553      IF (INT(12) .EQ. 1) THEN
554          NR = NR + 1
555          MUMAX = 0.1506
556          KS = 27.152
557          KSSO4 = 0.123

```

```

558      YLD = 0.04832
559  C      B = 0.021
560      KIH2S = 206.199
561
562      PHINHIB = 0
563      IF (PHINHIB .EQ. 1) THEN
564          PH_LL = 1.23
565          PH_UL = 1.23
566          M = 1.23
567          ZPHPPRSR = ((PH-PH_LL)/(PH_UL-PH_LL))*M
568      ELSE
569          ZPHPPRSR = 1
570      END IF
571      MU = MUMAX*PR/(KS+PR*(1+S(13)/KIH2S))*S(24)/(KSS04+S(24))*ZPHPPRSR
572      RXB(7,NR) = MU*XB(7)/XBMW(7)
573      RS(7,NR) = -MU*XB(7)/(YLD*SMW(7))
574      PR_CELL = -RXB(7,NR)*5/3
575      PR_SR = RS(7,NR)-PR_CELL
576      RS(24,NR) = 0.75*PR_SR
577      RS(5,NR) = -PR_SR
578      RS(16,NR) = -PR_SR
579      RS(14,NR) = -0.75*PR_SR
580      RS(22,NR) = -PR_SR*0.25 - PR_CELL*3/5
581      RS(21,NR) = PR_SR*0.25 - PR_CELL/5
582      RS(20,NR) = PR_CELL*3/5
583      RS(17,NR) = -PR_CELL
584
585      END IF
586
587  C-----Return rate to Aspen in Aspen format [kgmoles/s]
588  C      i.e. Sum rate matrices to get total rate vector
589  C      & multiply by reactor volume (with unit conversion)
590
591      DO 155 I = 1, NC
592          RATES(I) = 0D0
593      155 CONTINUE
594
595      DO 170 J = 1,NOXB
596          RRXB(J) = 0
597          DO 160 I = 1,NRXN
598              RRXB(J) = RRXB(J) + RXB(J,I)
599      160 CONTINUE
600          RATES(NXB(J)) = RRXB(J)/8.64E7*VOLRC
601      170 CONTINUE
602
603      DO 190 J = 1,NOXS
604          RRXS(J) = 0
605          DO 180 I = 1,NRXN
606              RRXS(J) = RRXS(J) + RXS(J,I)
607      180 CONTINUE
608          RATES(NXS(J)) = RRXS(J)/8.64E7*VOLRC
609      190 CONTINUE
610
611      DO 210 J = 1,NOS
612          RRS(J) = 0
613          DO 200 I = 1,NRXN
614              RRS(J) = RRS(J) + RS(J,I)
615      200 CONTINUE

```

```
616          RATES(NS(J)) = RRS(J)/8.64E7*VOLRC
617 210  CONTINUE
618
619      RETURN
620      END
621
```

Appendix D

Miscellaneous Calculations

Maximum Theoretical Sulphate Removal

Assuming that:

- No biological reactions other sulphate reduction occur,
- No biomass production of sulphate reducers occurs (i.e. all COD is used for sulphate reduction),
- All carbon in the substrate is oxidized to CO_2 ,
- All hydrogen in the substrate is oxidized to H_2O ,
- Sulphate is the sole source of oxygen,

then:

$$\begin{aligned} 1 \text{ mole COD} &\equiv 0.25 \text{ mole SO}_4^{2-} \\ 1 \text{ mole COD} \times 15.99 \text{ g COD/gmole COD} &\equiv 0.25 \text{ mole SO}_4^{2-} \times 96.06 \text{ g SO}_4^{2-}/\text{gmole SO}_4^{2-} \\ 1 \text{ g COD} &\equiv 1.5 \text{ g SO}_4^{2-} \end{aligned}$$

The actual sulphate removal will be less in a pure sulphate reducing system owing to biomass formations. In a mixed system sulphate removal per COD will be much less, particularly if the carbon source is complex and needs to pass through acidogenesis first.