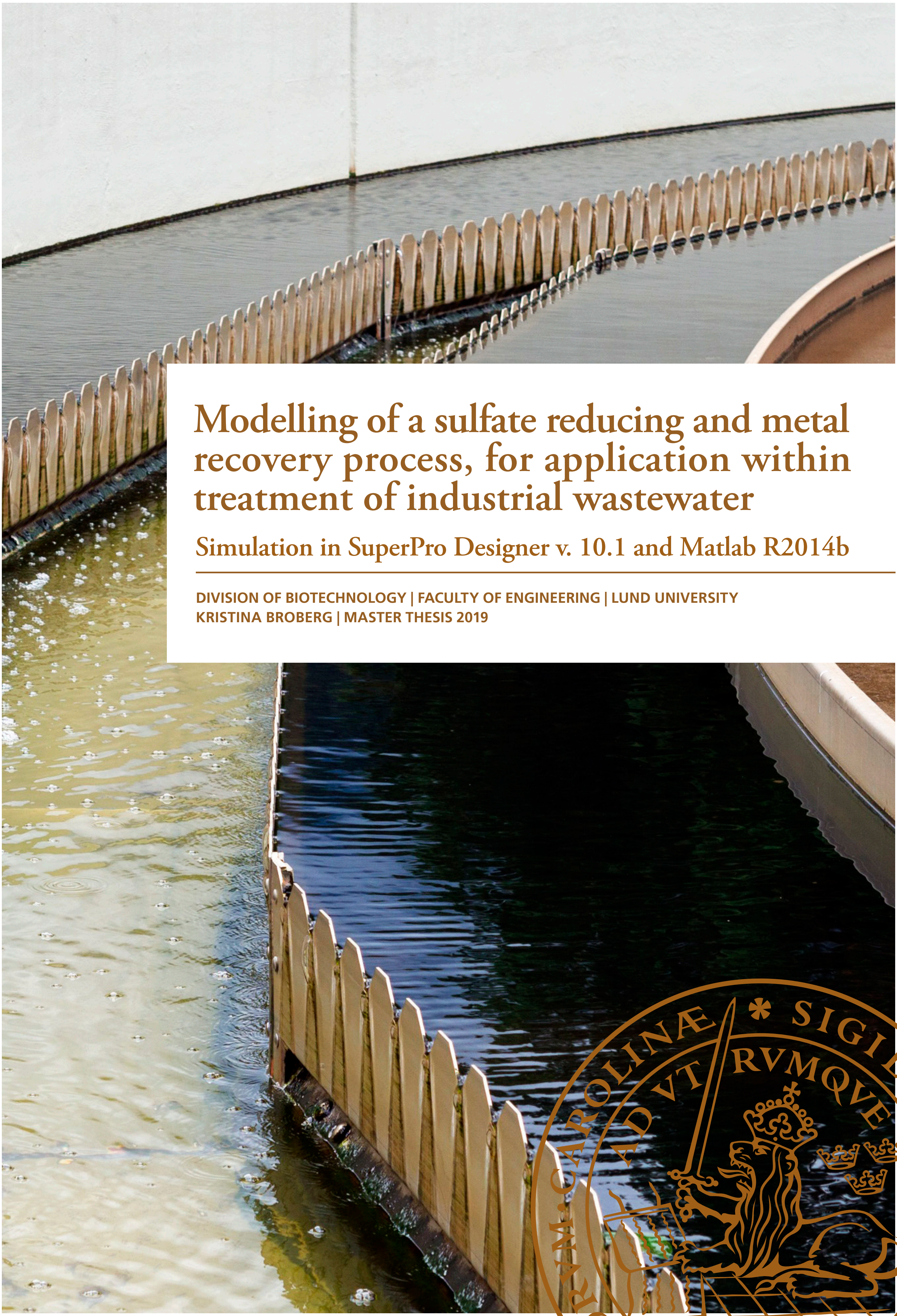


Modelling of a sulfate reducing and metal recovery process, for application within treatment of industrial wastewater

Simulation in SuperPro Designer v. 10.1 and Matlab R2014b

DIVISION OF BIOTECHNOLOGY | FACULTY OF ENGINEERING | LUND UNIVERSITY
KRISTINA BROBERG | MASTER THESIS 2019

Modelling of a sulfate reducing and metal recovery process, for application within treatment of industrial wastewater

Simulation in SuperPro Designer v. 10.1 and Matlab R2014b

Kristina Broberg

Master Thesis in Engineering, Biotechnology
Faculty of Engineering, Lund University
A collaboration with Research Institutes of Sweden (RISE)
Lund: 2019-01-25

Supervisors:	Carl Grey, Associate Professor at Division of Biotechnology, Lund University Erika Lönntoft, Research Institutes of Sweden (RISE)
Examiner:	Patrick Adlercreutz, Professor at Division of Biotechnology, Lund University

LUNDS UNIVERSITET
Lunds Tekniska Högskola

Acknowledgments

A special thank you to Erika for being my main supervisor at RISE and supporting and encouraging me through this process. Further, I would like to thank my supervisor Carl Grey at the Division of Biotechnology at Faculty of Engineering, Lund University for your support and for bringing up many ideas for solutions and aspects in the process. I would like to thank my other supervisors at RISE, Karin and Sudha, for being helpful and constantly interested in the progress of the project. I would like to thank RISE for providing me with the resources, time and space to execute my master thesis - it has been a very interesting and meaningful past six months.

A huge thank you to my friend, and office buddy Marika, for making the process a whole lot more fun, making the most out of THE office and for accepting my rants as things didn’t go the way I planned...several times a day.

As always, thank you to my family for endless support and my friends for keeping me sane.

Abstract

This thesis treats the development of an industrial scale models of a wastewater treatment process aiming at reducing the sulfate and heavy metal concentration in the effluent water from mining processes. The sulfate present in the process water is reduced using sulfate reducing bacteria (SRB) while the heavy metals are recovered through selective precipitation using the hydrogen sulfide produced by the SRB. Development of efficient treatment processes of both municipal, and maybe especially for industrial wastewaters such as leachate are an important step towards ensuring a future water supply for both the industry as well as for everyday consumers.

Two models of the process has been developed, one in the software SuperPro and another using the software Matlab. The SuperPro model was developed using a stoichiometric fermenter as its bioreactor whereas the Matlab model takes the kinetics of the bioreactor into consideration. The SuperPro model generated results that were consistent with the input data. The Matlab model on the other hand, did not display the correct Monod kinetic on the graphs generated when solving the differential equations.

The future steps of this project is to further develop the models. The SuperPro model needs to be validated against experimental results, and developed using a kinetic reactor instead of the current stoichiometric one. The Matlab model is in need of further development of the connection between the mass balances in the different steps of the process.

Sammanfattning

Denna masteruppsats syftar till utvecklandet av en industriellt, fullskalig modell över en vattenreningsprocess som syftar till simultan sulfatreduktion och tungmetallåtervinning från processvatten vid gruvdrift. Den mängd sulfat som finns tillgänglig i lakvattnet, omvandlas till vätesulfid med hjälp av sulfatreducerande bakterier (SRB). Tungmetallerna återvinns genom selektiv utfällning med hjälp av den vätesulfid som bildats av SRB.

Två modeller av processen har tagits fram, en baserad på mjukvaran SuperPro och den andra baserad på Matlab. SuperPro-modellen utvecklades kring en stökiometrisk bioreaktor medan Matlab-modellen utvecklats kring differentialekvationer som tar hänsyn till reaktionskinetiken. SuperPro-modellen genererade konsekventa och upprepningsbara resultat som överensstämde med indata. Matlab-modellen genererade precis som SuperPro-modellen konsekventa resultat, om än något oväntade – speciellt gällande den låga konsumptionen av acetat i modellen som innehöll båda substraten.

Framtida rekommendationer för detta projekt är vidareutveckling av dessa två modeller. SuperPro-modellen utvecklas bäst genom validering mot experimentell data samt genom införandet av en kinetiskt baserad bioreaktor istället för den nuvarande stökiometriska. Matlab-modellen behöver vidareutveckling främst inom kopplingarna mellan de beskrivande differentialekvationerna för modellens olika processteg.

Table of contents

List of Figures

List of Tables

Abbreviations and commonly used expressions

ABBREVIATION	EXPLANATION	UNIT
COD	Chemical oxygen demand, in this report the term is referring to the required amount of substrate associated with reduction of sulfate.	g
Theoretical COD	Theoretical chemical oxygen demand, refers to the theoretically (general) determined substrate requirement associated with sulfate reduction. Depends on substrate specific COD ratios (1.50 for methanol, 1.07 for acetate) and the general ratio is determined at 0.67 g COD/g SO_4^{2-} (Iowa State University, n.d.).	g
Experimental COD	Experimental chemical oxygen demand, refers to experimentally determined chemical oxygen demand (as previously defined).	g
$\mathbf{k_H}$	Henry's constant	atm*L/mol
$\mathbf{p_g}$	Partial gas pressure, specific to a gas	atm
Yg	Mole fraction of gas found in the liquid phase	-
n	Amount of substance	mol
R	Ideal gas constant	atm*L/K*mol
T	Temperature	K
V	Volume	L
c	Concentration	mol/L
$\mathbf{pK_a}$	Acid dissociation constant	-
$\mathbf{K_a}$	Acidic equilibrium constant	-
q	Volumetric flow, specified for each instance where its used	L/h
$\boldsymbol{\mu_{max}}$	Maximum growth rate	h^{-1}
$\mathbf{S_x}$	Liquid concentration of substance x	mol/L
$\mathbf{K_{s,x}}$	Saturation constant for substance x	mol/L
$\mathbf{I_{H2S}}$	Inhibition constant for hydrogen sulfide (unspecified species)	-
$\mathbf{I_{pH}}$	Inhibition constant for pH inhibition	-
X	Concentration of biomass	mol/L
$\mathbf{Y_{X/SO4}}$	Biomass formed on sulfate	-
$\mathbf{Y_{X/Ac}}$	Biomass formed on acetate	-
$\mathbf{r_x}$	Reaction rate for consumption or production of substance x	mol/L*h
$\mathbf{V_{liq}}$	Liquid volume	L
$\mathbf{V_{tank}}$	Liquid volume of the tank	L
$\mathbf{S_{red}}$	Concentration of reduced sulfur	mol/L
$\mathbf{S_{prim,X}}$	Primary/starting concentration of compound X in gaseous phase	mol/L
$\mathbf{S_{H+,loc}}$	Concentration of H^+ in a specific tank or setting	mol/L
$\boldsymbol{\rho_x}$	Rate of formation of compound x	mol/L*h
$\boldsymbol{\rho_{A,x}}$	Rate of acid-base equilibrium for compound x	mol/L*h
dt	Time step in differential equation	h

k	Reaction rate constant	h^{-1}
$\mathbf{k_{Lx}}$	Mass transfer rate coefficient for compound x	h^{-1}

1. Introduction and justification

During the past couple of summers and with the summer of 2018 in recent memory, water scarcity has begun to affect Sweden. During this past summer, consumers were urged to reduce their water usage, to avoid watering lawns and to avoid taking long showers. However, the everyday consumers are not the main users of freshwater in Sweden - the process and mining industry are. As a step towards ensuring the future water supply for industrial use as well as the supply and quality for everyday consumers, the development of sustainable and efficient water treatment processes is necessary.

With this in mind, the foundation for this project is the need for efficient and sustainable treatment processes of industrial process water. The main focus of this project is the sulfate reduction of process wastewater from the mining industry as well as a possibility of creating value from the wastewater stream through precipitation of metal sulfides. Further, the biological sulfate reduction used in this process has several other possible areas of application, such as the food technology industry which indicates good prospects for future use of the process (Hao, et al., 2014; Barrera, et al., 2015).

This project is based around modelling of the wastewater treatment process using two software's, SuperPro Designer and Matlab. By creating reliable models of processes, one can reduce the amount of experiments one actually has to perform and at the same time get more accurate predictions of the experiments one has to perform. Therefore, accurate models of real life processes are very powerful tools when developing safe and efficient processes.

1.1 Project associations

1.1.1 METGROW+

METGROW+ is an EU-project spanning over several different mining industries with two main focuses. The first branch aims at adapting and streamlining the mining process, trying to create the most cost- and resource efficient process. The second, focuses on innovation of new unit operations for mining processes. The project gathers knowledge from different partner organizations such as the mining industry, research institutes and social sciences.

METGROW+ have a focus on both industrially essential metals such as zinc (Zn), copper (Cu) and tin (Sn) as well as so called critical metals (for example chromium (Cr), cobalt (Co) and antimony (Sb)) (European Commission, 2018A; European Commission, 2018B; European Commission, 2018C).

The Matlab based model that is used as basis for this report and following simulation, was developed with METGROW+'s expected impact and objectives in mind (Kvarnström & Lönntoft, 2017).

1.1.2 SO_4-control

SO_4-control is a project sponsored by the European Commission and was launched by EIT raw materials in 2018. The project is a sequel project to the previous EIT based project Ariel (2015). (EIT RawMaterials, 2018A).

There are three main constituents to the SO_4-control project. The first, optimization of the leaching processes, focusing on sulfate control. The second, includes membrane technologies which allows for a high concentration of metal sulfates being formed. The third area focuses on developing solutions for sulfate reduction which is adapted to the Nordic climate (colder climate compared to existing processes) (EIT RawMaterials, 2018B).

This project and report aims at combining the third phase of SO_4-control, with the two previous ones in a complete process simulation.

1.2 Project description

1.2.1 Aim

The purpose of this master thesis is to further develop the sulfate reducing bacteria (SRB) continuously stirred tank (CSTR) process model, curtesy of RISE, through addition of up- and downstream process steps seen from the bioreactor in the simulation software SuperPro as well as in Matlab. As the process depends on the production of hydrogen sulfide, a very toxic gaseous compound, the process that is simulated should take the risks associated with the gas into consideration and minimize the possibilities of the gas escaping the reaction vessels.

1.2.2 Scope

The scope of the project is to develop an industrial scale SuperPro model of the sulfate reducing and metal recovering process as well as expansion of the existing Matlab script to include the precipitation of metal sulfides. Additionally, an investigation of the differences in output due to changes in substrate and substrate composition was carried out. Finally, two of the cases (case 1 and 3) were simulated using the full scale Matlab model which in turn was compared to the results received from the SuperPro simulation.

The scope of this project does not include any cost basis analysis or any analysis regarding the energy balances and consumption of the process. Further, the main focus of this report will be the performance of the bioreactor and sulfate reduction whereas the precipitation steps associated with the process will be of less significance

Disclaimer: The data used to run the simulations in this report was kindly provided by RISE. Due to existing confidentiality agreements, no additional information of the source of the data will be provided.

2. Background

2.1 The mining industry and water usage

In a report from 2000, Sweden Statistics concluded that everyday use made up 35% of the annual consumption of freshwater whereas process and mining industry required 55%. The same report estimated that the process and mining industry used approximately 1 500 Mm^3 ground and freshwater in 2000, to meet production demands (SCB, 2000). More recent numbers (2015) show that the total industry water disposal, over all industries and with varying degree of pollution, was approximated to 70.3 Mm^3 into the municipal wastewater system and approximately 812 Mm^3 into privately owned lakes or streams (SCB, 2017A). In 2015, the Swedish mining industry used 38 Mm^3 process water and an additional 11 Mm^3 for additional production related purposes (SCB, 2017B).

The sulfate concentration, the metals found in the process water and their concentrations vary greatly between mining sites due to the natural geological variation of the rock types present (Sveriges geologiska undersökning, n.d.; Boliden, 2018; V.Shadrunova & Orekhova, 2015). However, the leachate stream composition used in this report was adapted from previous RISE projects and contained 0.7 mmol Cu^{2+}/L, 17.4 mmol Zn^{2+}/L and 117.4 mmol/L of other metal ions.

2.2 Sulfate reducing microorganisms

Sulfate reducing microorganisms are members of the chemolithotrophic kingdom. This group includes both bacteria and archaea that utilize sulfate as their terminal electron acceptor, thus coupling anaerobic respiration to ATP synthesis. The reaction through which sulfated is reduced is commonly referred to as dissimilatory sulfate reduction. For purpose of this report the abbreviation SRB (sulfate reducing bacteria) includes both bacteria and archaea with the ability to reduce sulfate. Currently, more than 220 species of SRB with very varying characteristics and properties have been identified (Barton & Fauque, 2009).

2.2.1 Metabolism and metabolic pathways

There are two types of SRB with different electron-donor metabolisms - those with the ability to completely oxidize organic material to carbon dioxide (CO_2) and those that are incomplete oxidizers of organic material, with the final product being acetate. The SRB responsible for complete oxidation of organic material have proven to be able to utilize acetate as their main carbon source. These species perform energy generation through either a modified version of the citric acid cycle (*Desulfobacter postgatei*) or through the acetyl-CoA pathway (*Desmulfobacterium, Desulfotomaculum, Desulfococcus* species and *Desulfobacca acetoxidans*) (Muyzer & Stams, 2008).

Due to the differences in electron-donor metabolism, there are several organic substrates the can serve as electron donor to SRB. Methanol and acetate are the two that were considered for this work, under the assumption that usage of acetate resulted in complete oxidation of the substrate. Growth on methanol under mesophilic conditions, does however result in a low sulfate reduction and high methane production. Weijma *et al.* (2001), found that approximately 90% of the methanol resulted in methane and that only 0.4 g SO_4^{2-}/L×d was reduced to hydrogen sulfide under mesophilic conditions and when methanol was used as substrate.

Using methanol as carbon source, results in a low growth rate and a temperature sensitivity for the process (evaporation and spontaneous combustion are the main concerns). It is however a cheap substrate and its use reduces the risk of contamination and competition from non-sulfate reducing methanogens, which is seen as sufficient justification for its usage (Liamleam & Annachhatre, 2007; Weijma, 2000).

In methanol metabolism, acetate is not a byproduct, instead sulfide, carbon dioxide, methane and other compounds represent the final products (Liamleam & Annachhatre, 2007). An overview of the metabolism, and the connection between the methanol and acetate pathways is found in Figure 1. The transformation of methanol to acetate was crossed out since this reaction is assumed to not occur in the current system. The figure is adapted from Weijma (2000).

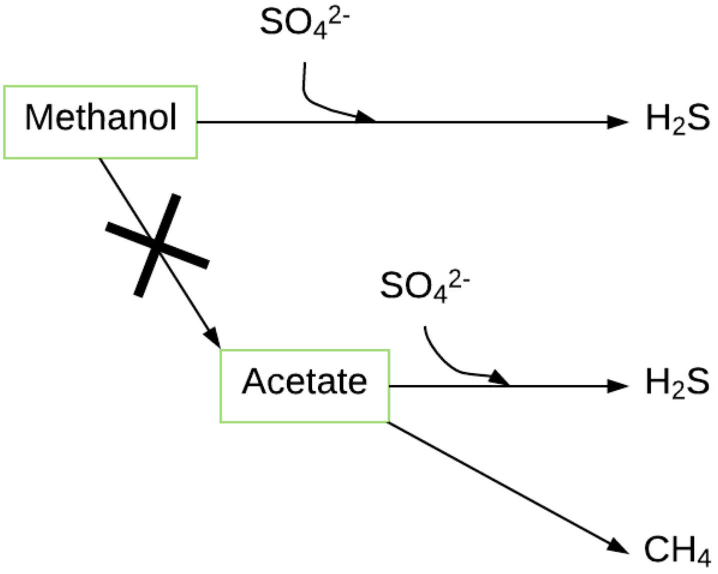


Figure 1 describes the main conversions associated with the substrates methanol and acetate, in SRB. The figure has been adapted from Weijma (2000).

Acetate is however a common intermediate product found in metabolic pathways of other substrates in SRB and is seen as an efficient substrate with respect to the common metabolism of many SRB. The acetate metabolism has no intermediates, just as the methanol metabolism, and is converted to sulfide directly. However, using acetate as substrate results in low growth rates that are comparable with the ones for methanol utilizing SRB.

2.2.2 Biomass formation

Biomass growth in the reactor is simulated to occur according to either of the reactions below, or a combination of both. Both reactions are a combination of ideal theoretical stoichiometric conditions and yields that were derived during the experiments of Nagpal *et al.* (1998) and Moosa *et al.* (2002). These experiments were conducted using ethanol as carbon source, it is assumed that the same biomass yield would be observed when methanol is used as substrate instead (Nagpal, et al., 1998; Moosa, et al., 2002). The resulting growth and production reaction is presented in (1).

$$CH_3OH + 0.71H_2SO_4 + 0.01NH_4 \rightarrow 0.05CH_{1.8}O_{0.5}N_{0.2} + 0.95H_2CO_3 + H_2O + 0.71H_2S \qquad (1)$$

Some of the investigated cases used acetate as carbon source. With acetate, (2) was the resulting growth and production reaction, combining both theoretical stoichiometric terms and the lab results of Nagpal *et al.* (1998).

$$CH_3COOH + H_2SO_4 + 0.04NH_4 \rightarrow 0.195CH_{1.8}O_{0.5}N_{0.2} + 1.805H_2CO_3 + 0.48H_2O + H_2S \qquad (2)$$

For simulation purposes the reactions are balanced on the reactant side by an additional term, H^+, which has no impact on the outcome of the reaction or process. Biomass is defined $CH_{1.8}O_{0.5}N_{0.2}$, since the composition of the SRB consortium is currently unspecified. Further, it is the generic formula for biomass found in SuperPro and a generally accepted formula used to biomass see for example Villadsen, *et al.*, (pg. 105, 2011).

2.2.3 Environmental growth requirements

Zink and copper are both essential trace elements but become toxic to SRB when the levels rise above those found in their natural growth environment (Barton & Fauque, 2009; Doble & Kumar, 2005). Lab scale experiments done by for example Hu *et al.* (2016) showed that when using sodium acetate as carbon source and a concentration of 40 mg Cu/L, a severe decrease in SO_4^{2-} removal was observed compared to experiments where no copper was added. The results indicated approximately a total of 1 mg/mL reduction in sulfate removal when copper was present compared to a final reduction of 375 mg/L (Hu, et al., 2016). With these results in mind, an important factor in the process is to keep the metal ions from entering the bioreactor and thus reducing the possible sulfate reduction.

As previously discussed, there are several different substrate options for the cultivation of SRB. According to Liamleam *et al.* (2007) amongst others, the theoretical requirement for sulfate reduction by SRB is 0.67 g chemical oxygen demand (COD) or electron donors (substrate) per g sulfate. However, the actual COD demand depend on several different factors such as what carbon source is used. For example, methanol have a theoretical COD of 1.5 g/g whereas acetate have a theoretical COD of 1.07 g/g (Iowa State University, n.d.).

See the table of "Abbreviations and commonly used expressions" on pg. 5 for a definition of the meaning of COD for the purpose of this project.

2.2.4 Growth requirements of SRB

SRB are present in many ecosystems due to their acceptance of a wide range of growth conditions. For example, their pH tolerance range from approximately 2 to 10, with an optimum pH range of pH 5-9 (Muyzer & Stams, 2008; Bai, et al., 2013). Acid mine drainage commonly has a pH of 2-4, which would require an increase of pH to attract more species of SRB (Bai, et al., 2013). Additionally, there are also many SRB that are thermostable. Due to their thermostable nature, most SRB exhibit their highest sulfate reduction at higher temperatures such as 65°C (Weijma, 2000).

A few SRB rich environments are mentioned in the following paragraphs, to exemplify the origin of the consortium used in the bioreactor. The common denominator for SRB is that they can be found in anoxic environments such as marine sediments, hydrocarbon seeps and wetlands. SRB's are also present at sites with acid mine drainage, oil fields and anaerobic wastewater treatment plants (Muyzer & Stams, 2008).

The sites at which SRB are present are usually also inhabited by other anaerobic microorganisms such as methanogens, homoacetogens and acetogenic bacteria which all require sulfate. The presence of methanogens causes the SRB to compete for carbon sources (mainly acetic acids), hydrogen and sulfur. If allowed enough time, SRB will outcompete methanogens in an environment containing acetate - however, this process may take years. Thus, to avoid competition and improve process characteristics acetate and other acetic acids should be avoided as the sole carbon source (Muyzer & Stams, 2008).

2.3 The process

The process aims at reducing the incoming level of sulfate from acid mine drainage while simultaneously recovering important metals which are found as metal ions (Cu^{2+}, Zn^{2+}, Me^{2+}) after the leaching process. For the purpose of the simulations, any figures or tables associated with the theory, method or result and discussion chapters the variable Me^{2+} represents a collection of various, unspecified metal ions, unless otherwise specified.

The wastewater treatment process is displayed in the flowchart presented below, see Figure 2, and can be divided into three main sections. Upstream of the bioreactor, the bioreactor and downstream processing. The upstream processing is represented by three metal precipitation steps, each followed by a clarification step. Tokuda *et al.* (2008), reported that the copper, zinc, nickel and tin containing wastewater from electroplating industries, when paired with sulfidation (using H_2S) resulted in metal sulfides. Further, they examined the possibility of selective metal precipitation using pH control for example that copper precipitated at pH 1.5, zinc at pH 4.5 and nickel at pH 5.5-6.0. Tokuda *et al.* showed that precipitation of the aforementioned metals were higher when H_2S was used for the sulfidation step, compared to when Na_2S was used (Tokuda, et al., 2008).

The bioreactor present in the real life process is a so called up-flow anaerobic sludge blanket reactor (UASB) but is simulated as a fermenter. The bioreactor, where sulfate is reduced to hydrogen sulfide by SRB (Hao, et al., 2014). The Matlab model of the bioreactor is based on findings of Barrera *et al.* (2015) as well as the anaerobic digester model 1 (ADM1) created by Batstone *et al.* (2002). The downstream processes are responsible for the final purification of the effluent water from the bioreactor. The final treatment includes filtration to remove any biomass and aeration to remove dissolved any hydrogen sulfide that is present in the system. Finally, an ultrafiltration unit is present to remove the elementary sulfur which is produced in the previous step (McVay, n.d.). In Figure 2, the black lines represent gas flow, the blue lines represent liquid flow and the orange ones represent product flow.

The process is operated in continuous mode to simulate the continuous inflow of leachate from the industrial site associated with the treatment plant. The process simulation will not take the piping dimensions, pressure drops or any other associates issues.

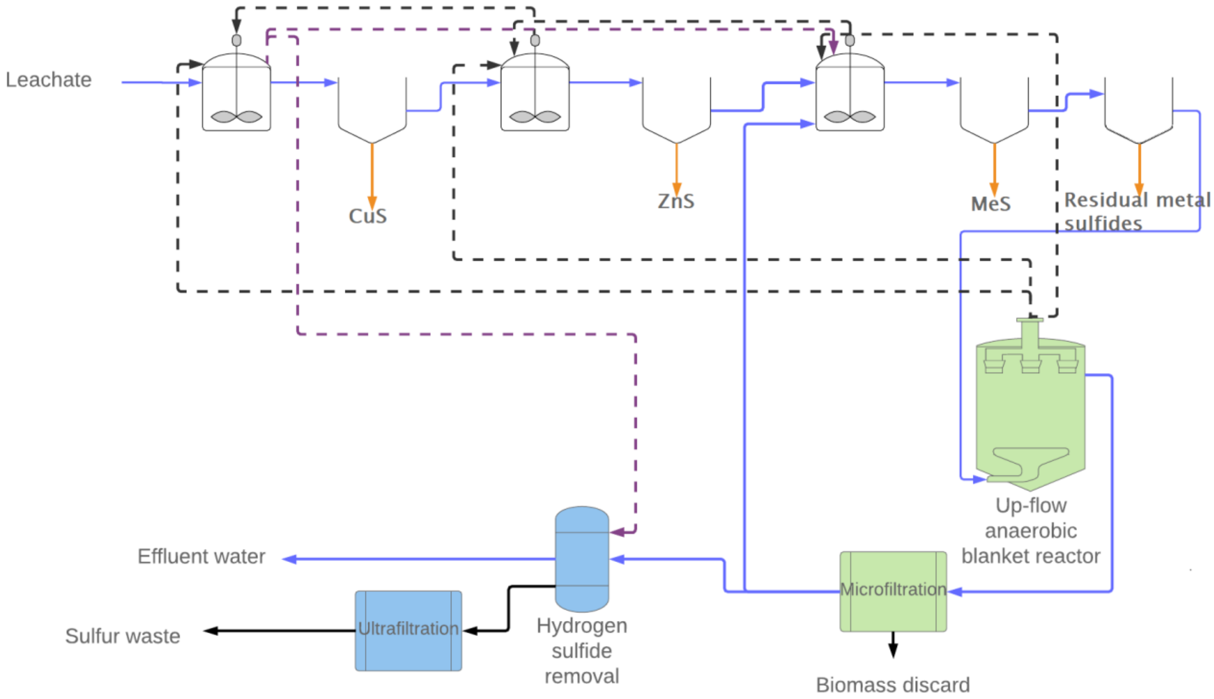


Figure 2 presents a flowchart of the sulfate reducing process. The initial steps of the process are precipitation reactions occurring in continuously stirred tanks, followed by separation of the formed metal sulfides using clarification. The process water is then allowed to enter into the UASB reactor, where the sulfate is transformed to hydrogen sulfide. The hydrogen sulfide is led from the reactor to each of the precipitation tanks whereas the water is partially led back to dilute the incoming sulfate stream to the reactor or to downstream processing. In the downstream processing steps, remaining hydrogen sulfide is removed from the process using aeration.

2.3.1 Metallic leachate

The inlet stream to the process, contains a mixture of metal ions, hydronium ions, sulfate ions and water. This stream originates from the mining process and will be referred to as “inlet stream” or “leachate”. Thus, in the case of this project, the stream will contain copper ions (Cu^{2+}) and zinc ions (Zn^{2+}) as well as various other metal ions (Me^{2+}) depending on the composition of the mineral ore found in the deposit.

2.3.2 UASB reactor

The bioreactor used in the process is an up-flow anaerobic sludge blanket (UASB) reactor. The reactor converts organic material under anaerobic conditions and is commonly used for wastewater treatment with biogas being the desired product. This reactor type can be used with SRB to produce H_2S (g) instead of biogas from process wastewater.

The reactor has an influx of wastewater through channels and holes at the bottom, a simple schematic over an example reactor design can be seen in Figure 3. The influx is relatively high which results in continuous mixing of the reactor. Further, the reactor contains granules of support material (carriers or carrier material) onto which the inoculated biomass attaches and forms a biofilm as the SRB continuously proliferates. As the SRB reduce sulfate, the generated H_2S (g) rises to the top of the reactor, where it is lead out and further engaged in the process. Simultaneously, as the water progresses upwards through the reactor it is continuously purified. The water found at the top of the reactor is seen as “clean” (containing <150 mg/L SO_4^{2-} according to project definition) and is lead out of the reactor (The International Water Association, 2018).

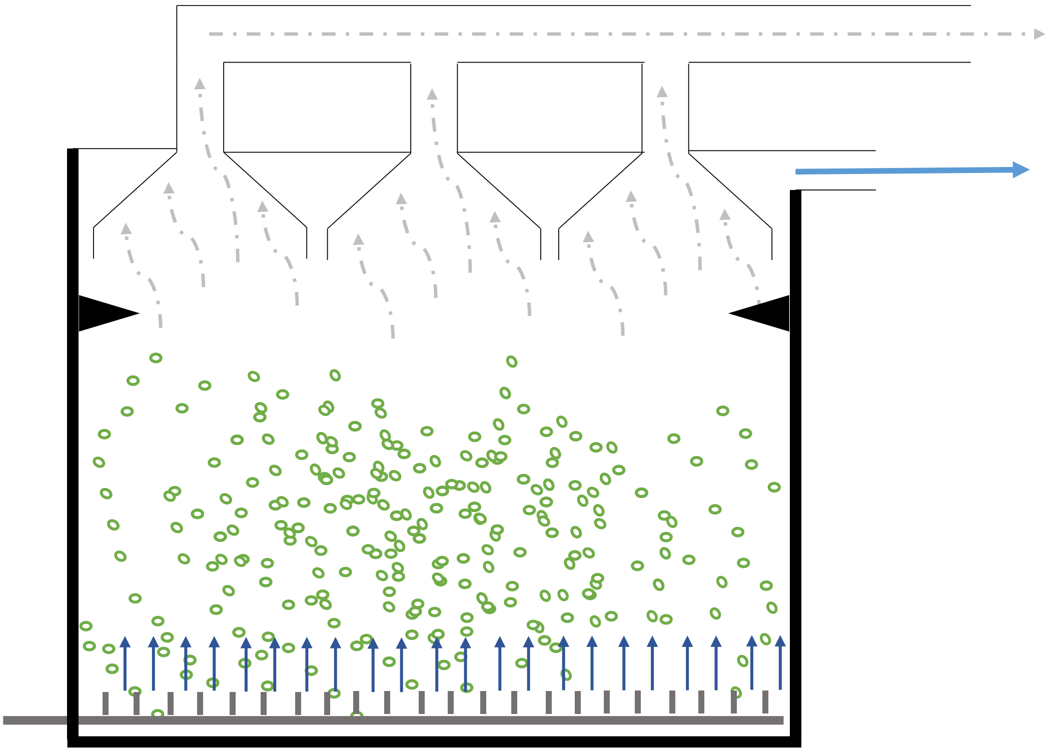


Figure 3 describes the processes occurring in an up-flow anaerobic sludge blanket reactor. The black lines represents the contours of the reactor vessel. The grey fixture at the bottom of the reactor, is the inlet pipes for the process water. The blue arrows at the bottom represents the inflow and rise of water. The green circles represents the SRB containing carrier material. The dotted lines being led out of the reactor through the separators, represents the formed mixture of hydrogen sulfide, carbon dioxide and other gaseous compounds. The blue line exiting the reactor, top right, represents the treated effluent water. The figure was adapted from figures found at The International Water Association and Sustainable Sanitation and Water Management (Eawag (Swiss Federal Institute of Aquatic Science and Technology); Dorothee Spuhler (seecon international gmbh); Sustainable Sanitation and Water Management Toolbox , 2018; The International Water Association Publishing, 2019).

The UASB reactor enables the reduction of sulfate into hydrogen sulfide. The growth rate, as well as the production rates, are expected to follow Monod kinetics. Further, the rate expressions are based on the adapted ADM1 (anaerobic digestion model no. 1) created by Jeppson and Rosén (2011). These expressions are used in creating the Matlab model of the process and can be found in the section “Method for modelling in Matlab”. However, the fermenter that is present in the SuperPro simulation, will be simulated as a stoichiometric fermenter (see Assumptions under Method - SuperPro).

2.3.2.1 Biofilm formation

The UASB reactor uses biofilm on carrier material as the microbial source. A biofilm is a sheet of cells that have attached to a surface (Donlan, 2002). The biofilm is produced by SRB that are capable of biofilm formation and which are introduced to the reactor through a pulse addition at the start of the process. The biomass will continue to grow until all of the carriers are covered. Throughout the continuous biomass growth, any additional biofilm that has been produced will not be able to attach to the carrier material which causes a bleed off of biomass and a natural regulation of its concentration. It is important that bleed off from the carrier material is lower than the generation time of the biofilm culture otherwise washout will occur (D’Acunto, et al., 2011).

As previously mentioned, there are two main types of bacteria that are able to survive under the stated process conditions - methanogens and SRB. Thus, there is possible competition in the bioreactor. The basis of the UASB reactor is the conditions that are provided are specified so that the SRB outcompetes for example methanogens. Therefore, for large scale processes, it is very important to check that the consortium used for inoculation is completely free of contaminants as the reactor is expected to reach a 100% performance level instantly after inoculation.

2.3.3 Precipitation of metal sulfides

The H_2S (g) that is formed in the UASB reactor, is led to precipitation vessels with the goal of producing insoluble metal - sulfide complexes. When dissolved in water, H_2S (g) forms a weak acidic solution according to (3) and (4).

$$H_2S(aq) + H_2O(l) \rightarrow HS^-(l) + H_3O^+(l) \qquad \text{pKa} = 7 \qquad (3)$$
$$HS^-(l) + H_2O(l) \rightarrow S^{2-}(l) + H_3O^+(l) \qquad \text{pKa} = 14.1 \qquad (4)$$

(National Center for Biotechnology Information, 2018)

Bivalent metal ions such as Cu^{2+} and Zn^{2+} reacts with the completely reduced sulfur, S^{2-} and forms an insoluble complex (Tokuda, et al., 2008). In each of the precipitation steps, the (5) will take place.

$$S^{2-} + Me^{2+} \rightarrow MeS \qquad (5)$$

Due to the high pKa associated with the formation of S^{2-} in an aqueous solution and the low solubility product (log(Ksp) =-34.8 for CuS and $-11.5 \leq \log(K_{sp}) \leq -8.7$ for ZnS), it is very unlikely to find S^{2-} ions present in the precipitation tanks. The reaction taking place is most likely (5) because of the pH in the precipitation vessels. However, there is also a possibility of (6) occurring due to the pH in the solution.

$$Me^{2+} + HS^- \rightarrow MeS + 2H^+ \qquad (6)$$

Tokuda *et al.* (2008), approximated the selectivity to 95.5% for Cu at pH 1.5 and 87.4% for Zn at pH 4.5 when the flow rate of H_2S (g) was set to 350 mL/min and its concentration 5000 ppm. The starting metal ion concentrations were 100 mg/L each of Cu and Zn (Tokuda, et al., 2008). The precipitation rate is assumed to be instantaneous and irreversible for this project. However, Al-Tazari *et al.* (2003) showed that the precipitation rate of CuS is directly correlated to the mass transfer rate of H_2S from gaseous phase into the reaction zone. They further showed that the crystal size was increasing with the concentration of H_2S entered into the reactor but was decreasing with both the specific surface area and the liquid side mass transfer rate (Al-Tarazi, et al., 2003).

2.3.4 Particle separation

The leachate is expected to contain other metal ions than Cu^{2+} and Zn^{2+} such as Ni^{2+}, Sn^{2+} and As^{3+}. These ions both disturb the selective precipitation of Cu^{2+} and Zn^{2+} and are toxic to SRB specifically - therefore they should be prevented from entering the bioreactor. Tokuda *et al.* (2008) found that Sn^{2+} co-precipitates with Cu^{2+} at pH 1.5 and that Ni^{2+} precipitates with a 94.7% selectivity at pH 6.7 (Tokuda, et al., 2008).

To prevent the residual metal ions from entering the bioreactor, an additional precipitation tank is required. The pH is raised to approximately pH 7 which should result in metal sulfide precipitation of the residual metal ions.

Separation of the precipitate from the bulk liquid can be achieved by several different techniques relying, on the difference in density of the precipitate in relation to the other dissolved particles and solutes. One technique that can be applied is continuous sedimentation in a clarifying tank. This process allows heavier particles to settle at the bottom of the clarification tank (Suez Water Technologies & Solutions, 2018; Diehl, 2001; Wouda, et al., 1977; Oregon State Government, n.d.).

Separation steps is added after each precipitation tank, to remove the solid particles (crystals) that have been formed. Crystallization is a process that occurs when an outside force is applied which forces the separate particles to form crystal structures. The outside force, is known as supersaturation of a liquid. In the final steps of crystallization, the crystals form larger structures which in turn can be separated by means of density as well as size. For cost efficiency, an industrial sedimentation technique called clarification is used. Clarification is commonly applied within the field of wastewater treatment (Al-Tarazi, et al., 2003; Homostomsky & Jones, 1995).

Clarification consists of two main steps - flocculation and sedimentation. During flocculation, a flocculation agent is added at the inlet and is fully dispersed in the inlet stream due to rapid mixing. The flocculation agent recommended for this process is polymeric and ideally resulting in a low ratio of polymer to metal precipitate as well as easy release of the metal precipitate from the polymer after sedimentation.

The floc aggregates are only able to grow to a certain size, which is determined by several different parameters such as the coagulant dose and type, shear rate and time, temperature and pH (Lindquist, 2003).
It is necessary that the floc achieves a greater density than the density of water, otherwise no sedimentation will occur. Thus, the floc density affects the settling velocity which needs to be greater than the overflow rate if the floc are going to settle (Hammer & Hammer, 2011).

This project uses so called lamella or IP (inclined plate) clarifiers. A lamella clarifier maximizes the sedimentation area while increasing its space efficiency, covering approximately 10% of the area of a traditional clarifier (Nordic Water, 2018). The basic principle of lamella clarifiers is that inclined plates, or lamellas, are stacked closely together in the tank. Water enters the system from a point lower than the top of the lamellas and is forced up along the lamellas. As the water rises up the lamellas, particles sediments and flows in the opposite direction into a collection point - where the sediment can be collected either continuously or in a semi continuous manner. The clarified water is led out at the top of the clarifier (Suez , 2018).

2.3.5 Purification of effluent H_2S/CO_2 stream by leading it through the precipitation tanks

Both H_2S and CO_2 exists in an equilibrium with one liquid and one dissolved gas component as well as in an acid/base liquid equilibrium with the conjugating aqueous base(s) of each species. Both the gas/liquid and the acid/base equilibrium depend on pH as well as temperature. Since the temperature and pressure is assumed to be kept constant over the process (25°C), the equilibria is assumed to only vary with the pH.

The amount of gas that is dissolved in the liquid can be described by Henry’s law, see Equation 1.

Equation 1 - Henrys Law.

$$k_H = \frac{p_g}{Y_g}$$

Where k_H - Henry's constant (temperature dependent), p_g - partial pressure of the gas and Y_g - mole fraction of the gas found in the liquid phase.

The partial gas pressure can be found using the ideal gas law, see Equation 2.

Equation 2 - Ideal gas law.

$$p_g = \frac{nRT}{V} = cRT$$

Where n - moles of gas, R - the ideal gas constant, T - the temperature and c - concentration of the gas.

The concentration of acid and its conjugate base is determined by the acid base equilibrium which is connected to the pH of the solution. Equations 3 and 4 show a general formula for deriving the acid dissociation constant and concentrations.

Equation 3

$$K_a = \frac{[A^-][H^+]}{[HA]}$$

Equation 4

$$pK_a = -\log K_a$$

In the processes, there is a stream that leads recycled water and gas phase components from the bioreactor, into the third precipitation tank. This stream is meant to act as pH regulating in the third precipitation tank and is expected to control the pH to approximately pH 7. As the pH is approximately 7 in the third precipitation tank, CO_2 (g) is expected to dissolve itself in a greater extent in the water. Thus, the H_2S (g) fraction will increase in the gas mixture leaving the precipitation tank – meaning that the residual gas that goes into the second precipitation tank, will contain a lower fraction CO_2 (g) compared to H_2S (g).

2.3.6 An alternative way of gas separation - H_2S/CO_2 gas separation

As sulfate is removed from the process water in the bioreactor, H_2S (g) and simultaneously CO_2 is formed. The gas mixture is led out of the reactor and then into each of the precipitation tanks where it's mixed with the leachate and where S^{2-} and HS^- reacts with for example Cu^{2+} or Zn^{2+} to form metal sulfides, according to (5) and (6).

H_2S (g) is the only gaseous species of interest to this process. For optimization purposes, a purification step where CO_2 is removed could be added. Carbon dioxide in an undesired component in the process both as it reduces the purity of H_2S (g) in the gas mixture but also as its overall pKa is approximately 6.35 indicating that additional pH control will be necessary in precipitation tanks needing lower pH. However, commonly it is CO_2 that is recovered and H_2S is removed - which implicates increased costs for the less common recovery of H_2S (g). Carbon dioxide recovery is achieved through either absorbing the H_2S in cold water or by using amine compounds to absorb the gas in a pressure swing adsorption process, coupled with molecular sieves (Mamrosh, et al., n.d.; Ibrahim & Raj, 2016; Heck, et al., 2018).

Purification of H_2S (g) can be obtained when the gas is present at relatively low levels. The process, called The Dow process (from the Dow Chemical Company), is a scrubber process that uses a low residency time to reduce the co-absorption of CO_2. However, the separated H_2S will not be in its gaseous state but rather as a salt present in an aqueous solution (National Center for Biotechnology Information, 2018; Kohl & Nielsen, 1997).

2.3.7 Effluent water: filtration and purification

The water exiting the bioreactor is filtered to remove any biomass that has been removed from the biofilm. The filtration method is a semi-continuous microfiltration unit with a cut-off at 0.2 μm, corresponding to the size of the biomass. The removal of microbes from the effluent stream is a crucial process step as it ensures that the recycled water does not introduce microbial growth in a precipitation tank and that no microbes are being emitted from the process (Minnesota Rural Water Association, n.d.).

General goal effluent stream values are set according to the values displayed in Table 1. The values are justified by a ruling made by SVEA HOVRÄTT Mark- och miljööverdomstolen on the levels of sulfate, copper and zinc that are acceptable to be released into the sewers connected to the analysis point (SVEA HOVRÄTT Mark- och miljööverdomstolen, 2016).

Table 1 displays the emission limits for each of the substances sulfate, copper and zinc as a mean value over 30 days. The limits are specific to the analysis point but was used as reference values in this project.

Compound	Mean value (30 days)
Sulfate (SO_4^{2-})	150 mg/L
Copper (Cu^{2+})	0.5 μg/L + bioavailability
Zink (Zn^{2+})	5.5 μg/L + bioavailability

As will be discussed later on, the presence of H_2S and HS^- in the water is undesirable in many aspects concerning both environmental and safety risks. These substance have to be removed from the effluent water prior to it exiting the treatment process. There are several processes that can be applied to remove H_2S and HS^- from water. An important aspect is the magnitude of the water that is processed every day resulting in a demand for a simple and inexpensive yet efficient treatment process.

Removal of H_2S and HS^- can be achieved through a process called aeration or degasification where, in this case, the process water is allowed to react with oxygen. The reaction which takes place is (7).

$$H_2S + 0.5O_2 \rightarrow S_0 + H_2O \tag{7}$$

Where S_0 represents elementary sulfur. As the process is depending on inexpensive solutions, air will be used as the source of oxygen. Even though the reaction scheme above displays a 1:1 ratio of S to O, the real requirement is 1:5 when using air as oxygen makes up approximately 20% of the gas mixture of air. Efficiency of the process can be increased by pressurizing the reaction vessel and thus forcing the gas to interact more with the aqueous solution (McVay, n.d.).

The elementary sulfur that is produced needs to be removed, preferably through a filtration process - either ultra- or nanofiltration can be applied for this purpose. For economic purposes ultrafiltration is recommended as its purchasing and operational costs are slightly lower than those of a reverse osmosis unit (Minnesota Rural Water Association, n.d.).

2.3.8 Recycle streams

Streams can be recycled for any number of reasons. One of them being the increased control over the selectivity of the reactions that are occurring. Another is the opportunity to limit the raw material usage, by reentering the raw material that hasn’t been utilized in the previous reaction step.

Recycling of a stream is defined by material from a stream downstream is being re-entered into a stream upstream from the unit operation. An important factor when considering recycling of a stream in industrial processes, is the recycling ratio. The recycling ratio is found by dividing the recycled stream with the feed stream into which the recycled stream is being added (Price, 2005).

There are several recycling streams present in the SuperPro simulation of the process. As seen in Figure 2, a liquid stream from the bioreactor is reentered into the third precipitation tank for dilution of the sulfate concentration entering the reactor as well as to balance the pH in the tank to approximately pH 7. Further, there is recycling of the gas mixture of hydrogen sulfide and carbon dioxide from precipitation tank 3 to 2, tank 2 to tank 1 and from tank 1 some of the remaining hydrogen sulfide is transferred back to tank 3 whereas a portion is removed from the process and treated. The toxicity of the hydrogen sulfide along with increasing resource efficiency are the main reasons behind streams being recycled in this process.

2.3.9 Scale up

The process is simulated using two different magnitudes of flow as specified in the scope of this report. Scale up of processes are known to be accompanied by so called scale up issues. The main scale up issues associated with this process are: whether or not sufficient mixing will be accomplished in the bioreactor and the precipitation tanks and if the mixing time in the smaller scale studies can effectively be transferred to the larger scale operations.

Another problem associated with the scale up of the process is the gas transfer in the bulk liquid. This concerns both the precipitation tank where H_2S (g) is used for the continuous precipitation of Me^{2+} to MeS and the bioreactor where H_2S is produced inside the cells and travels through the bulk liquid - ending up in either a dissolved form of HS^- or in gaseous phase and removed from the reactor through the vent (Villadsen, et al., 2011).

2.4 Environmental impact and risk assessment

This process relies on a substantial H_2S (g) production, to ensure proper metal recovery and sulfate removal. However, the production of hydrogen sulfide is associated with many risks as H_2S (g) is a very toxic gaseous compound. The toxicity is first and foremost associated with inhalation of the gas and its short term exposure limit is 15 ppm. Therefore, substantial measures have to be taken to ensure that no gas is let out into the working environment around the plant. These measures includes, but are not limited to, having hydrogen sulfide sensors in the facility environment, constant measurement and control over the amount of H_2S (g) produced as well as keeping all unit operations which will use the gas as closed units (AFROX, 2011).

The effluent water from the bioreactor will be split into two streams, where one will enter into the previous precipitation steps and the other is the process effluent water, as seen in Figure 2. The recycling of water as well as hydrogen sulfide is a step to increase the efficiency and reduce the amount of waste that is produced by the process. In the final version of this process, as much as possible should be recycled as well as the production of hydrogen sulfide should be optimized to reduce the amount that needs downstream purification.

The effluent water contains H_2S in liquid and gaseous form as well as HS^- in solution, due to the neutral pH of pH 7. To avoid H_2S entering for example the municipal wastewater treatment system, as mentioned before, a commonly employed technique is aeration. Subjecting the water to air and/or oxygen will result in the formation of elementary sulfur. The solid particles could then be removed through ultrafiltration or by an activated carbon filter (Saha, et al., 2017; Farm Water Supply Branch Alberta Agriculture and Forestry, 2018).

Another risk that is associated with the process is the use of SRB. Because the process contains microorganisms it is important that these are removed and destroyed according to protocol. If removal of the microbes would fail, there is a risk of SRB contamination.

The low pH as well as the H_2S production requires the process equipment to meet a special set of standards as both of these conditions are corrosive on common stainless steel. This applies for both the bioreactor as well as the precipitation tanks.

3. Method & materials

This thesis combines a case study based on a SuperPro model of the process as well as a process simulation in Matlab. The SuperPro model is reflected in both the literature study, presented here as well as under “2. Background” and as specific scenarios and cases presented in Table 5 and under “3. Results”. The majority of the project consisted of experimental work in terms of modelling and simulation of the process models which are presented in the following section.

The literature study was curated by two main search engines: LUBsearch and Google as well as a small number of physical literature references. Google, was mainly used when no previous information was available or when additional general information was required. LUBsearch, was used to scan the scientific field for information - i.e. previous studies regarding sulfate reducing bacteria and metal precipitation using H_2S (g). Key search terms included SRB, UASB, metal sulfide precipitation using H_2S and/or any combination of these and similar entries.

The result of the literature study is presented in the previous chapter “2. Background” as well as assumptions and equations in this chapter.

In general, the so called methodology of abduction was applied. Abduction theory is a combination of the induction and deduction strategies where induction strategy is represented by the finding of support for one's hypothesis and deduction originates in finding connections in the data that has been gathered. Abduction is thus the most commonly applied practice in research methodology as it includes the concept of trial and error (Duc, M. Le, 2011).

This project, even if thoroughly defined in its scope was based on a somewhat stiff simulation software and limited prior knowledge which resulted in various trial and error situations along the project. The methodology of trial and error was only applied when the software did not present functions that were seen as necessary as well in situations when for example the presented unit operation was not able to run with the provided input data.

3.1 Method - behind SuperPro

3.1.1 SuperPro Designer

SuperPro Designer v.10.03 was used as one of two simulation software’s in this project. This software was created with mainly biotechnological and environmental applications in mind, such as fermentation and enzymatic processes as well as wastewater treatment. SuperPro Designer, provides a number of unit operations essential to bioprocessing as well as common process operations such as mixing, splitting and temperature control (Intelligen Inc. , 2018).

The software allows for detailed design specifications to be made for each unit operation used in the process. The user is also prompted to choose the mode of operation of the process - batch or continuous, at the start of the simulation. It is possible to mix the two modes but it requires a high degree of knowledge regarding how continuous unit operations work in batch processes and vice versa. Mixing of the two process modes, may result in difficulties to converge the process due to the demands of the specific unit operation and its process mode, might not be fulfilled. In addition to the design specifications, it is possible for the user to define several other parameters affecting the process such as economics, labor, process associated transports and waste management (costs, hazards etc.). Further, the user does not have to specify the size of unit operations as this is done automatically when applying the software's default settings and is based on the user specified inlet stream flows and time constants (Intelligen Inc. , 2018).

In general, the software and the simulation performed will be referred to as "SuperPro" and "SuperPro simulation" or "SuperPro model", in the remainder of this report.

3.1.2 Process assumptions, conditions & parameters

The assumptions made for this process was divided into two main categories - assumptions made for the process in general and assumptions made specifically for the SuperPro simulation. The assumptions are presented in no particular order, below.

3.1.2.1 General Process Parameters and Assumptions

- T = 25°C throughout the process.
- No buffering effect from ions, such as HCO_3^-.
- All reactions involving metal ions hand hydrogen sulfide were assumed to be instantaneous and irreversible.
- $c_{CuS,in} = c_{ZnS,in} = c_{MeS,in} = 0$ mol/L.
- No need to replace biofilm – the culture was assumed to be lasting indefinitely.
- H_2S and CO_2 production was assumed to occur according to the previously presented stoichiometric relationship.
- Only the reactions of $Me^{2+} + H_2S$ (aq) = $MeS + 2H^+$ and $Me^{2+} + H_2S$ (g) = $MeS + 2H^+$ were assumed to occur. Where H_2S (aq) included all available species of H_2S in an aqueous solution.
- As previously mentioned, Me^{2+} included various other metal ions which were assumed to precipitate a 100% at pH 7.
- The overall assumption was that an increased pH would cause the metal ions being transferred from the previous process step to fully precipitate.
- No need to add trace minerals as they were assumed to be present in the leachate entering the process.

3.1.2.2 SuperPro Process Parameters and Assumptions

- Cu = Cu^{2+}, Zn = Zn^{2+}, Me = Me^{2+} (where Me^{2+} was simulated with the same properties as Zn^{2+}), see Appendix A.1.
- In general, no ions were simulated, only pure compounds and mixtures were used with the exception of NH_4^+ as nitrogen source and H^+ for the purpose of balancing the stoichiometric reactions.
- It was assumed that pH was constant in all tanks and thus no need for additional pH control (not including the acid/base additions made to each tank separately).
- For modeling purposes it was assumed that only SRB would inhabit the bioreactor, that no contamination would occur and that there would be no competition with non-SRB methanogens.

- It was assumed that the same biomass yield would be observed in this process as the one Nagpal *et al.* (1998) found using ethanol or acetate as carbon source.
- The reactor was simulated as a stoichiometric fermenter under the assumption that it would work as a UASB reactor.
- For the purpose of clarification CuS(s), ZnS(s) and MeS(s) were simulated with the particle diameters presented in Table 3, thus excluding any type of crystallization or flocculation for the simulation of the process.

3.1.3 Building the SuperPro model

The model developed in SuperPro was subject to previously stated assumptions, predetermined steps in model development as well as an iterative process as the model is very co-dependent on its different parts.

The bioreactor was primarily based on the Aspen Plus model created by Kvarnström and Lönntoft (2017) as well as a primal SuperPro model of the process made by RISE. The SuperPro model at hand was built in sequence according to the three process development phases mentioned in the beginning of the background: precipitation steps, bioreactor and post treatment of the effluent water from the bioreactor.

To achieve a more accurate representation of the process, it was run as a continuous process. In reality the process is somewhere between continuous and semi-continuous because of the unit operations that are involved.

3.1.3.1 Inlet stream and component definitions

The first step when creating a SuperPro model is the definition of the pure components and mixtures that are present in the process. For this model the following components were defined and unless otherwise stated, they received the properties of their reference components as found in the SuperPro database.

In addition to the specified compounds and stock mixtures, SuperPro automatically calls for oxygen, nitrogen and water to be present as registered components and mixtures. The components and stock mixtures are found in Appendix A.1.

In addition to the definition of the components, the (primary) inlet streams of each of the unit operations have to be defined. The inlet streams to the process are leachate, substrate, NaOH and air. The species and the overall stream composition of the leachate stream can be found in Appendix A.2. The substrate stream contained a varying composition of carbon source (methanol and/or acetate), water, NH_4 and H^+ (for stoichiometric purposes). The inlet air stream, only contained air and was allowed to be adjustable to achieve a 100% conversion in the simulated aeration unit. The NaOH stream contained a solution of 50% w/w NaOH.

3.1.3.2 Metal sulfide precipitation and separation

The development of the precipitation steps and the following separation of the metal sulfides was the second step when developing the process model. These process steps were simulated using two unit operations - a CSTR (continuously stirred tank reactor) and an IP (inclined plate) clarifier. In each of the CSTR vessels reactions (8)-(10), were applied. In this case, Me^{2+} and MeS represents the various species of metal (sulfide). The specie H_2S (aq) is assumed to contain all of the species that are present of H_2S in water, for example H_2S (aq), H_2S (g) and HS^- (aq).

$$Me^{2+} + H_2S(g) \rightarrow MeS + 2H^+ \tag{8}$$

$$Me^{2+} + H_2S(aq) \rightarrow MeS(aq) + 2H^+ \tag{9}$$

$$MeS(aq) \rightarrow MeS(s) \tag{10}$$

The different phase specifications were applied to simulate the natural occurrence of species in different physical states as well as the transfer between these states. The conversion factor for each of the metal precipitates were based on the results of Tokuda *et al.* (2008) and are presented in Table 2. The conversion of the unidentified metal precipitates at pH 7, was set to 100% under the assumption that the high pH would cause all remaining metal ions to precipitate.

Table 2 displays the conversion factors used to define each of the metal precipitation reactions. The precipitation of general metal ions to MeS was set to 100% because of the pH being 7 in the final precipitation tank as well as due to the unknown composition of the species "general metal ions". Data from Tokuda et al. (2008).

Metal precipitate	Conversion factor (%)
CuS	96.9
ZnS	96
MeS	100

The temperature of each of the precipitation tanks was set to 25°C. The retention time of each precipitation tank was set to 10.5 hours to allow for all of the metal ions to be precipitated and simultaneously simulate the potential holdup caused by the slow overflow rate of the IP clarifiers (Kobayashi, et al., 1989).

The IP clarification units did not allow for specification of retention or holdup time but generated a so called settling velocity, based on the density and particle size specifications applied during the registration of pure components and stock mixtures. These parameters are presented in Table 3, where the properties of the MeS is an approximation made to account for the size distribution that applies to the various metal sulfides that "MeS" is thought to include.

Table 3 describes the various particle densities and diameters used to appropriately size the clarifiers and account for the sedimentation processes (Singh & Singh, 2012; Ajibade & L.Botha, 2016; American Elements, 2018; National Center for Biotechnology Information, 2019).

Metal precipitate	Particle density (g/cm^3)	Particle diameter (µm)
CuS	4.76	0.0173
ZnS	4.09	0.015
MeS	4.09	0.02

As previously stated, the retention time determined for the precipitation tanks was assumed to include the potential hold up associated with the clarification units. The model was adjusted for 98% removal of the metal sulfide particles in each of the clarification steps (Suez, 2018).

3.1.3.3 The bioreactor

The bioreactor was modeled using the previously developed Matlab model (Kvarnström and Lönntoft, 2017), which contained the reactions and flows of the bioreactor, as guidance. The Matlab model simulated the reactor behavior over the course of one process run, assumed to last 1000 h.

The bioreactor was built using the reactions (11)-(15), with a determined conversion efficiency of 100%.

$$CH_3OH + 0.71H_2SO_4 + 0.01\,NH_4 \rightarrow 0.05CH_{1.8}O_{0.5}N_{0.2} + 0.95H_2CO_3 + H_2O + 0.71H_2S(g) \quad (11)$$

$$CH_3COOH + H_2SO_4 + 0.04NH_4 \rightarrow 0.195CH_{1.8}O_{0.5}N_{0.2} + 1.805H_2CO_3 + 0.48H_2O + H_2S(g) \quad (12)$$

$$CH_3COOH + CH_3OH + 1.71H_2SO_4 + 0.05NH_4 \rightarrow 0.245CH_{1.8}O_{0.5}N_{0.2} + 2.755H_2CO_3 + 1.48H_2O + 1.71H_2S(g) \quad (13)$$

$$H_2CO_3\,(aq) \rightarrow CO_2\,(g) + 2H_2O \quad (14)$$

$$H_2S\,(g) \rightarrow H_2S\,(aq) \quad (15)$$

Reactions (11)-(13), describes the combined reaction for biomass and product formation. Biomass formation was associated with a low yield since the primary motive of the process was product formation and a biomass growth large enough to sustain the biofilms. Reactions (14) and (15) were added to simulate the dissolution of H_2S in the aqueous phase and for the transfer of H_2CO_3 to CO_2 (g). The conversions for reactions (11)-(15) are specified in Table 4 and were found under the assumption of gas-liquid equilibrium being present, by using Henry's Law (Equation 1) in combination with the acid base equilibrium constants (Equation 2).

Table 4 displays the degree of conversion associated with reactions (11)-(14).

Reaction		Conversion (%)
Biomass and product formation	[(11) and (12)]	100
$\boldsymbol{H_2S\,(g) \rightarrow H_2S\,(aq)}$	[(14)]	44 (at pH 7)
$\boldsymbol{H_2CO_3\,(aq) \rightarrow CO_2\,(g) + 2H_2O}$	[(13)]	18 (at pH 7)

The reactor temperature was set to 37°C and no aeration was applied to simulate anaerobic conditions. The hydraulic retention time was set to 10 hours (Kaksonen, et al., 2004). The working volume was defined to be 85% of the reactor volume (Nizami, et al., 2011).

All gas that was produced, was led out from the reactor and into the precipitation vessels, where it was allowed to react with the divalent metal ions.

There are several inhibitory parameters associated with the SRB growth rate equation, as described by Kvarnström and Lönntoft (2017). The full growth equation is displayed in Equation 5, modified to use methanol as substrate, instead of the previously used ethanol. Equation 6, was present in the work of Kvarnström and Lönntoft (2017), and was based on the same kinetics as Equation 5 but used acetate as carbon source. The basis of the equation is Monod growth kinetics and non-competitive terms added to describe inhibition of the growth process.

Equation 5 – Growth equation for growth on methanol as carbon source.

$$\mu = \frac{\mu_{max}}{Y_{SO4/MeOH}} \frac{S_{MeOH}}{K_{m,MeOH} + S_{MeOH}} \frac{S_{SO4}}{K_{m,SO4} + S_{SO4}} X \times I_{H2S} \times I_{pH}$$

Equation 6 - Growth equation with acetate as carbon source.

$$\mu = \frac{\mu_{max}}{Y_{SO4/MeOH}} \frac{S_{Acetate}}{K_{m,Acetate} + S_{Acetate}} \frac{S_{SO4}}{K_{m,SO4} + S_{SO4}} X \times I_{H2S} \times I_{pH}$$

Equation 7 - Hydrogen sulfide coupled inhibition.

$$I_{H2S} = \frac{1}{1 + \frac{S_{H2S}}{K_i}}$$

Where K_i is the inhibitory coefficient/concentration associated with the hydrogen sulfide concentration.

Equation 8 - pH coupled inhibition.

$$I_{pH} = \frac{1 + 2 \times 10^{0.5(pHLL - pHUL)}}{1 + 10^{(pH - pHUL)} + 10^{(pHLL - pH)}}$$

Where pH_{LL} is the lower pH limit and pH_{UL} the upper pH limit to support the growth of the SRB.

The process was simulated using a stoichiometric fermenter, a kinetic fermenter and a kinetic batch fermenter with various degree of adaption of the kinetic expression. The stoichiometric fermenter only used the stoichiometric equations (11)-(13) and no additional kinetic expressions. Additionally, the process was tested with a kinetic fermenter, with both the stoichiometric expressions in (11)-(13) as well as a modified version of Equation 6 where only the hydrogen sulfide coupled inhibition term (Equation 7) was used - as this was the limit of the SuperPro kinetic expression. Finally, a separate flowsheet containing a single batch kinetic reactor was created, where the same reactions were entered as for the previously described kinetic continuous fermenter.

3.1.3.4 Effluent water purification

The final process development step concerned purification of the water leaving the bioreactor. The effluent liquid stream from the reactor was led through a semi-continuous microfiltration unit in which the biomass was separated. The cycle time of the filtration unit was set to 1440 min and a retentate concentration of 1.5 g solids/L.

The permeate was then divided into an effluent stream and a stream that was recycled back into the second precipitation unit. The outgoing water from the bioreactor was used to dilute the ingoing leachate of the third precipitation tank. A similar methodology was used by for example Velasco *et al.* (2008), when the optimal COD/SO_4^{2-} ratio was examined. The recycling stream served as a dilution agent of the SO_4^{2-} entering the bioreactor, as well as a pH control ensuring a buffering effect on the pH to approximately pH 7.

The effluent stream, along with residual H_2S (g), was led into an aeration unit, which was simulated using an equilibrium CSTR. As previously mentioned, the air flow used in the unit was set as adjustable for convenience. The aeration unit aimed at removing HS^- and H_2S (g) and H_2S (aq) from the effluent stream (McVay, n.d.). Reaction (16) was entered in the unit operation.

$$H_2S + 0.5O_2 \rightarrow S_0 + H_2O \tag{16}$$

Where S_0 represents elementary sulfur. As the process requires approximately 5 mg O_2/L when air is used, this was simulated by allowing the air stream to be adjustable. The produced sulfur was then removed using an ultrafiltration unit, as a nanofiltration unit was not available in SuperPro.

3.1.4 Evaluation of the SuperPro model - case trials

To investigate the different outcomes in the SuperPro model, six cases over two primary scenarios were developed. The cases are presented in Table 5 and the scenarios are listed below. Cases 1-3 are based on the theoretical demand of 0.67 g COD/g SO_4^{2-} whereas cases 4-6 are based on an assumption of a higher theoretical demand of 1 g COD/g SO_4^{2-}. The first and forth case of each scenario used methanol as its substrate, the second and fifth used acetate and the third and sixth cases used a mixture of both methanol and acetate.

As previously described for the purpose of simulating the growth and production reactions on methanol and acetate (cases 3 and 6), their respective growth reactions (11) and (12), had to be combined into a single reaction, see reaction (13) below.

$$CH_3COOH + CH_3OH + 1.71H_2SO_4 + 0.05NH_4 \rightarrow 0.245CH_{1.8}O_{0.5}N_{0.2} + 2.755H_2CO_3 + 1.48H_2O + 1.71H_2S \tag{13}$$

The scenarios that were developed and tested in a primary phase are displayed below.

- A scenario with non-optimized carbon source or clarifiers. This case was developed under the assumption that no sulfate would be lost during the precipitation and clarification.
- A scenario with optimized amount of substrate. This case was developed from previous lab results the group at RISE had reported.

Throughout the process it was discovered that the unit operation IP clarifier, impacted the process a lot - especially the throughput of SO_4^{2-}. After the realization that there in fact was loss of sulfate associated with the precipitation and clarification processes, two additional cases were developed.

- A scenario where the clarifiers were optimized (approximately 2% w/w overall loss from the incoming leachate stream) without any optimization regarding the carbon source. The carbon source was based on the theoretical need according to the COD specified in Table 5.
- A scenario with both clarifier and carbon source optimization.

Table 5 displays the optimal COD per case and the associated theoretical COD per substrate.

Case	Optimal COD (g/g)	Carbon source	Theoretical COD per substrate
Case 1	0.67	Methanol	1.5
Case 2	0.67	Acetate	1.07
Case 3	0.67	Methanol and acetate (1:1)	1.5 and 1.07
Case 4	1	Methanol	1.5
Case 5	1	Acetate	1.07
Case 6	1	Methanol and acetate (1:1)	1.5 and 1.07

Table 6 displays a theoretical outcome for the scenario regarding non-optimized clarifiers or carbon source as well as the one with optimized clarifiers.

Table 6 displays the theoretical outcome of the SuperPro simulation, based on scenarios 1 and 2, listed above as well as the cases found in Table 5.

Scenario	Case	Ingoing SO_4^{2-} (mol/h)	Removed SO_4^{2-} (%)	Produced H_2S (mol/h)	Ingoing carbon source (mol/h)	Produced biomass (mol/h)
Non-optimized carbon source or clarifiers	1	19 354	100	19 354	26 725	1 336
	2	19 348	100	19 348	19 982	3 897
	3	19 354	100	19 354	13 363 Methanol 13 363 Acetate	6 548
	4	19 356	100	19 356	39 892	1 995
	5	19 355	100	19 355	29 836	5 818
	6	19 356	100	19 356	19 946 Methanol 19 946 Acetate	9 733
Optimized clarifiers	1	32 321	100	32 321	44 630	2 232
	2	32 331	100	32 331	33 392	6 511
	3	32 230	100	32 230	22 253 Acetate 22 253 Methanol	10 904

3.2 Modelling in Matlab

Previously, a Matlab script describing the bioreactor as a batch process had been developed by Kvarnström and Lönntoft (2017). In this thesis, a section describing a continuous version of the process, some alterations to the bioreactor mass balances and the precipitation reactions were added.

The basis of the bioreactor simulation was the adapted ADM1d model developed by Mu *et al.* (2007), on the basis of the original ADM1 model created by Batstone *et al.* (2002). ADM1d originates from ADM1 which is a kinetic model of the processes occurring in an UASB reactor (Mu, et al., 2007). The model takes both liquid and gas components into account in the kinetic equations which build the model. The ADM1d was further adapted to fit the sulfate reducing process which has been previously described by Kvarnström and Lönntoft (2017).

The additions to the Matlab model followed the same structure as the previously developed bioreactor model by Kvarnström and Lönntoft (2017). That is: one .m file containing the model and differential equations, one .m file containing the parameters used in the model and a final .m file containing the ode (ordinary differential equation) solver.

Equations 5 to 8, had previously been implemented in the Matlab script that described the bioreactor. And was used again, for the same purposes when needed to describe the acid-base and gas-liquid relationships in each of the precipitation tanks.

3.2.1 Definition of Matlab assumptions

Just as the SuperPro model, the Matlab simulation were subject to a set of assumptions as well as many user defined parameters associated with the differential equations and expressions of reaction kinetic. The assumptions are presented in this section and the parameters associated with the models are stated in the following section (3.2.2 Definition of Matlab parameters).

Firstly, the Matlab model was partially based on literature data adapted for the bioreactor script of Kvarnström and Lönntoft (2017). The values associated with constants found in Kvarnström and Lönntofts' (2017) model, were kept intact. Additionally, literature values for the precipitation reactions were adapted in accordance with the ones that Kvarnström and Lönntoft (2017) used when performing their simulation in Aspen Plus.

The constants that were assigned literature values were, as previously discussed, primarily connected to the bioreactor model created by Kvarnström and Lönntoft (2017). However, this model operated under two different conditions compared to the one developed in this thesis. First, the model was developed under the assumption that the process would run as a batch process. Secondly, the earlier model operated in the size of lab scale compared to the present one, which operated in "full" industrial scale (due to the size of the leachate stream). Thus, a major assumption was that these literature values would be completely transferrable from the previous model into the new one. However, as the main purpose of developing this model was to prove that it generated expected results, the literature data can easily be substituted with more accurate process constants as they become available.

Additionally, the model was developed under the assumption that the same yield constants for biomass growth on ethanol and methanol could be used, due to the lack of literature values. As discussed in the previous paragraph, this assumption was made based on the goal for the model being to get it to function. Further, as can be seen in both of the stoichiometric reactions developed, their main goal is not biomass growth but rather production of hydrogen sulfide (as well as carbon dioxide). The biomass that is produced is produced mainly for maintenance of the culture found on the carrier materials.

Constants or values associated with stream sizes and the volume of reaction vessels were results from the SuperPro simulation. It was possible to finally, implement SuperPro values into the Matlab model as the models were developed stepwise after one another which allowed the SuperPro model to be fully finished first.

Additional assumptions that corresponds with the ones made for the SuperPro model are stated in the list below.

- The model was developed under black box assumptions, only considering the bioreactor and precipitation steps of the process. No considerations were taken to the mass and energy balances associated with any of the other unit operations present in the SuperPro model, for example clarification or filtration units.
- The temperature was assumed to be constant in all parts of the process, $T = 25°C = 298$ K.
- The process was assumed to run continuously.
- The bioreactor on which the new model was based, was assumed to contain all of the necessary equations and relationships describing substrate consumption and metabolite production.
- As the terms "Me" and "MeS" were a collection of various species of metal ions, they needed to be assigned generalized parameter values. Since zinc was more abundant in the leachate stream than copper, the terms were assigned the same values as zinc and ZnS wherever it was necessary.

3.2.2 Definition of Matlab parameters

The parameters that were used in the Matlab simulations are stated in Table 7, below. The table contains the constants name in Matlab, the size and unit of the constant as well as the source of the data. The parameters presented in this table, can be found in the attached version of the script as well in Appendix A.4 and Appendix A.5.

Table 7 displays the constants associated with the differential equations, reaction rate equations and other expressions found in the Matlab files. The table contains the variable name, its value and unit, explanation and from where the value was assigned.

Variable	Value/unit	Explanation	Reference
Flow parameters - Overall process			
q_{flow}	123 547 (L/h)	Volumetric leachate flow into the process at the first precipitation tank.	SuperPro results.
q_{s_vol}	1 673.95 (L/h)	Volumetric substrate flow.	SuperPro results.
q_{g_vol}	506 817 (L/h)	Volumetric gas flow out of the bioreactor.	SuperPro results.
q_{in}	54.77 (mol/L)	Overall molar concentration of the stream entering the first precipitation tank.	Based on SuperPro results.
q_s	22.32 (mol/L)	Overall molar concentration of the substrate stream entering the bioreactor.	Based on SuperPro results.
q_p	0.055 (mol/L)	Overall molar concentration of the product stream leaving the precipitation tank.	Based on SuperPro results.
q_g	0.041 (mol/L)	Overall molar concentration of the gas stream leaving the bioreactor.	Based on SuperPro results.

Coefficients for: Methanol degrading SRB: mSRB // Acetate degrading SRB: aSRB			
$\mu_{max,MeOH}$	0.013 (h^{-1}) 0.612 (h^{-1})	Maximum specific growth rate of mSRB (methanol utilizing bacteria).	Nagpal *et al.* (1998) Barrera *et al.* (2002)
$Y_{MeOH/SO4}$	0.0208 (g/g)	Yield for biomass on sulfate, mSRB, Y_{SO4X}	Nagpal *et al.* (1998)
$Y_{XMeOH/SO4}$	0.0104 (g/g)	Yield of mSRB biomass on methanol.	Nagpal *et al.* (1998)
$K_{s,MeOH}$	0.0045 (mol/L)	Saturation constant for methanol (approximated to be equal to that of ethanol).	Nagpal *et al.* (1998)
$K_{s,SO4}$	0.0085 (mol/L) 0.000297 (mol/L)	Saturation constant for sulfate.	Nagpal *et al.* (1998) Barrera *et al.* (2002)
$\mu_{max,Ac}$	0.063 (h^{-1})	Maximum specific growth rate of acetate consuming SRB.	Moosa *et al.* (2002)
$Y_{Ac/SO4}$	0.58 (g/g)	Yield for acetate on biomass, Y_{ACX}.	Moosa *et al.* (2002)
$K_{s,Ac}$	0.0035 (mol/L)	Saturation constant for acetate.	Barrera *et al.* (2002)
Coefficients for: BIOREACTOR INGREDIENT FEEDS/FLOWS			
X_{mSRBin}	1.0954×10^{-6} (mol/L)	Inflow of biomass. A low value (>0) is necessary for the simulation to be able to run.	Based on SuperPro.
X_{aSRBin}	1.0954×10^{-6} (mol/L)	Inflow of biomass. A low value (>0) is necessary for the simulation to be able to run.	Based on SuperPro.
$S_{MeOH,in}$	0.0742 (mol/L)	Inflow concentration of methanol.	Based on SuperPro.
$S_{Ac,in}$	0.0742 (mol/L)	Inflow concentration of acetate.	Based on SuperPro.
$S_{SO4,in}$	0.1269 (mol/L)	Inflow concentration of sulfate.	Based on SuperPro and leachate stream data from RISE partner.
$Y_{XAc/SO4}$	0.0208 (g/g)	Yield for sulfate on biomass, aSRB.	Nagpal *et al.* (1998)
$S_{red,in}$	1.0954×10^{-9} (mol/L)	Reduced sulfur (liquid phase) concentration inflow.	Based on SuperPro.
$S_{IC,in}$	1.0954×10^{-9} (mol/L)	Inorganic carbon concentration inflow.	Based on SuperPro.
$k_{dec,mSRB}$	0 (h^{-1})	Decay coefficient for methanol utilizing SRB.	Moosa *et al.* (2002)
$k_{dec,aSRB}$	0 (h^{-1})	Decay coefficient for acetate utilizing SRB.	Moosa *et al.* (2002)
Mass transfer gas-liquid parameters: BIOREACTOR			
$k_{La,H2S}$	10	Mass transfer coefficient for H_2S.	Nagpal *et al.* (1998)

$k_{La,CO2}$	10	Mass transfer coefficient for CO_2.	Nagpal *et al.* (1998)
$k_{H,H2S}$	10.7	Henry’s constant for H_2S.	Nagpal *et al.* (1998)
$k_{H,CO2}$	25.88	Henry’s constant for CO_2.	Nagpal *et al.* (1998)
T	298 (K)	Temperature.	Based on SuperPro.
R	0.082057	Ideal gas constant.	-
V_{liq}	343 288 (L)	Liquid volume.	SuperPro.
C_{gas}	0.0409 (mol/L)	Gas concentration in outgoing flow from the reactor	Based on SuperPro
P_{tot}	1 (atm)	Total pressure from all gases, corresponding to the external pressure.	-
The bioreactor: Acid-base coefficients			
$k_{ab,H2S}$	1×10^{15}	Acid base kinetic parameter for H_2S.	-
$K_{a,H2S}$	1.075×10^{-7}	Acid-base equilibrium coefficient for H_2S.	-
$K_{a,CO2}$	4.97×10^{-7}	Acid-base equilibrium coefficient for HCO_3^-.	-
$K_{ab,CO2}$	1×10^{12}	Acid base kinetic parameter for HCO_3^-.	-
The bioreactor: Inhibition parameters			
$pH_{LL,mSRB}$	6	Lower pH limit where the group of organisms (mSRB) are 50% inhibited.	
$pH_{UL,mSRB}$	7.5	Higher pH limit where the group of organisms (mSRB) are 50% inhibited.	
$pH_{LL,aSRB}$	6	Lower pH limit where the group of organisms (aSRB) are 50% inhibited.	
$pH_{UL,aSRB}$	7.5	Higher pH limit where the group of organisms (aSRB) are 50% inhibited	
pH_{bio}	7	pH in the bioreactor, for the pH inhibition term.	
$K_{i,H2S}$	5.13×10^{-3}	Inhibition constant for the H_2S inhibition term.	Barrera *et al.* (2002)
	0.003875		Kaksonen *et al.* (2002)
	1.54		Nagpal *et al.* (1998)
Precipitation steps: Acid-base parameters			
$pH_{1.5}$	1.5	pH 1.5 in the first precipitation tank.	Tokuda *et al.* (2008)
$pH_{4.5}$	4.5	pH 4.5 in the second precipitation tank.	Tokuda *et al.* (2008)
$pH_{7.2}$	7.2	pH 7.2 in the third precipitation tank.	Tokuda *et al.* (2008)

Precipitation steps: Precipitation rate constants and conversion yields/coefficients			
k_{Cu}	2.4156 (h^{-1})	Precipitation rate constant for CuS precipitation.	Tokuda *et al.* (2008)
k_{Zn}	2.9448 (h^{-1})	Precipitation rate constant for ZnS precipitation, also used for MeS precipitation.	Tokuda *et al.* (2008)
Y_{CuS}	0.9660	Conversion achieved for the CuS precipitation.	Tokuda *et al.* (2008)
Y_{ZnS}	0.9600	Conversion achieved for the ZnS precipitation.	Tokuda *et al.* (2008)
Y_{MeS}	1	Conversion achieved for the MeS precipitation.	Tokuda *et al.* (2008)
Precipitation steps: Molar flows of Cu^{2+}_{in}, Zn^{2+}_{in} and Me^{2+}_{in} (from RISE partner)			
$S_{Cu,in}$	7.2126×10^{-4} (mol/L)	Ingoing concentration of Cu^{2+}.	RISE partner.
$S_{Zn,in}$	0.0174 (mol/L)	Ingoing concentration of Zn^{2+}.	RISE partner.
$S_{Me,in}$	0.1174 (mol/L)	Ingoing concentration of Me^{2+}.	RISE partner.
Precipitation steps: Tank /unit operation parameters			
$V_{liq,bio}$	1.8198×10^{6} (L)	Total volume of all of the bioreactor units simulated in SuperPro.	SuperPro.
$V_{liq,tank1}$	1.73034×10^{6} (L)	Total volume of all of the first precipitation tank vessels simulated in SuperPro.	SuperPro.
$V_{liq,tank2}$	1.72602×10^{6} (L)	Total volume of all of the second precipitation tank vessels simulated in SuperPro.	SuperPro.
$V_{liq,tank3}$	3.21651×10^{6} (L)	Total volume of all of the third precipitation tank vessels simulated in SuperPro.	SuperPro.
V_{gas}	3.0936×10^{5} (L)	Gas volume out of the bioreactor.	SuperPro.

3.2.3 Flow relationships

As a step in implementing a continuous model, flow expressions were developed to simulate the transfer of the liquid between the tanks. The dynamics were described in volumetric units of L/h. The flow dynamics are presented below in Equation 9 to 17.

Equation 9

$$q_{p1} = Z_1 q_{in}$$

Equation 10

$$q_{p2} = Z_2 q_1$$

Equation 11

$$q_{p3} = Z_3 q_2$$

Where Z_1-Z_3 represents the fraction of reduction of the liquid stream through product separation, a number which was retrieved from the clarifier mass balances found in the SuperPro model.

Equation 12

$$q_1 = q_{in}$$

Equation 13

$$q_2 = q_1 - q_{p1}$$

Equation 14

$$q_{re3} = F_x$$

Equation 15

$$q_3 = q_2 - q_{p2} + q_{re3}$$

Equation 16

$$q_{in,bio} = q_3 - q_{p3} + q_s$$

Equation 17

$$q_{out} = q_{in,bio} - q_{re3}$$

Where q_1-q_3 and $q_{in,b}$ represented inflows into the different tanks and q_{out} the effluent stream from the bioreactor. The variable q_{re3} represented the recycled stream from the bioreactor into the third precipitation tank, this was assigned the constant F_x (F_x = 121 m^3/h = 121 000 L/h) based on results from the SuperPro model. The value that was assigned to this stream was assumed to result in sufficient dilution of the incoming sulfate stream as well as control the pH around pH 7. An additional effluent stream was added to the script to represent the outgoing gas flow from the bioreactor. This was given the term q_g, the unit L/h and was collected from the SuperPro script.

3.2.4 Adapting the bioreactor mass balances

The bioreactor model was adapted to fit the data retrieved from the SuperPro simulation as well as a continuous process mode. The bioreactor model developed by Kvarnström and Lönntoft (2017) was directly based off of the Jeppson and Rosén (2011) adaption of the ADM1 model developed by Batstone *et al.* (2002). The equations associated with the previous model, are stated with the adapted versions following directly below each of them.

In the model developed by Kvarnström and Lönntoft (2017), the biomass formation using the substrates was described according to Equation 18. However, to account for the stoichiometry associated with the coupled sulfate and methanol growth, the yield that Nagpal *et al.* (1998) found was divided with 0.75, resulting in Equation 19. Equation 20 was thus created to describe the specifically coupled sulfate consumption.

Equation 18 - Biomass formation on methanol.

$$r_{MeOH} = \frac{\mu_{max}}{Y_{SO4/MeOH}} \frac{S_{MeOH}}{K_{m,MeOH} + S_{MeOH}} \frac{S_{SO4}}{K_{m,SO4} + S_{SO4}} X \times I_{H2S} \times I_{pH}$$

Equation 19 – Adapted, biomass formation on methanol.

$$r_{MeOH} = \frac{\mu_{max}}{\frac{Y_{X/SO4}}{0.75}} \frac{S_{MeOH}}{K_{m,MeOH} + S_{MeOH}} \frac{S_{SO4}}{K_{m,SO4} + S_{SO4}} X \times I_{H2S} \times I_{pH}$$

Equation 20 - Relationship between sulfate consumption rate and methanol consumption, based on the stoichiometry of biomass production on methanol.

$$r_{SO4} = 0.75 r_{MeOH}$$

The differential equation describing sulfate consumption was adapted to account for the different substrates and their coupled rate equations. The previously used equations is labeled 20 and the adapted version as Equation 21.

Equation 21 - Sulfate reduction.

$$\frac{dS_{SO4}}{dt} = \frac{q_{in,bio}}{V_{liq}} \left(S_{SO4,in} - S_{SO4}\right) - \left(1 - Y_{\frac{X}{MeOH}}\right) Y_{\frac{X}{SO4}} r_{MeOH}$$

Equation 22 - Sulfate reduction, adapted.

$$\frac{dS_{SO4}}{dt} = \frac{q_{in,bio}}{V_{liq}} \left(S_{SO4,in} - S_{SO4}\right) - r_{SO4}$$

Where r_{SO4} contain the stoichiometry associated with the coupled sulfate and methanol consumption.

The differential equation describing the production of hydrogen sulfide was altered to consider the consumption of S_{red} in the metal precipitation equations. The resulting differential equation is presented in Equation 23.

Equation 23 - Production of hydrogen sulfide (reduced sulfur, liquid)

$$\frac{dS_{red}}{dt} = \frac{q_{in,bio}}{V_{liq}} \left(S_{red,in} - S_{red}\right) + \left(1 - Y_{\frac{X}{MeOH}}\right) r_{SO4} - r_{H2St} - \left(1 - Y_{\frac{X}{Acetate}}\right) r_{acetate} - \rho_{Me^{2+}}$$

Where r_{H2S} is the mass transfer rate of H_2S (g) (Equation 33), $Y_{X,MeOH}$ is the yield of biomass on methanol, $Y_{X,Acetate}$ is the biomass yield on acetate and ρ_{Me2+} is the rate of metal sulfide formation (Equation 25).

3.2.5 Modelling of metal precipitation

Finally, the precipitation reactions were described according to the model developed by Tokuda *et al.* (2008) with some modifications to fit the purpose of this simulation, see Equation 24. The metal ions and the reduced sulfide species were assumed to react in a 1:1 ratio, which explains the structure found in Equation 25. To avoid that the script generated any form of negative concentrations, an if-statement which conditioned the Me^{2+} concentration to be equal to or greater than 0.

Equation 24 - Removal of metal ions.

$$\frac{dS_{Me^{2+}}}{dt} = \frac{q}{V_{tank}} \left(S_{Me^{2+}_{in}} - S_{Me^{2+}}\right) - \rho_{Me^{2+}}$$

Where q represents the ingoing stream to that specific precipitation tank (q_1, q_2 or q_3) and P_{Me2+} represent the uptake of Me^{2+} into metal sulfide complexes, see Equation 25.

Equation 25 - Rate of metal sulfide formation.

$$\rho_{Me^{2+}} = k\left(S_{Me^{2+}} + S_{S_{red,tank}}\right)$$

In which S_{Sred} was added to describe the reaction between H_2S (aq) and Me^{2+}, instead of the H_2S (g) present in the gas/liquid interface as described by Tokuda et al (2008).

For each of the precipitation tanks, the same type of acid-base and gas-liquid equilibrium describing equations as were found in the bioreactor model, were added. The equations are presented below in Equation 26 to 35 and for demonstrative purposes Me^{2+} will be used to represent an arbitrary species of metal ions. The following rate equations were based on the equations developed by Batstone *et al.* (2002) for the ADM1 reactor, and adapted to fit the purpose of this process.

Acid-base expressions and equilibrium expressions are stated below in Equation 26 – 32.

Equation 26 – Description of the relation between the concentration of H^+ and the pH in the tank

$$S_{H^+_{tank}} = 10^{-pH_{tank}}$$

Equation 27 – Specific relation describing the availability of H^+ in a specific tank

$$S_{H^+_{in,tank}} = S_{H^+_{tank}} + \frac{S_{H^+_{bio}}}{3}$$

Equation 28 – Description of the availability of the reduced sulfur (liquid), specific to a tank

$$S_{Sred,tank} = \frac{S_{Sred,bio}}{3}$$

Where the term $S_{red,tank}$, refers to the availability of all reduced species of sulfur present in the liquid phase of the specific tank.

Equation 29 – H_2S (g) presence in a specific precipitation tank

$$S_{H_2S,tank} = S_{Sred,tank} - S_{HS^-_{tank}}$$

Equation 30 – CO_2 (g) presence in a specific precipitation tank

$$S_{CO_2,tank} = S_{IC,tank} - S_{HCO3^-_{tank}}$$

Equation 31 – Dissociation equation, HS^-

$$\rho_{A,HS^-_{tank}} = K_{ab}(S_{HS^-_{tank}}\left(S_{H^+_{in,tank}} + K_{a,H_2S}\right) - K_{a,H_2S} \times S_{Sred,tank}$$

Equation 32 – Dissociation equation, HCO_3^-

$$\rho_{A,HCO3^-_{tank}} = K_{ab}(S_{HCO3^-_{tank}}\left(S_{H^+_{in,tank}} + K_{a,CO_2}\right) - K_{a,CO_2} \times S_{IC,tank}$$

Mass transport for H_2S (g) from liquid to gas is described by Equation 33.

Equation 33 – Mass transport of H_2S (g) from liquid to gas

$$r_{H_2St,tank} = k_l a(S_{prim,H_2S} - S_{H_2S,tank})$$

Dissociation rate expressions for each of the precipitation tanks are described in Equation 34 – 35.

Equation 34 – Dissociation rate, HS$^-$

$$\frac{dS_{HS^-_{tank}}}{dt} = -\rho_{A,HS^-_{tank}}$$

Equation 35 – Dissociation rate, HCO$_3^-$

$$\frac{dS_{HCO3^-_{tank}}}{dt} = -\rho_{A,HCO3^-_{tank}}$$

4. Result & Discussion

4.1 SuperPro model and simulation

A SuperPro model was developed for a small, laboratory scale, process and another was created for an industrial sized leachate flow. The sizes were chosen as they corresponded to cases where real process data could be applied. The lab scale process is shown in Figure 4 and the larger in Figure 5. The main difference between the smaller and larger models were that there were no pumps or a compressor simulated in the smaller case - as the flows were too small to size the equipment properly. It is possible to recreate the same scenarios and cases that are presented later on in this section, with the smaller scale model. However, the small case model was not investigated for the different scenarios and cases since the leachate stream had contents that were specified for an industrial scaled process.

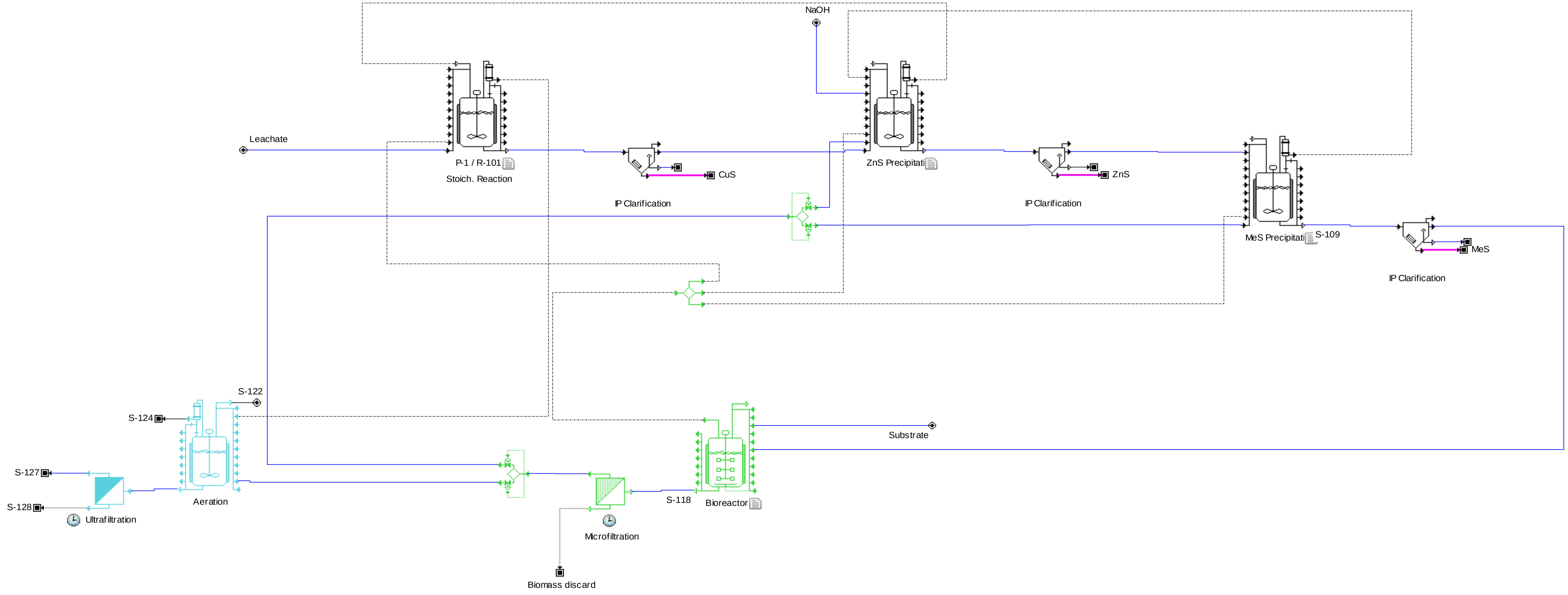


Figure 4 displays a flowsheet over the small scale process. The unit operations in black make up the section "Precipitation", the green unit operations belong to the section “Bioreactor” and the blue unit operations belong to “Effluent treatment”. Liquid flows are displayed as blue lines, gaseous flows are displayed as black dotted lines. Purple, solid lines represent the product flow of various metal sulfides and light grey lines represent waste streams.

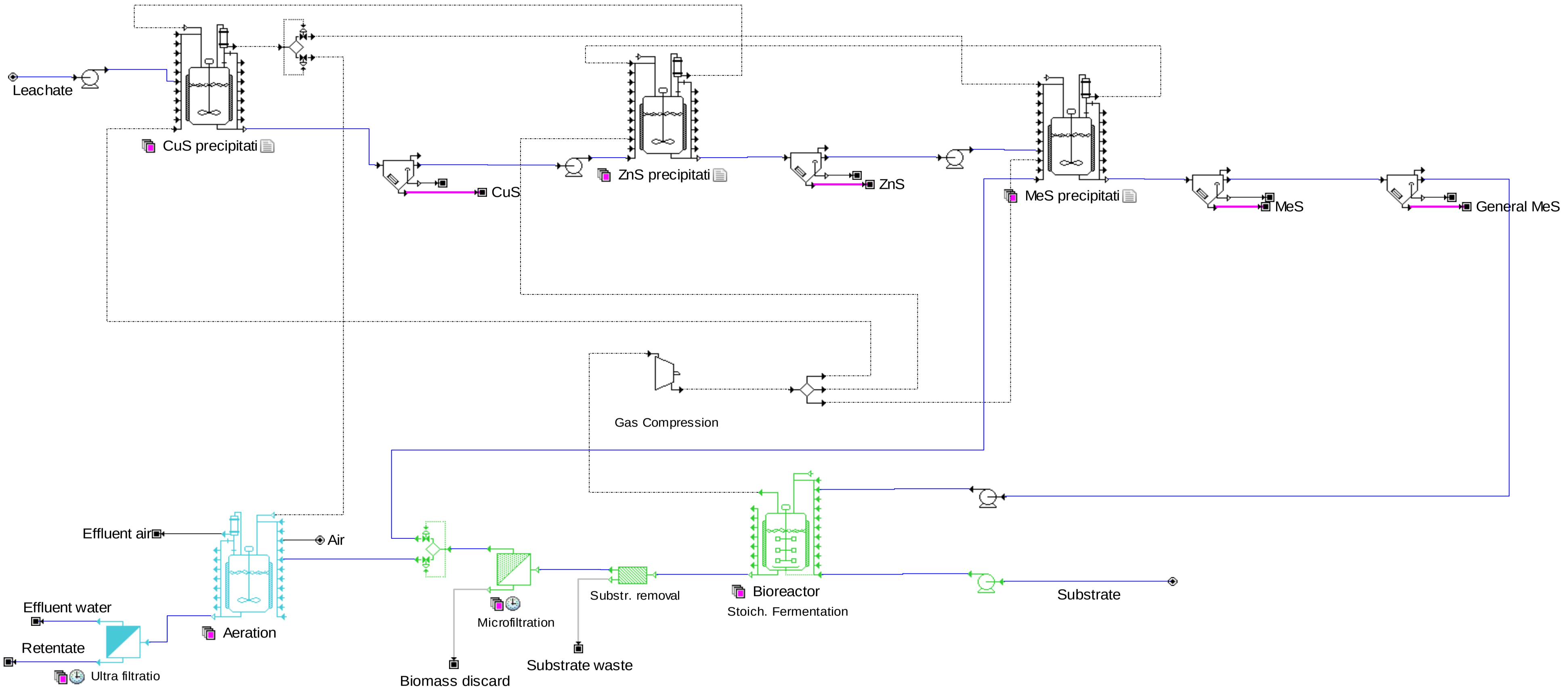


Figure 5 displays the flowsheet created for the larger scale process. The main difference from the small scale process is the addition of pumps as well as a compressor for the gas flow. The black unit operations make up the section "Precipitation", the green unit operations belong to the section “Bioreactor” and the blue unit operations belong to “Effluent treatment”. Liquid flows are displayed as blue lines, gaseous flows are displayed as black dotted lines. Purple lines represent the product flow of various metal sulfides and light grey lines represent waste streams.

The output stream named “Substrate waste”, was included to control the usage of carbon source in the reactor. It was not possible to get the reactor to use all of the carbon source without entering into an error mode. Therefore, it was necessary to keep a slight surplus of the carbon source flow. The surplus of carbon source is thus expected to be used in an actual version of the process where it is assumed to partake in metabolic processes producing a variety of carbon based byproducts, such as CO_2 and some CH_4(g) amongst others. The failure to meet exactly 100% usage of carbon source, might have been due to the condition that 100% of the sulfate should be removed. If another condition of the sulfate reduction would have been applied to the SuperPro model, the substrate usage might have been consistently 100% without any associated errors.

A variety of cases, displayed in Table 5, were examined under two primary scenarios. The first scenario, which was the baseline for all the cases was a case without optimized clarifiers and where theoretical concentrations for the carbon sources was used. The second scenario that was examined, was optimization of the carbon source with the default clarifier settings. As previously mentioned, it was discovered that the outgoing solids containing feed stream from the clarifiers, intuitively, impacted the process severely. A higher degree of solids in the product stream resulted in a higher degree of sulfate being present in the following process steps. Thus, an additional scenario that was examined was based on the belief that the outgoing product flow from the clarifier, should be optimized (approximately 2% w/w overall loss of sulfate up until the bioreactor) so that as much sulfate as possible were kept in the ongoing process stream. The final scenario examined the optimized clarifier settings coupled with an optimized feed of the carbon source feed.

The values associated with the resulting SuperPro simulation are displayed in Table 8. The resulting “Material & Stream” and “Equipment” reports of the simulation using optimized carbon source and default settings of the clarifiers, case 1 are displayed in Appendix A.3 and A.4 respectively. Additionally, Appendix A.3 displays the flowchart related to the “Material & Stream” and “Equipment” reports, which contains the names of all streams and equipment. These reports are included as an example of the results one can generate from a SuperPro simulation. The results are all based on this type of reports, each generated specifically for its scenario and case number.

As seen in Table 8, SuperPro consistently produced too much H_2S to the amount of sulfate available in the process feed stream into the bioreactor. Since there is a consistent overproduction of 0.94% of H_2S, this can be explained by the software’s usage of either too few or too many significant digits in the calculations due to the reaction stoichiometry defined for the process.

Further, as displayed below, the resulting substrate usage across for cases 1, 2 and 5 in scenario 1 are almost 100%, the exception being cases 3, 4 and 6. Worth noting, is that two of the cases with deviating results, in scenario 1 are the ones containing multiple substrates. This could be an indication that SuperPro have some difficulties processing the multiple substrates and the entered stoichiometric reaction (13). However, as seen from the results of the remaining scenarios, the substrate usage improved as the clarifier settings and amount of substrate were optimized.

Table 8 displays the results from the SuperPro simulation for all different scenarios.

Scenario	Case	Used substrate (%)	Produced biomass (% of expected)	Removed SO_4^{2-} (%)	Produced H_2S (% of expected)
Scenario 1 (non-optimized)	1	100	99.9	100	98
	2	97.8	100	100	101
	3	85.5	50	100	101
	4	68.8	100	100	101
	5	65.5	100	100	101
	6	57.3	50	100	101
Scenario 2 (optimized carbon source, non-optimized clarifiers)	1	100	100	100	101
	2	100	100	100	101
	3	100	50	100	101
Scenario 3* (optimized clarifiers)	1	100	99.9	97.4	98.3
	2	97.8	100	100	101
	3	85.5	50	100	101
Scenario 4* (optimized clarifiers + optimized carbon source)	1	100	99.9	100	101
	2	100	100	100	101
	3	100	50	100	101

*The optimized C-source results applies for example to both case 1 and case 4, as they are based on the same principles and reactions. Therefore, only one results for cases 1-3 are displayed for these scenarios.

Figure 6, displays the substrate usage by the bioreactor for each of the cases across the four scenarios. According to the figure, the substrate usage corresponds well to the theoretically predicted one, see Table 6. The deviating cases in scenarios 1 and 3, were case 3 and 6, which contained multiple substrates. This indicates that the method for predicting the substrate need, based on COD/SO_4^{2-} ratio and multiple substrate was incorrect. However, due to the ability to reach near 100% substrate usage in scenarios 2 and 4 (both optimizing the carbon source), indicated that SuperPro is able to differentiate properly between the substrates.

The resulting COD/SO_4^{2-} ratios for each of the scenarios are found in Table 9, below. The result in Table 9, is displayed for case 1-3, as the overall result suggests that SuperPro generated solutions that are in close proximity to the predetermined COD/SO_4^{2-} ratio. However, with the results presented here there is no need to model the process using a COD/SO_4^{2-} ratio of 1 g/g as the bioreactor only is based on stoichiometry, with a predetermined 100% conversion efficiency for the combined growth and product reaction.

Table 9 displays the resulting COD/SO_4^{2-} ratios for the different cases in the scenario of optimized carbon source usage and default clarifier settings.

Carbon source (theoretical g COD/g SO_4^{2-})	**COD/SO_4^{2-} final ratio (g/g)** **Scenario 1**	**Scenario 2**	**Scenario 3**	**Scenario 4**
Metanol (0.67 g COD/g SO_4^{2-})	0.67	0.68	0.67	0.69
Acetate (0.67 g COD/g SO_4^{2-})	0.67	0.66	0.67	0.66
Metanol + Acetate (1:1) (0.67 g COD/g SO_4^{2-})	0.78	0.67	0.78	0.67
Metanol (1 g COD/g SO_4^{2-})	1.00	-	-	-
Acetate (1 g COD/g SO_4^{2-})	1.00	-	-	-
Metanol + Acetate (1:1) (1 g COD/g SO_4^{2-})	1.18	-	-	-

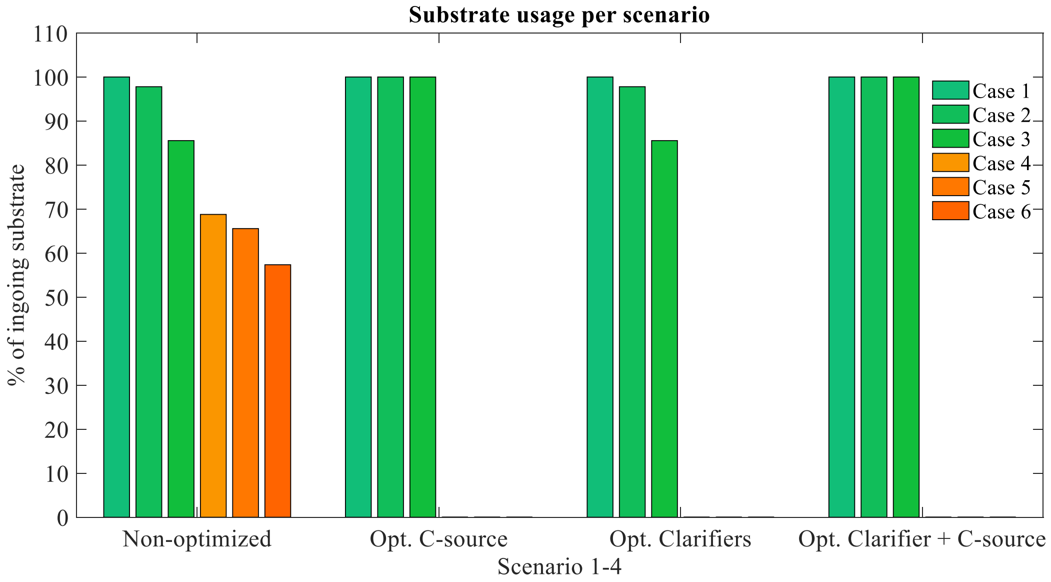


Figure 6 displays the substrate usage for each case in every scenario. Case 1 achieved almost 100% substrate usage across all scenarios, the same is true for case 2 excluding scenario 3. Case 3, reaches approximately 100% substrate usage in scenario 2 and 4 but is approximately only 80% for scenarios 1 and 3. Case 4 through 6, were only tested for the non-optimized conditions as their results would be the same in case 1 through 3.

Figure 7, displays the total production of H_2S for case 1 across the four scenarios. Case 1, was chosen specifically since methanol is the preferred carbon source. It reduces the risk of competition between SRB and non-SRB methanogens. As previously discussed, another reason was because of the overall highest substrate usage across the scenarios was associated with methanol as substrate. This raises the question if the model is better adapted to methanol because the growth equation was based on results from experiments using ethanol. Another approach would be to discuss whether or not these results simply depend on the growth/product reaction for methanol is better stoichometrically balanced compared to the one for acetate.

The production of H_2S depends on two main parameters: the ingoing sulfate concentration and the ingoing substrate concentration. The larger the amount of sulfate that enters the bioreactor, the higher amount of H_2S is produced, provided that the increased sulfate concentration is matched with a proportionally increased amount of carbon source. This trend is clearly presented in Figure 7. The scenarios 1 and 2 contained a lower sulfate inflow of sulfate to the bioreactor compared to scenarios 3 and 4 where a higher amount of sulfate entered the bioreactor due to optimization of the clarifiers. Thus, scenarios 1 and 2 were associated with lower production of H_2S.

Figure 7, clearly illustrates that it is vital to have good knowledge of all components, unit operations and flows when creating a model in SuperPro. As seen in Figure 8, the production of H_2S increases across the scenarios however, the amount of utilized H_2S remains the same as expected. Since too little is known about the full scale process, it is difficult to determine which of these four scenarios that corresponds to reality best. However, it is most likely that an approach of optimizing the amount of carbon source will be used – as this is what is currently applied in the lab trials.

Figure 7, displays that there is a higher fraction of unused H_2S associated with the scenarios 3 and 4, compared to 1 and 2. However, the amount of used H_2S is the same across all of the cases, approximately 20 000 mol. This result raises the question of what is more desirable. It seems as if though scenarios 1 and 2 are better optimized against the process, as less H_2S is unused and thus there is less need for treatment of the residual hydrogen sulfide. On the other hand, the excess gas produced in scenario 3 and 4 could be stored and used to top-up the produced H_2S if there was be a lower amount of sulfate in the leachate. Alterntively, the unused gas in scenario 3 and 4 could be stored and sold.

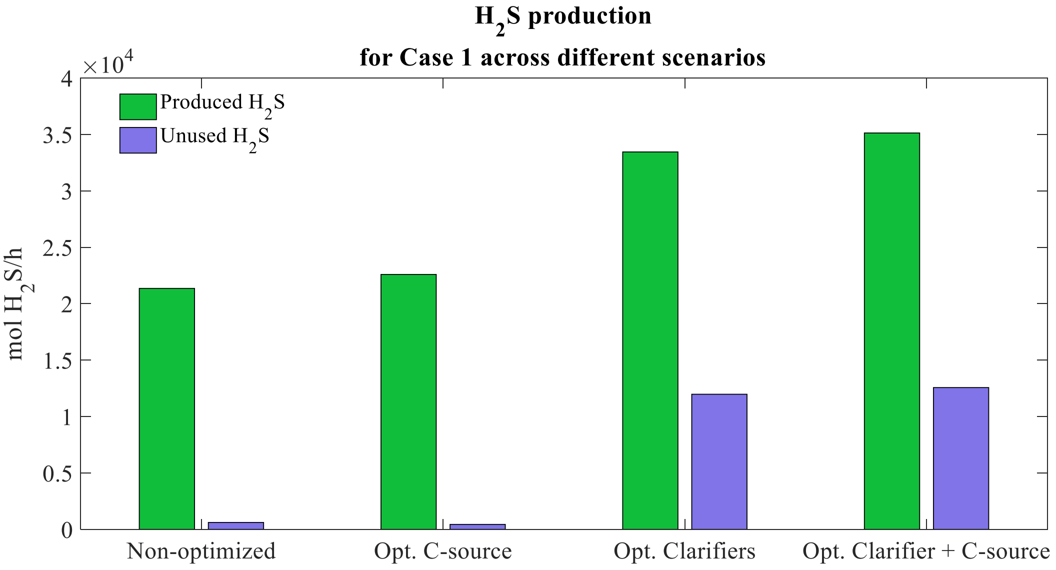


Figure 7 displays the total hydrogen sulfide production and usage for case 1 (methanol) in each of the four scenarios.

Figure 8, displays the amount of each metal sulfide that was recovered from each clarifier. As seen in Figure 8, the first separation is expected to contain the overall purest sample of metal sulfide (CuS) whereas the following separation steps will be associated with having an increasing amount of impurities. A 100% overall recovery of both CuS and ZnS is expected whereas the general MeS is expected to have a lower overall recovery as it only passes through two clarifiers compared to four for CuS and ZnS.

For the unknown, various MeS that are removed in the last two clarifiers, the recovery is approximately 99.96 % which results in some residual MeS that will enter the bioreactor. Presumably, this should not affect the performance of the bioreactor as the MeS is an irreversibly formed compound. However, due to the constant inflow of the relatively heavy MeS, there might be sedimentation at the bottom of the bioreactor or on the carrier material – resulting in occasional maintenance of the bioreactor. Further, with the current process design there is a slim chance of for example MeS being released into the environment.

A possible solution or process improvement, regarding MeS entering the reactor is to add a microfiltration step before the bioreactor. This would remove any residual MeS from the stream entering the bioreactor thus reducing the risk of sedimentation in the reactor.

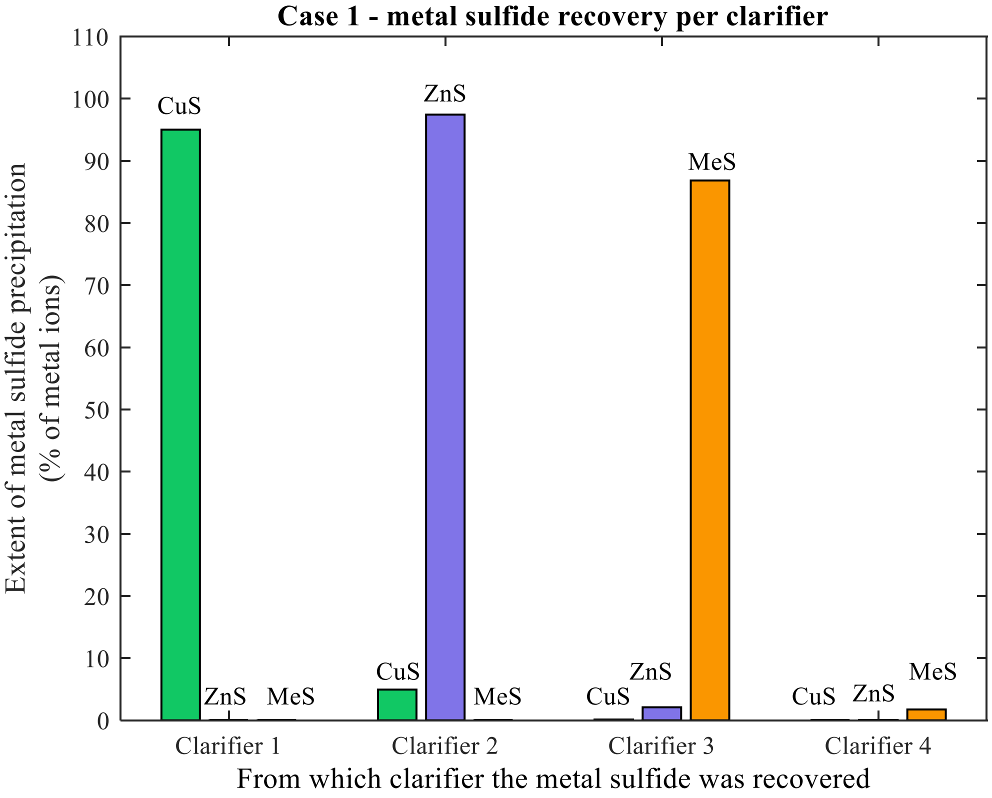


Figure 8 displays the recovery of each species of metal sulfide associated with each of the clarifiers. As seen in the figure, no recovery of either CuS or ZnS is expected in the two latter clarifiers whereas there is an expected residual MeS to be found in the influx stream to the bioreactor.

4.2 Weaknesses associated with the SuperPro model

Throughout this work several weaknesses of the software and its interface have been noticed. Amongst them, is the inability to implement correct pH control for each of the process steps. At present moment, this has to be done by hand, through theoretical determination of the amount of acid or base that is needed to reach the desired pH, under the assumption that the input is homogenous and constant in its composition.

Another noticeable weakness of the software, is the difficulties associated with using the kinetically based fermentation units and bioreactors. As presented here, several attempts were made to incorporate the kinetic unit operations into the model but all failed. This weakness is made up partially of the inadequate inhibitory terms that are provided by the software. Another part of the problem, is that even though the kinetic expression was adapted to fit the growth terms provided by the software, nothing was produced in the reactor.

Apart from the limitation in the usage of the kinetic mode, there is a limitation associated with the unit operations that are available when simulating fermentation processes. The real life process is based on an UASB reactor but it was simulated using a stoichiometric fermenter. A welcome improvement would be to increase the number of fermentation based unit operations from the standard fermenter, CSTR and bioreactor to include other commonly used reactor types such as an UASB reactor, fluidized bed reactor or a membrane reactor which has purposes in both microbial and enzymatic applications.

Additionally, the software does not automatically divide soluble substances into their separate species when entered into an aqueous solution, this concerns for example soluble salts, acids and bases. Thus, for the correct species to be formed or used as reactants, it is necessary to define these under the "Pure components". However, this does not ensure that the software allows the species to exist under the correct form during the reaction, as this is based on what reference component was chosen during the registration process.

4.3 Matlab model and simulation

The Matlab code can be found in its entirety in Appendix A.5 and A.6. Appendix A.5 references the script based on the SuperPro model using the optimized carbon source methanol (Scenario 2: optimized carbon source, default clarifier settings - case 1). Appendix A.6 displays the extended script where the optimized mixture of methanol and acetate were used as carbon source (Scenario 2: optimized carbon source, default clarifier settings - case 3). As the code is based on the previously referenced bioreactor Matlab model developed by Kvarnström and Lönntoft, the same method was used to expand the script to include the precipitation steps as well. The model was based on the collaboration of three main function files: the model file, the file containing the parameters and the solver which used an ODE (ordinary differential equation) solver.

For the purpose of this simulation an ode15s solver was used due to the stiffness of the problem, but this can be changed to either an ode23s or ode45s depending on the scenario the model is adapted to.

As previously mentioned, two separate versions of the script were developed. The first, was based on the stream values associated with case 1 in Scenario 2 (default settings of the clarifiers, optimized substrate usage). The second version of the script, combined acetate and methanol as substrates. Additionally, values from the SuperPro script containing non-optimized clarifiers but an optimized substrate feed. The two adaptions of the script were used to see if, and what differences the usage of two substrates would make compared to one.

Figure 9, displays the resulting curves over the production of reduced sulfur (liquid) and biomass as well as the consumption of methanol and sulfate. The overall appearance of the graphs is as expected, the curves representing of the products are increasing whereas the concentration of the substrate and sulfate are decreasing. The shape of the curves are thought to be a result of the applied kinetics. Approximately 10 h into the process run, at the same time as the methanol consumption reaches its steady state condition, there is an unexpected spike in the concentration of reduced sulfur (liquid). This might be caused by the connection of the bioreactor and precipitation mass balances, due to the reaction kinetics that are applied to the script as these are based on literature data from lab scale experiments (see for example Nagpal *et al.* (1998) and Moosa *et al.* (2002)).

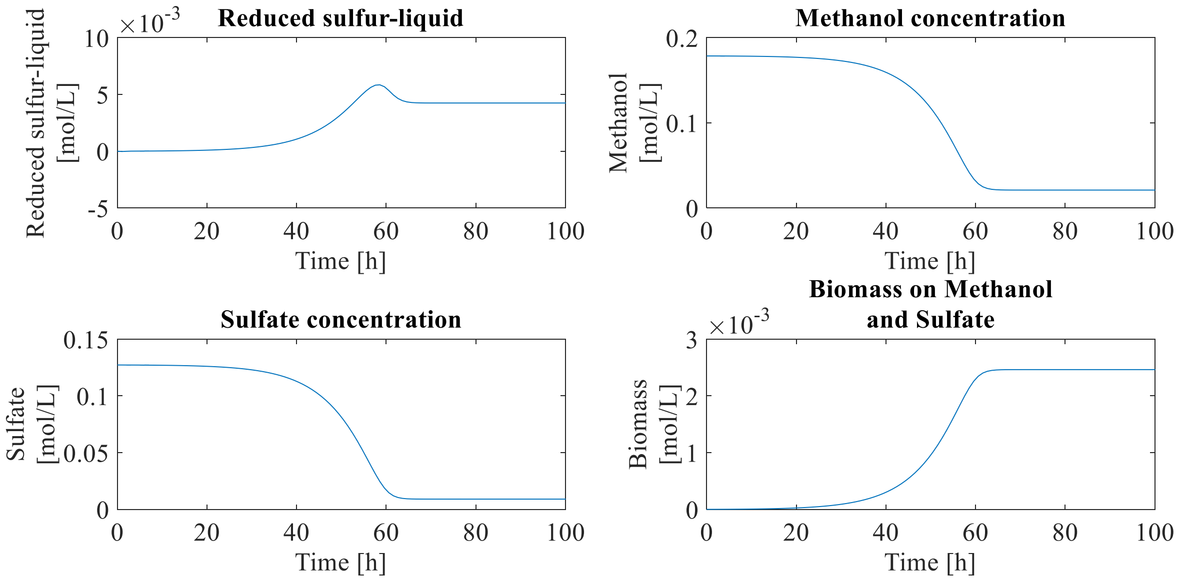


Figure 9 displays the resulting curves over sulfate and methanol consumption as well as reduced sulfur (liquid) and biomass production.

Figure 10, displays the resulting production of H_2S (g) in the upper left corner of the subplot, and the produced H_2S (g) compared to the production of reduced sulfur species in liquid phase, found in the lower right corner. The figure clearly displays, that the majority of the produced hydrogen sulfide is present in the liquid phase and only a limited amount is available as gas at pH 7.

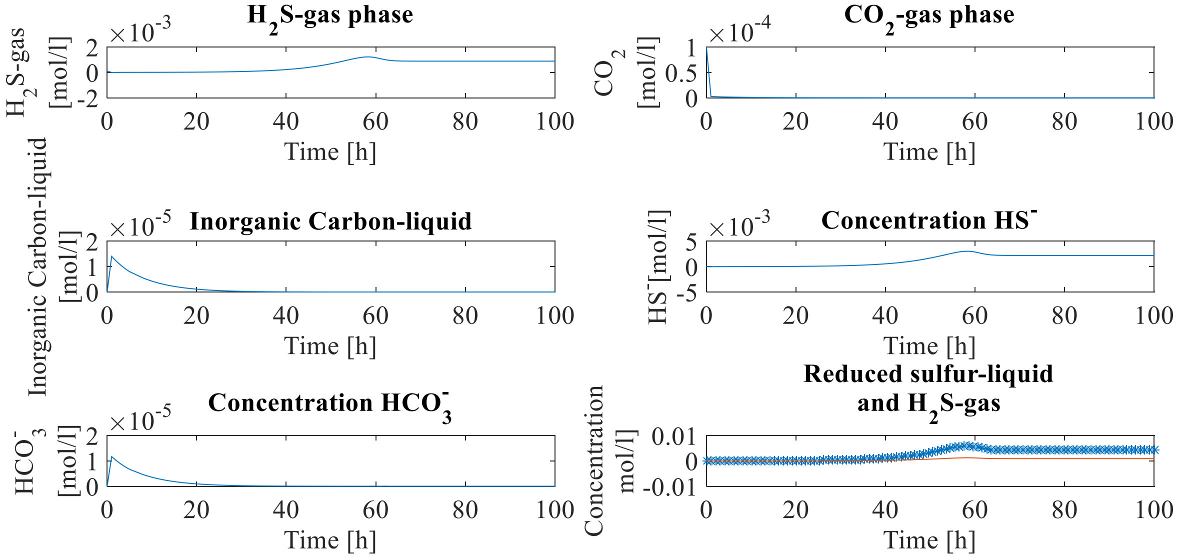


Figure 10 displays the resulting concentrations of the produced H_2S (g), HS^-, CO_2 (g), inorganic carbon (liquid) and HCO_3^-. As well as a comparison between the concentrations of produced H_2S (g) (—) and reduced sulfur (liquid) (*).

The reduction in metal ion concentrations, due to the precipitation of metal ions, are presented in Figure 11. The graphs adapted an overall linear model due to the rate equation of metal precipitation being described as a first degree kinetic expression. The figure shows that within the first 5 h of the process, the reduction of metal ions reach 100% meaning that the residual concentration of ions is 0 mol/L.

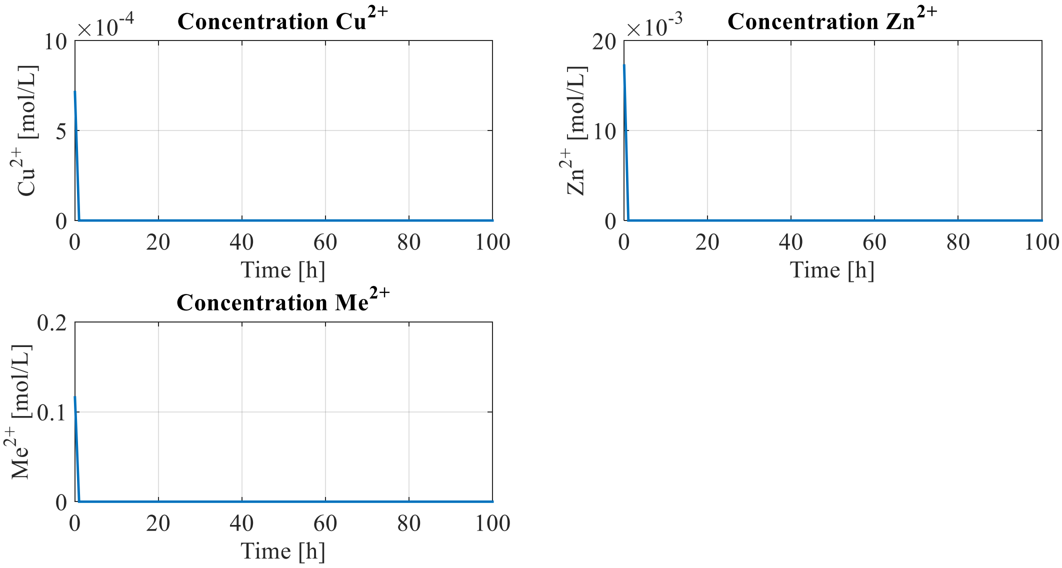


Figure 11 displays the reduction in concentration for copper, zinc and metal ions over a period of 100 h.

Figure 12, displays the resulting concentrations of methanol, acetate and sulfate due to consumption as well as the resulting concentrations from the production of reduced sulfur liquid and biomass for the specified conditions of scenario 2, case 3. The overall resulting shape of the curves matches the expectations for substrate uptake and production of metabolites. The following results should be regarded as uncertain due to the acetate consumption doesn't reach the expected amount. This could be caused by the definition of the reaction kinetics, as previously discussed, since they are adapted for a lab scale setting.

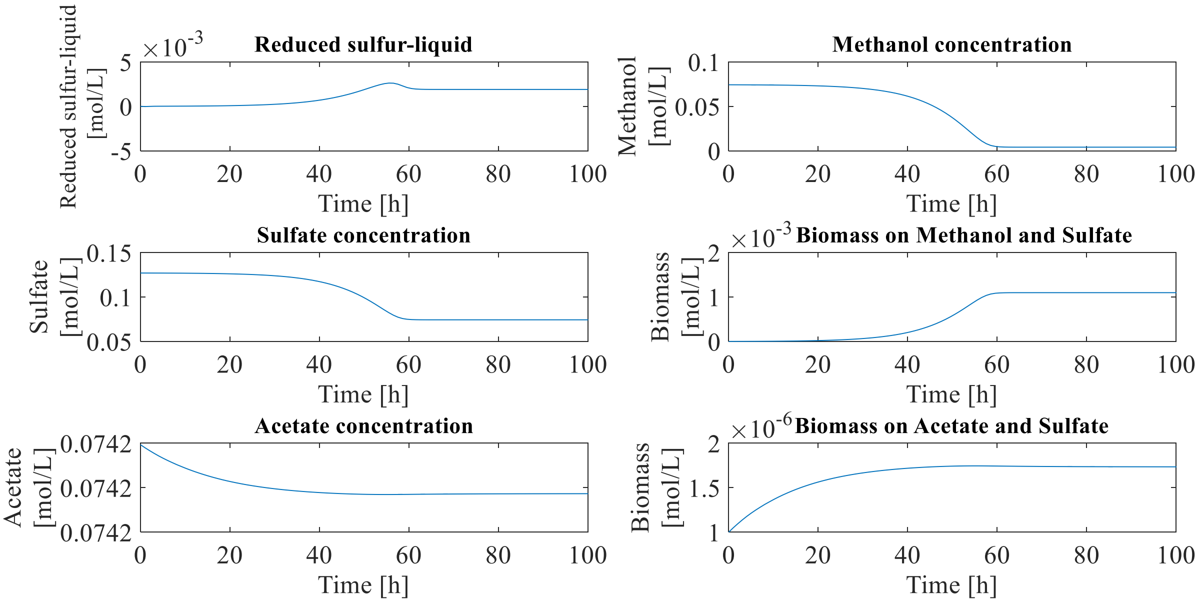


Figure 12 displays the reduction of sulfate, methanol and acetate across time and the simultaneous production of biomass (on both methanol and acetate) and reduced sulfur (liquid).

Further, Figure 13 displays the resulting the specific production of H_2S (g) in the upper left corner of the subplot, and the produced H_2S (g) compared to the production of reduced sulfur species in liquid phase, found in the lower right corner. The figure displays, as previously, that the majority of the produced H_2S is present in the liquid phase and only a limited amount is available as gas at pH 7. The discussion regarding the results when using a single substrate is valid in this segment as well.

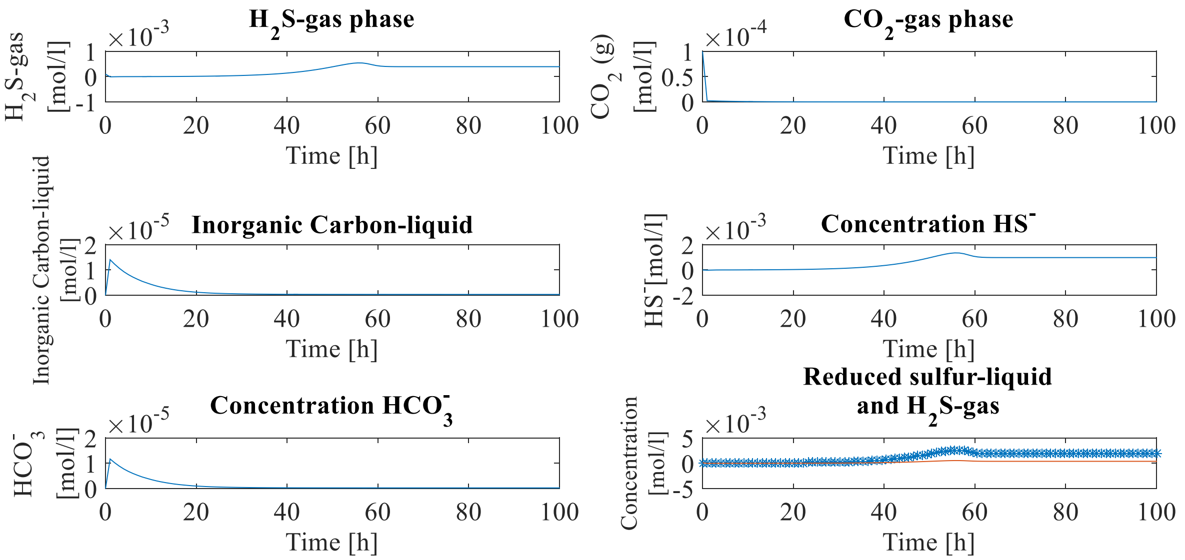


Figure 13 displays the resulting concentrations of the produced H_2S (g), HS^-, CO_2 (g), inorganic carbon (liquid) and HCO_3^-. As well as a comparison between the concentrations of produced H_2S (g) and reduced sulfur (liquid).

The reduction of metal ion concentrations, due to precipitation reactions, are presented in Figure 14. The graphs adapted an overall linear model due to the rate equation of metal precipitation being described as a first degree kinetic expression and have the same appearance as previously presented for case 1. The same discussion regarding the allowed effluent concentrations applies to this simulation as well.

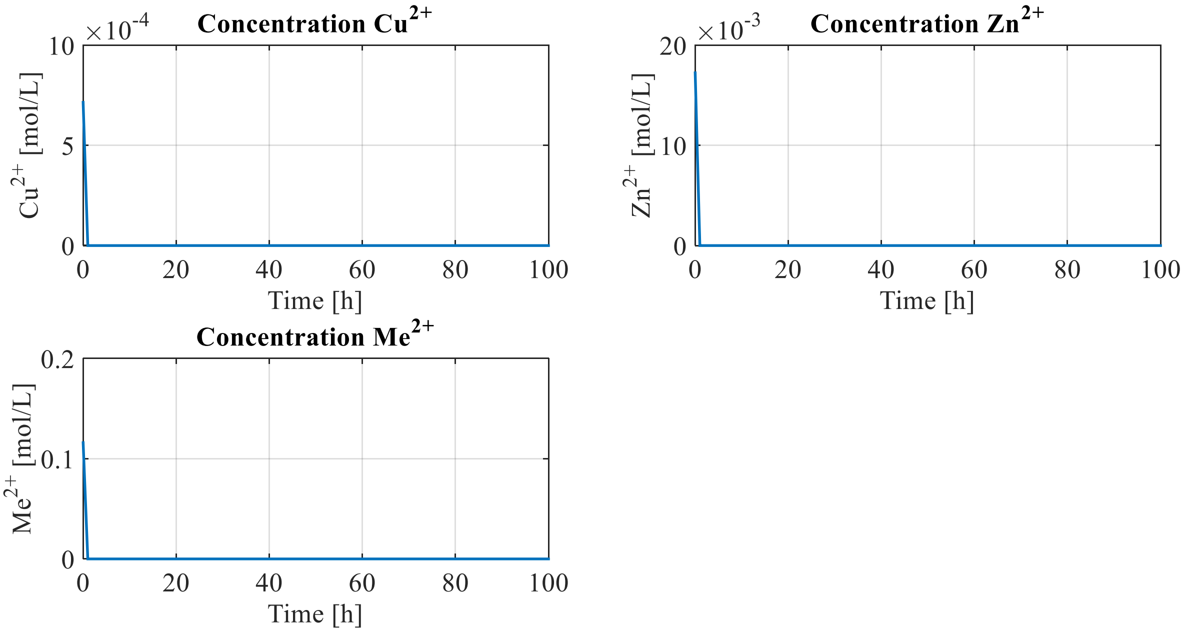


Figure 14 displays the reduction in concentration for copper, zinc and metal ions for a period of 100 h.

The bioreactor model which the current model was based on, contained various parameters from several lab studies such as Nagpal *et al.* (1998) and Moosa *et al.* (2000). However, these parameters were first of all adapted to lab scale experiments and not to industrial scale processes. Further, parameters such as $K_{i,H2S}$ and μ_{max} had a large impact on the appearance of the curve. The results presented above, belong to $K_{i,H2S} = 5.13\times10^{-3}$ mol/L and $\mu_{max} = 0.612$ h^{-1} (Nagpal, et al., 1998; Barrera, et al., 2015).

Since the Matlab model presented in this report, is based on the model developed by Kvarnström and Lönntoft (2017), a comparison could be made between the results gathered here and the ones achieved in the previous model. The parameters that will be compared includes the substrate consumption, biomass production and production of reduced sulfur liquid (S_{red} in the Matlab files).

There were many differences being expected between the results of the current model and the previous script as the previous model was developed for a batch process whereas the current one simulated a continuous process. The current process also involves the three precipitation tanks which results in that the produced S_{red} is used in other parts of the process. Finally, the flows and amounts associated with the current process are approximately 1000 times greater than the ones found in the previous bioreactor model which accounts for the differences in scale on the y-axis. Thus, main comparison between the current model and the previous one concerns the overall behavior of the curves rather than specific curve behavior.

Figure 15, displays the model results reported by Kvarnström and Lönntoft (2017). The behavior of the curves in their model, is mainly due to the process being in batch mode. However, the curves over consumption and production all exhibit their expected behavior. Batch mode processes have no inflow or outflow from the process – i.e. what is entered at the start of the process is what will be available for the course of its duration. Thus, there is an expectation of continuous decrease of the substrate concentrations as well as a constant increase of the produced species such as reduced sulfur (liquid) and biomass, since no steady state of the processes will be reached.

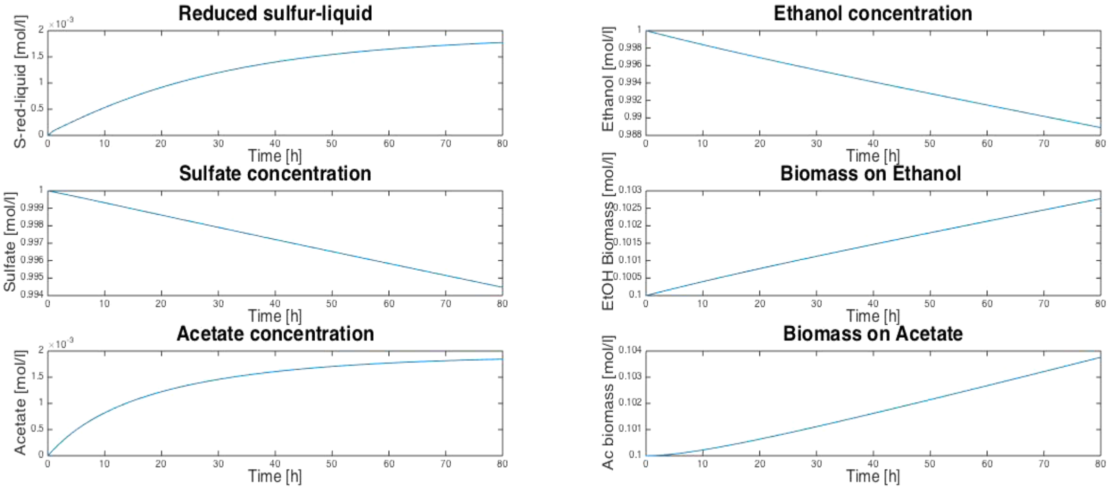


Figure 15 displays the produced reduced sulfur (liquid) and biomass on both methanol and acetate as well as the reducing concentrations of ethanol, acetate and sulfate for a batch run of the bioreactor. The figure is a result from the script developed by Kvarnström and Lönntoft (2017).

4.4 Weaknesses associated with the Matlab model

In its current state, the Matlab model is not fully functioning. There is something that is bugging within the model, most likely due to the addition of linked metal precipitation differential equations rather than the software itself. The main concern is the unexpected spike of “reduced sulfur – liquid” present at approximately 60 h. Thus, additional debugging of the script is necessary.

Further, the actual process of precipitation and the most likely rate limiting step of crystal formation has not been taken into consideration. The current reaction rates assumes that the precipitation occurs linearly, which is probably not true, it’s most likely some sort of an exponential relationship due to the steps of supersaturation and crystal formation kinetics.

For the model to accurately describe the unit operations many more parameters and relationships have to be defined compared to the current model, if it’s supposed to work alongside the SuperPro model – which has default values one can operate the model with if certain parameters remain unknown. Additionally, all of the parameters are currently defined for the process should be defined and validated for industrial scale utilization, and not necessarily based on the experimental/literature values as they currently are.

5. Reflection and future recommendations

The developed models are adaptable by removing, silencing or adding one or more of the precipitation steps. This is encouraged if for example only CuS is desired or if it is acceptable to let all of the remaining metal ions precipitate together at a higher pH.

The accuracy of a SuperPro model relies heavily on the amount of detail that is applied to the process parameters and setup of the different unit operations and compounds that take part in the process. A higher level of detail thus leads to a more accurate model and prediction of the performance of the process. A future recommendation is therefore to increase the level of detail found in each of the unit operations as well as for each of the compounds used in the process. As of now, the level of detail used in the simulation should be considered as a source of error.

In the SuperPro model, the biomass filtered off from the process could be destroyed and reentered into the bioreactor as nitrogen source, to increase the overall resource efficiency of the process.

As previously mentioned, enabling the kinetic mode in either of the bioreactor/fermenter unit operations available would bring an increased level of control into the process in SuperPro. It would be easier to resemble the real life bioreactor compared to a model which doesn’t rely on as many assumptions, first and foremost considering economic aspects of the process. Further, improvements would be more accurate gas liquid and acid base reactions that would extend specifically to each of the unit operations. Again, this will result in greater model accuracy and simultaneously bring accuracy to the economic aspects of the process even though they weren’t considered for this project.

Another scenario which would be interesting to examine, is the comparison between the model in a semi-continuous mode with the existing one in continuous mode to see the effects on economics as well as the sizing and utilization of the unit operations.

Previously, the referenced verdict for the allowed levels of SO_4^{2-}, Cu^{2+} and Zn^{2+} emissions into the analysis point, was determined to be the reference point. As can be seen in Figure 9, approximately 100% removal of the copper and zinc ions through their sulfide equivalents. A slightly lower recovery was found for the overall removal of MeS, as the general metal sulfides only passes through two clarifiers. However, the overall removal of metal sulfides is due to the approximations of the process which states that hydrogen sulfide is produced in excess and that 100% of the metal ions will react will available hydrogen sulfide, resulting in the irreversible precipitation of metal sulfides. A more reliable and correct SuperPro model would take experimental results of both the time scale and extent of the precipitation reactions - thus a more accurate outgoing metal sulfide flow would be achieved.

Further, as there is a desire to optimize the hydrogen sulfide production there might be an increased amount of SO_4^{2-} in the effluent water compared to what the current SuperPro simulation states. However, this could be approached in either of two ways - either nothing is done and the SO_4^{2-} is kept at the maximum acceptable effluent level, hydrogen sulfide is produced in excess and later removed from the process. Another way of approaching this is through the installation of holding tanks for both the leachate stream which will ensure a constant, optimized flow of SO_4^{2-} into the process or by installation of storage units for the produced hydrogen sulfide which could either be sold or reentered into the process if the metal ion concentration would spike for some reason.

Finally, the main recommendation is to calibrate and verify the models using lab and experimental data from for example process trial runs as well as partial process experiments as this will increase the accuracy and credibility of the process.

6. Conclusions

With the current findings the Matlab model seem to be a better prediction of the process compared to the SuperPro simulation as the Matlab model takes both the acid base and gas liquid transfer equilibria into account. However, the main disadvantage of the Matlab model is that it is currently not describing the process correctly when the precipitation steps are included. Additionally, there are no fully integrated descriptive equations for the unit operations associated with the process which reduced the accuracy of the model further.

The SuperPro model is more advanced compared to the Matlab model, since it incorporates the mass balances of each specific unit operation into its solution. The SuperPro model is more accurate with respect to the black box model of the process and generates expected results based on the input parameters. For this reason, and because of its interface SuperPro is a more intuitive software to build a software around – as the user instantly can view and review its unit operations.

Based on the results presented and the discussion points that have been raised under both results and as future recommendations the conclusion of this master thesis is that currently, the SuperPro model is a sufficiently accurate representation of the process and due to its layout it is fairly easy to alter depending on the end goal. Further, the Matlab model is in need more work to adequately provide descriptions of the process and its parameters. However, once these models are fully functioning they will provide a powerful simulation tool to predict and analyze process data.

References

Eawag (Swiss Federal Institute of Aquatic Science and Technology); Dorothee Spuhler (seecon international gmbh); Sustainable Sanitation and Water Management Toolbox , 2018. *Upflow Anaerobic Sludge Blanket Reactor (UASB).* [Online] Available at: https://sswm.info/water-nutrient-cycle/wastewater-treatment/hardwares/semi-centralised-wastewater-treatments/upflow-anaerobic-sludge-blanket-reactor-(uasb) [Accessed 03 01 2019].

AFROX, 2011. *Material safety data sheet (MSDS) Hydrogen sulfide (H2S).* [Online] Available at: http://www.afrox.co.za/en/images/Hydrogen_Sulphide_tcm266-27660.pdf [Accessed 06 12 2018].

Ajibade, P. A. & L.Botha, N., 2016. Synthesis and structural studies of copper sulfide nanocrystals. *Results in Physics,* Volume 6, pp. 581-589.

Al-Tarazi, M., Heesink, A. B. M. & Versteeg, G. F., 2003. Precipitation of metal sulphides using gaseous hydrogen sulphide: mathematical modelling. *Chemical Engineering Science,* Volume 59, pp. 567-579.

American Elements, 2018. *Copper Sulfide.* [Online] Available at: https://www.americanelements.com/copper-sulfide-1317-40-4 [Accessed 11 01 2019].

Bai, H. et al., 2013. Bioremediation of copper-cntaining wastewater by sulfate reducing bacteria coupled with iron. *Journal of Environmental Management,* Volume 129, pp. 350-356.

Barrera, L. E. et al., 2015. Modeling the anaerobic digestion of cane-molasses vinasse: Extension of the Anaerobic Digestion Model No. 1 (ADM1) with sulfate reduction for a very high strength and sulfate rich wastewater. *Water Research,* Issue 71, pp. 42-54.

Barton, L. L. & Fauque, G. D., 2009. Chapter 2 Biochemistry, Physiology and Biotechnology of Sulfate-Reducing Bacteria. *Advances in Applied Microbiology,* Volume 68, pp. 41-98.

Boliden, 2018. *Metals for modern life.* [Online] Available at: https://www.boliden.com/globalassets/media/337-6514-metals-for-modern-life-2017-sv_final.pdf [Accessed 21 10 2018].

D’Acunto, B., Esposito, G., Frunzo, L. & Pirozzi, F., 2011. Dynamic modeling of sulfate reducing biofilms. *Computers & Mathematics with Applications,* 62(6), pp. 2601-2608.

Diehl, S., 2001. *Operating charts for continuous sedimentation I: control of steady states.* [Online] Available at: http://www.maths.lth.se/matematiklth/personal/diehl/sdjem1_final_draft.pdf [Accessed 21 10 2018].

Doble, M. & Kumar, A., 2005. Chapter 13 - Treatment of Waste from Metal Processing and Electrochemical Industries. In: *Biotreatment of Industrial Effluents.* s.l.:Elsevier, pp. 145-155.

Donlan, R. M., 2002. Biofilms: Microbial Life on Surfaces. *Emerging Infectuous Diseases,* 8(9), pp. 881-890.

Duc, M. Le, 2011. *Induktion, deduktion och abduktion,* s.l.: s.n.

EIT RawMaterials, 2018A. *Terrafame Group launches a new research project supported by EIT RawMaterials.* [Online] Available at: https://eitrawmaterials.eu/terrafame-group-launches-new-research-project-supported-by-eit-rawmaterials/ [Accessed 21 10 2018].

EIT RawMaterials, 2018B. *SO4Control. Scale-up of Solution for Mining Water Sulfate Control with Side-product Recovery.* [Online] Available at: https://eitrawmaterials.eu/project/so4control/ [Accessed 21 10 2018].

European Commission, 2018A. *METGROW+ About.* [Online] Available at: http://metgrowplus.eu/the-project/ [Accessed 21 10 2018].

European Commission, 2018B. *METGROW+ EXPECTED IMPACT.* [Online] Available at: http://metgrowplus.eu/expected-impact/ [Accessed 21 10 2018].

European Commission, 2018C. *METGROW+ OBJECTIVES.* [Online] Available at: http://metgrowplus.eu/objetives/ [Accessed 21 10 2018].

Farm Water Supply Branch Alberta Agriculture and Forestry, 2018. *Removing Hydrogen Sulfide Gas from Water.* [Online] Available at: https://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/agdex1160 [Accessed 28 10 2018].

Hammer, M. J. & Hammer, M. J., 2011. *Water and wastewater technology.* 7 ed. s.l.:Upper Saddle River, N.J. : London : Prentice Hall ; Pearson Education .

Hao, T.-w.et al., 2014. A review of biological sulfate conversions in wastewater treatment. *Water Research,* Issue 65, pp. 1-21.

Heck, H. H., Hall, M. L., Santos, R. d. & Tomadakis, M. M., 2018. Pressure swing adsorption separation of H2S/CO2/CH4 gas mixtures with molecular sieves 4A, 5A, and 13X. *Separation Science and Technology,* 53(10), pp. 1490-1497.

Homostomsky, J. & Jones, A. G., 1995. Penetration model of gas-liquid reactive precipitation of calcium carbonate crystals. *Transactions of I. Ch. E. M. E,* 73(A), pp. 241-245.

Hu, X. et al., 2016. Treatment of simulation of copper-containing pit wastewater with sulfate-reducing bacteria (SRB) in biofilm reactors. *Environmental Earth Sciences,* 75(1305).

Ibrahim, S. & Raj, A., 2016. Kinetic Simulation of Acid Gas (H2S and CO2) Destruction for Simultaneous Syngas and Sulfur Recovery. *Industrial & Engineering Chemistry Research,* 55(24), pp. 6743-6752.

Intelligen Inc. , 2018. *SuperPro Designer - overview.* [Online] Available at: http://intelligen.com/superpro_overview.html [Accessed 12 11 2018].

Iowa State University, n.d. *Appendix 1. Theoretical Oxygen Demand (TOD) Values of Organic Compounds (adapted from Pitter and Chudoba, 1990).* [Online] Available at: http://home.eng.iastate.edu/~tge/ce326/6.13.4.5.appendx1.doc [Accessed 03 01 2019].

Jones, A. G., Hostomsky, J. & Zhou, L., 2014. On the effect of liquid mixing rate on primary crystal size during the gas-liquid precipitation of calcium carbonate. *Chemical Engineering Science,* Volume 47, pp. 3817-3824.

Kaksonen, A. H., Franzmann, P. D. & Puhakka, J. A., 2004. Effects of Hydraulic Retention Time and Sulfide Toxicity on Ethanol and Acetate Oxidation in Sulfate-Reducing Metal-Precipitating Fluidized-Bed Reactor. *Biotechnology and Bioengineering,* 86(3), pp. 52-59.

Kobayashi, M., Kametani, H. & Yamada, K., 1989. Precipitation of CuS from Acidic Cu2+ Solution with Activated Sulphur Powder by Disproportionation Reaction. *Journal of the Mining Institute of Japan,* Volume 6, pp. 54-67.

Kohl, A. L. & Nielsen, R. B., 1997. Chapter 5 Alkaline Salt Solutions for Acid Gas Removal. In: G. P. Publishing, ed. *Gas Purification 5th ed..* s.l.:Elsevier, pp. 402-404.

Kvarnström, H. & Lönntoft, E., 2017. *Modelling of Metal Recovery using Sulfate Reducing Bacteria,* Lund, Sweden: Dicision of Applied Microbioloy, Lund University.

Liamleam, W. & Annachhatre, A. P., 2007. Electron donors for biological sulfate reduction. *Biotechnology Advances,* 25(5), pp. 452-463.

Lindquist, A., 2003. *About water treatment.* Helsingborg: Kemira kemwater.

Mamrosh, D. L. et al., n.d. *Removal of Hydrogen Sulfide and REcovery of Carbon Dioxide from Geothermal Non-Condensable Gas Using Water.* [Online] Available at: http://www.trimeric.com/assets/removal-of-hydrogen-sulfide-and-recovery-of-carbon-dioxide-from-geothermal-non-condensable-gas-using-water.pdf [Accessed 04 10 2018].

McVay, R. D., n.d. *Processes for Removal of Hydrogen Sulfide Found in Florida Ground Water Sources.* [Online] Available at: https://www.google.com/url?sa=t&rct=j&q=&esrc=s&source=web&cd=1&ved=2ahUKEwj654_z1qbfAhXClSwKHQSBAdcQFjAAegQICRAC&url=http%3A%2F%2Fwww.frwa.net%2Fuploads%2F4

%2F2%2F3%2F5%2F42359811%2Fremovalofhydrogensulfide010511.doc&usg=AOvVaw2H7dBSrtJZgHNmnWK2Gode [Accessed 05 11 2018].

Minnesota Rural Water Association, n.d. *Membrane Filtration.* [Online] Available at: https://www.mrwa.com/WaterWorksMnl/Chapter%2019%20Membrane%20Filtration.pdf [Accessed 21 10 2018].

Moosa, S., Nemati, M. & Harrison, S. T. L., 2002. A kinetic study on anaerobic reduction of sulphate, Part I: Effect of sulphate concentration. *Chemical Engineering Science,* Volume 57, pp. 2773-2780.

Mu, S., Zeng, Y., Tartakovsky, B. & Wu, P., 2007. Simulation and control of an upflow anaerobic sludge blanket (UASB) reactor using ADM1-based distributed parameter model. *Industrial & Engineering Chemical Research*, 46(5), pp. 1519-1526.

Muyzer, G. & Stams, A. J. M., 2008. The ecology and biotechnology of sulphate-reducing bacteria. *Nature Reviews Microbiology,* pp. 1-15.

Nagpal, S., Chuichulcherm, S., Livingston, A. & Peeva, L., 1998. Ethanol Utilization by Sulfate-Reducing Bacteria: An Experimental and Modeling Study. *Biotechnology and Bioengineering,* Volume 70, pp. 533-543.

Nagpal, S., Chuichulcherm, S., Peeva, L. & Livingston, A., 2000. Microbial Sulfate Reduction in a Liquid-Solid Fluidized Bed Reactor. *Biotechnology and bioengineering,* Volume 70, pp. 370-380.

National Center for Biotechnology Information, 2018. *Hydrogen Sulfide.* [Online] Available at: https://pubchem.ncbi.nlm.nih.gov/compound/hydrogen_sulfide#section=Solubility [Accessed 04 10 2018].

National Center for Biotechnology Information, 2018. *Sodium Bisulfide.* [Online] Available at: https://pubchem.ncbi.nlm.nih.gov/compound/sodium_hydrosulfide#section=Boiling-Point
[Accessed 21 10 2018].

National Center for Biotechnology Information, 2019. *Zinc sulfide.* [Online] Available at: https://pubchem.ncbi.nlm.nih.gov/compound/zinc_sulfide#section=Solubility [Accessed 11 01 2019].

Nizami, A.-S., Singh, A. & Murphy, J. D., 2011. Design, commissioning and start-up of a sequentially fed leach bed reactor complete with upflow anaerobic sludge blanket digesting grass silage. *Energy and Fuels,* Volume 25, pp. 823-834.

Nordic Water, 2018. *Lamella Separator LS.* [Online] Available at: https://www.nordicwater.com/sv/product/lamellaseparator-ls/ [Accessed 04 10 2018].

Oregon State Government, n.d. [Online] Available at: https://www.oregon.gov/oha/PH/HEALTHYENVIRONMENTS/DRINKINGWATER/OPERATIONS/TREATMENT/Documents/Sedimentation_Clarification.pdf [Accessed 28 10 2018].

Price, R. M., 2005. *Recycle and Bypass Processes.* [Online] Available at: http://facstaff.cbu.edu/rprice/lectures/recycle.html [Accessed 28 10 2018].

Saha, U. et al., 2017. *Your household water quality: removal of hydrogen sulfide and sulfate.* [Online] Available at: http://extension.uga.edu/publications/detail.html?number=C858-15&title=Removal%20of%20Hydrogen%20Sulfide%20and%20Sulfate [Accessed 06 12 2018].

SCB, 2000. *Vattenräkenskaper, Rapport 2000:6,* Stockholm, Sweden: SCB.

SCB, 2017A. *Industrins vattenutsläpp, 1000-tal kubikmeter efter typ av recipient och år, oregelb.* [Online] Available at: http://www.statistikdatabasen.scb.se/pxweb/sv/ssd/START__MI__MI0902__MI0902F/UtslappVatten/table/tableViewLayout1/?rxid=f45f90b6-7345-4877-ba25-9b43e6c6e299# [Accessed 21 10 2018].

SCB, 2017B. *Industrins vattenanvändning efter näringsgren SNI 2007, typ av användning, tabellinnehåll och år, oregelb.* [Online] Available at: http://www.statistikdatabasen.scb.se/pxweb/sv/ssd/START__MI__MI0902__MI0902E/VattenAnvSN

I2007/table/tableViewLayout1/?rxid=f45f90b6-7345-4877-ba25-9b43e6c6e299 [Accessed 21 10 2018].

Singh, L. R. & Singh, S. A., 2012. Particle Size Effect on TL Emission of ZnS Nanoparticles and Determination of Its Kinetic Parameters. *Journal of Nanomaterials.*

Suez , 2018. *degremont water handbook - lamellar sedimentation.* [Online] Available at: https://www.suezwaterhandbook.com/water-and-generalities/fundamental-physical-chemical-engineering-processes-applicable-to-water-treatment/sedimentation/lamellar-sedimentation [Accessed 04 10 2018].

Suez Water Technologies & Solutions, 2018. *External Treatment: Chapter 05 - Clarification.* [Online] Available at: https://www.suezwatertechnologies.com/handbook/chapter-05-clarification [Accessed 04 10 2018].

Suez, 2018. *Suez degremont water handbook - clarification.* [Online] Available at: https://www.suezwaterhandbook.com/processes-and-technologies/biological-processes/suspended-growth-cultures/clarification [Accessed 21 10 2018].

SVEA HOVRÄTT Mark- och miljööverdomstolen, 2016. *DOM 2016-10-18 Stockholm, Mål nr M 4160-15.* [Online] Available at: http://www.markochmiljooverdomstolen.se/Domstolar/markochmiljooverdomstolen/M%204160-15%20Dom%202016-10-18.pdf [Accessed 28 10 2018].

Sveriges geologiska undersökning, n.d. *Bergarter.* [Online] Available at: https://www.sgu.se/om-geologi/berg/bergarter/ [Accessed 21 10 2018].

The International Water Association Publishing, 2019. *Up Flow - Anaerobic Sludge Blanket Reactor (UASB).* [Online] Available at: https://www.iwapublishing.com/news/flow-anaerobic-sludge-blanket-reactor-uasb [Accessed 03 01 2019].

The International Water Association, 2018. *Up Flow - Anaerobic Sludge Blanket Reactor (UASB).* [Online]
Available at: https://www.iwapublishing.com/news/flow-anaerobic-sludge-blanket-reactor-uasb [Accessed 21 10 2018].

Tokuda, H. et al., 2008. Study on reaction kinetics and selective precipitation of Cu, Zn, Ni and Sn with H2S in single-metal and multi-metal systems. *Chemosphere,* Volume 73, pp. 1448-1462.

Tokuda, H. et al., 2008. Study on reaction kinetics and selective precipitation of Cu, Zn, Ni and Sn with H2S in single-metal and multi-metal systems.. *Chemosphere,* Issue 73, pp. 1448-1452.

V.Shadrunova, I. & Orekhova, N. N., 2015. A Process for Advanced Recycling of Water Originating from Mining Operations, with Metal Recovery. *Mine Water and the Environment,* 34(4), pp. 478-484.

Weijma, J., 2000. *Methanol as electron donor for thermophilic biological sulfate and sulfite reduction* , Wageningen, The Netherlands: Wageningen University.

Wikipedia, 2018. *Sulfate-reducing microorganisms.* [Online] Available at: https://en.wikipedia.org/wiki/Sulfate-reducing_microorganisms [Accessed 21 10 2018].

Villadsen, J., Nielsen, J. & Lidén, G., 2011. *Bioreaction Engineering Principles.* 3rd, pg. 105 ed. London: Springer Science+Business Media, LLC.

Villadsen, J., Nielsen, J. & Lidén, G., 2011. Chapter 11 Scale-Up of Bioprocesses. In: *Bioreaction Engineering Principles 3rd edition.* London: Springer Science+Business Media, LLC, pp. 497-541.

Wouda, T., Rietema, K. & Ottengraf, S., 1977. Continuous sedimentation theory: Effects of density gradients and velocity profiles on sedimentation efficiency. *Chemical Engineering Science,* 32(4), pp. 351-358.

Appendix A.1 – Pure components and stock mixtures, defined in SuperPro

Local name	Database reference component	Additional information
Cu2+	Copper	
Zn2+	Zinc	
Me2+	Zinc	Average molecular weight was set to 58.693 g/mol.
Na	Sodium	
Hydr. sulf.	Hydrogen sulfide	
H2S(aq)	Hydrogen sulfide + water	
Sulfur	Sulfur	
CuS	Zinc sulfate	Mw: 95.606 g/mol, physical properties of CuS
CuS(s)	Zinc sulfate	Mw: 95.606 g/mol, physical properties of CuS. Simulates the solidification of the metal precipitate.
ZnS	Zinc sulfate	Mw: 97.474 g/mol, physical properties of ZnS
ZnS(s)	Zinc sulfate	Mw: 97.474 g/mol, physical properties of ZnS. Simulates the solidification of the metal precipitate.
MeS	Zinc sulfate	Mw: 90.753 g/mol, physical properties of ZnS.
MeS(s)	Zinc sulfate	Mw: 90.753 g/mol, physical properties of ZnS. Simulates the solidification of the metal precipitate.
Water	Water	
Biomass	Biomass	
Air	Air	Stock mixture in SuperPro
NH4	Ammonium sulfate	Mw: 18.052 g/mol. Simulates nitrogen source for biomass production.
H+	Water	Mw: 1.008 g/mol, to balance reactions
Sulfuric acid	Sulfuric acid	Simulates SO_4^{2-}, Mw: 96.06 g/mol
H2CO3	Carbon dioxide and water.	Mw: 62.026 g/mol, physical properties of CO_2 and water. Includes all species of CO_2 found in the aqueous phase i.e. CO_2 (aq), CO_2 (g) and HCO_3^-.
CO2(aq)	Carbon dioxide	Simulation of the dissolved CO_2 found in the aqueous phase
Methanol	Methanol	Carbon source in fermentation.
Acetic acid	Acetic acid	Acetate. Carbon source in fermentation.

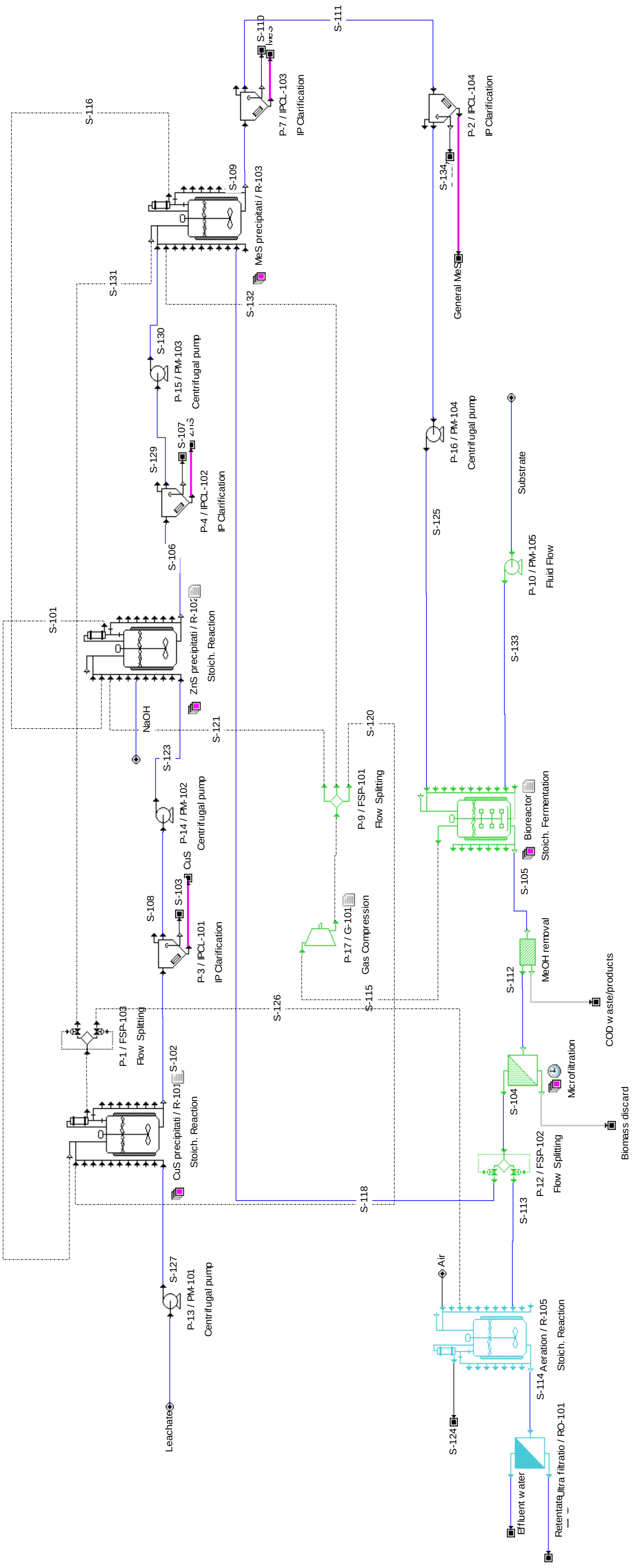
Appendix A.2 – Leachate stream composition

Species	Concentration/Flow entering the process
SO_4^{2-}	33 311 (mol/h)
Cu^{2+}	89 (mol/h)
Zn^{2+}	2147 (mol/h)
Me^{2+}	14 500 (mol/h)
Water	121 000 (kg/h)

Appendix A.3 – SuperPro flowchart and “Material and stream” report

Primarily, a flowchart over case 1, scenario 2 (optimized carbon source, default settings on clarifiers) is displayed as a guide to the following “Material & Stream” report. The flowchart serves as a guidance for the “Equipment” report found in Appendix A.4 as well.

The “Material & Stream” report references a so called main branch, which is the same as the overall flow direction of the model. Additionally, the report talks about “Sections”. There are three sections present in the SuperPro model: “Precipitation”, “Bioreactor” and “Effluent treatment”. The sections are a result of the buildup of the SuperPro model, as it was created according to the steps precipitation/upstream processing, the bioreactor and finally the downstream processing/effluent treatment.


Leachate
S-127
P-13 / PM-101
Centrifugal pump
CuS precipitati / R-101
Stoich. Reaction
S-102
P-1 / FSP-103
Flow Splitting
S-101
S-116
S-131
S-126
P-3 / IPCL-101
IP Clarification
S-108
S-103
CuS
P-14 / PM-102
Centrifugal pump
S-123
NaOH
S-121
ZnS precipitati / R-102
Stoich. Reaction
S-106
P-4 / IPCL-102
IP Clarification
S-129
S-107
ZnS
P-15 / PM-103
Centrifugal pump
S-130
S-132
MeS precipitati / R-103
S-109
P-7 / IPCL-103
IP Clarification
S-110
MeS
S-111
P-2 / IPCL-104
IP Clarification
S-134
General MeS
P-16 / PM-104
Centrifugal pump
S-125
S-120
P-9 / FSP-101
Flow Splitting
P-17 / G-101
Gas Compression
S-115
S-118
Substrate
P-10 / PM-105
Fluid Flow
S-133
Bioreactor
Stoich. Fermentation
S-105
MeOH removal
S-112
COD waste/products
Microfiltration
S-104
Biomass discard
P-12 / FSP-102
Flow Splitting
S-113
Air
Aeration / R-105
Stoich. Reaction
S-114
S-124
Ultra filtratio / RO-101
Effluent water
Retentate

Materials & Streams Report *for Case 1- MeOH opt- 0.67 RISE PARTNER data*

1. OVERALL PROCESS DATA

Annual Operating Time	7920.00 h
Operating Days per Year	330.00
MP = Undefined	

2.1 STARTING MATERIAL REQUIREMENTS (per Section)

Section	Starting Material	Active Product	Amount Needed (kg Sin/kg MP)	Molar Yield (%)	Mass Yield (%)	Gross Mass Yield (%)
Main Section	(none)	(none)	Unknown	Unknown	Unknown	Unknown
Precipitation	(none)	(none)	Unknown	Unknown	Unknown	Unknown
Bioreactor	(none)	(none)	Unknown	Unknown	Unknown	Unknown
Effluent treatment	(none)	(none)	Unknown	Unknown	Unknown	Unknown

Sin = Section Starting Material. Aout = Section Active Product

2.2 BULK MATERIALS (Entire Process)

Material	kg/yr	kg/h	kg/kg MP
Air	792000	100.00	
Cu2+	44849	5.66	
H+	76859	9.70	
Me2+	6740631	851.09	
Methanol	6966586	879.62	
NaOH (50% w/w)	14	0.00	
NH4	39327	4.97	
Sulfuric Acid	25874640	3267.00	
Water	958320000	121000.00	
Zn2+	1111651	140.36	
TOTAL	999966558	126258.40	

2.3 BULK MATERIALS (per Section)

SECTIONS IN: Main Branch

Precipitation

Material	kg/yr	kg/h	kg/kg MP
Cu2+	44849	5.66	
Me2+	6740631	851.09	
NaOH (50% w/w)	14	0.00	
Sulfuric Acid	25874640	3267.00	
Water	958320000	121000.00	
Zn2+	1111651	140.36	
TOTAL	992091786	125264.11	

Bioreactor

Material	kg/yr	kg/h	kg/kg MP
H+	76859	9.70	
Methanol	6966586	879.62	
NH4	39327	4.97	
TOTAL	7082772	894.29	

Effluent treatment

Material	kg/yr	kg/h	kg/kg MP
Air	792000	100.00	
TOTAL	792000	100.00	

2.4 BULK MATERIALS (per Material)

Air	% Total	kg/yr	kg/h	kg/kg MP
Effluent treatment (Main Branch)				
Aeration	100.00	792000	100.00	
TOTAL	100.00	792000	100.00	

Cu2+	% Total	kg/yr	kg/h	kg/kg MP
Precipitation (Main Branch)				
P-13	100.00	44849	5.66	
TOTAL	100.00	44849	5.66	

H+	% Total	kg/yr	kg/h	kg/kg MP
Bioreactor (Main Branch)				
P-10	100.00	76859	9.70	
TOTAL	100.00	76859	9.70	

Me2+	% Total	kg/yr	kg/h	kg/kg MP
Precipitation (Main Branch)				
P-13	100.00	6740631	851.09	
TOTAL	100.00	6740631	851.09	

Methanol	% Total	kg/yr	kg/h	kg/kg MP
Bioreactor (Main Branch)				
P-10	100.00	6966586	879.62	
TOTAL	100.00	6966586	879.62	

NaOH (50% w/w)	% Total	kg/yr	kg/h	kg/kg MP
Precipitation (Main Branch)				
ZnS precipitati	100.00	14	0.00	
TOTAL	100.00	14	0.00	

NH4	% Total	kg/yr	kg/h	kg/kg MP
Bioreactor (Main Branch)				
P-10	100.00	39327	4.97	
TOTAL	100.00	39327	4.97	

Sulfuric Acid	% Total	kg/yr	kg/h	kg/kg MP
Precipitation (Main Branch)				
P-13	100.00	25874640	3267.00	

TOTAL	100.00	25874640	3267.00	
Water	**% Total**	**kg/yr**	**kg/h**	**kg/kg MP**
Precipitation (Main Branch)				
P-13	100.00	958320000	121000.00	
TOTAL	100.00	958320000	121000.00	
Zn2+	**% Total**	**kg/yr**	**kg/h**	**kg/kg MP**
Precipitation (Main Branch)				
P-13	100.00	1111651	140.36	
TOTAL	100.00	1111651	140.36	

2.5 BULK MATERIALS: SECTION TOTALS (kg/h)

Raw Material	Main Section	Precipitation	Bioreactor	Effluent treatment
Air	0.00	0.00	0.00	100.00
Cu2+	0.00	5.66	0.00	0.00
H+	0.00	0.00	9.70	0.00
Me2+	0.00	851.09	0.00	0.00
Methanol	0.00	0.00	879.62	0.00
NaOH (50% w/w)	0.00	0.00	0.00	0.00
NH4	0.00	0.00	4.97	0.00
Sulfuric Acid	0.00	3267.00	0.00	0.00
Water	0.00	121000.00	0.00	0.00
Zn2+	0.00	140.36	0.00	0.00
TOTAL	0.00	125264.11	894.29	100.00

2.6 BULK MATERIALS: SECTION TOTALS (kg/yr)

Raw Material	Main Section	Precipitation	Bioreactor	Effluent treatment
Air	0	0	0	792000
Cu2+	0	44849	0	0
H+	0	0	76859	0
Me2+	0	6740631	0	0
Methanol	0	0	6966586	0
NaOH (50% w/w)	0	14	0	0
NH4	0	0	39327	0
Sulfuric Acid	0	25874640	0	0
Water	0	958320000	0	0
Zn2+	0	1111651	0	0
TOTAL	0	992091786	7082772	792000

3. STREAM DETAILS

Stream Name	**Leachate**	**S-127**	**Substrate**	**S-133**
Source	**INPUT**	**P-13**	**INPUT**	**P-10**
Destination	**P-13**	**CuS precipitati**	**P-10**	**Bioreactor**

Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	25.00	25.00
Pressure (bar)	1.01	1.04	1.01	1.04
Density (g/L)	1013.90	1013.90	793.83	793.83
Total Enthalpy (kW-h)	3571.30	3571.34	20.98	20.98
Specific Enthalpy (kcal/kg)	24.53	24.53	20.18	20.18
Heat Capacity (kcal/kg-°C)	0.98	0.98	0.81	0.81
Weight Percent (mass)				
Cu2+	0.00	0.00	0.00	0.00
H+	0.00	0.00	1.09	1.09
Me2+	0.68	0.68	0.00	0.00
Methanol	0.00	0.00	98.36	98.36
NH4	0.00	0.00	0.56	0.56
Sulfuric Acid	2.61	2.61	0.00	0.00
Water	96.60	96.60	0.00	0.00
Zn2+	0.11	0.11	0.00	0.00
Component Flowrates (kg/h)				
Cu2+	5.66	5.66	0.00	0.00
H+	0.00	0.00	9.70	9.70
Me2+	851.09	851.09	0.00	0.00
Methanol	0.00	0.00	879.62	879.62
NH4	0.00	0.00	4.97	4.97
Sulfuric Acid	3267.00	3267.00	0.00	0.00
Water	121000.00	121000.00	0.00	0.00
Zn2+	140.36	140.36	0.00	0.00
TOTAL (kg/h)	125264.11	125264.11	894.29	894.29
TOTAL (L/h)	123547.33	123547.33	1126.55	1126.55
Stream Name	**S-125**	**S-115**	**S-105**	**S-119**
Source	**P-16**	**Bioreactor**	**Bioreactor**	**P-17**
Destination	**Bioreactor**	**P-17**	**MeOH removal**	**P-9**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	24.98	25.00	25.00
Pressure (bar)	1.04	1.01	1.01	10.01
Density (g/L)	1003.38	1.58	998.92	15.66
Total Enthalpy (kW-h)	4415.20	81.01	4429.92	81.01
Specific Enthalpy (kcal/kg)	24.80	86.80	24.87	86.80
Heat Capacity (kcal/kg-°C)	0.99	0.22	0.99	0.22
Weight Percent (mass)				

Biomass	0.00	0.00	0.02	0.00
Carb. Dioxide	0.00	53.51	0.00	53.51
CuS (s)	0.00	0.00	0.00	0.00
H+	0.03	0.00	0.03	0.00
H2CO3	1.14	0.00	1.80	0.00
H2S (aq)	0.07	0.00	0.26	0.00
Hydr. Sulfide	0.00	46.49	0.00	46.49
MeS (s)	0.00	0.00	0.00	0.00
Methanol	0.00	0.00	0.00	0.00
NH4	0.00	0.00	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	1.25	0.00	0.00	0.00
Water	97.51	0.00	97.89	0.00
ZnS (s)	0.00	0.00	0.00	0.00
Component Flowrates (kg/h)				
Biomass	0.00	0.00	33.81	0.00
Carb. Dioxide	0.00	429.71	0.00	429.71
CuS (s)	0.00	0.00	0.00	0.00
H+	41.77	0.00	42.03	0.00
H2CO3	1747.08	0.00	2758.89	0.00
H2S (aq)	103.69	0.00	397.01	0.00
Hydr. Sulfide	0.00	373.31	0.00	373.31
MeS (s)	0.47	0.00	0.47	0.00
Methanol	0.00	0.00	0.00	0.00
NH4	0.14	0.00	0.27	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	1918.27	0.00	0.00	0.00
Water	149341.53	0.00	150011.76	0.00
ZnS (s)	0.00	0.00	0.00	0.00
TOTAL (kg/h)	153152.95	803.02	153244.24	803.02
TOTAL (L/h)	152636.50	506816.52	153409.40	51288.67
Stream Name	**S-121**	**S-132**	**S-120**	**S-116**
Source	**P-9**	**P-9**	**P-9**	**MeS precipitati**
Destination	**ZnS precipitati**	**MeS precipitati**	**CuS precipitati**	**ZnS precipitati**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	25.00	24.98
Pressure (bar)	10.01	10.01	10.01	1.01
Density (g/L)	15.66	15.66	15.66	1.80
Total Enthalpy (kW-h)	26.73	26.73	27.54	47.52
Specific Enthalpy (kcal/kg)	86.80	86.80	86.80	61.47

Heat Capacity (kcal/kg-°C)	0.22	0.22	0.22	0.20
Weight Percent (mass)				
Carb. Dioxide	53.51	53.51	53.51	100.00
Hydr. Sulfide	46.49	46.49	46.49	0.00
Component Flowrates (kg/h)				
Carb. Dioxide	141.80	141.80	146.10	665.06
Hydr. Sulfide	123.19	123.19	126.93	0.00
TOTAL (kg/h)	265.00	265.00	273.03	665.06
TOTAL (L/h)	16925.26	16925.26	17438.15	369680.38
Stream Name	**NaOH**	**S-123**	**S-101**	**S-106**
Source	**INPUT**	**P-14**	**ZnS precipitati**	**ZnS precipitati**
Destination	**ZnS precipitati**	**ZnS precipitati**	**CuS precipitati**	**P-4**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	24.98	25.00
Pressure (bar)	1.01	1.04	1.01	1.01
Density (g/L)	1525.30	1013.94	1.43	1016.64
Total Enthalpy (kW-h)	0.00	3555.88	5.85	3559.15
Specific Enthalpy (kcal/kg)	19.07	24.53	110.19	24.38
Heat Capacity (kcal/kg-°C)	0.76	0.98	0.24	0.97
Weight Percent (mass)				
Carb. Dioxide	0.00	0.00	10.60	0.00
Cu2+	0.00	0.00	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.00
H+	0.00	0.00	0.00	0.00
H2CO3	0.00	0.03	0.00	0.93
H2S (aq)	0.00	0.00	0.00	0.01
Hydr. Sulfide	0.00	0.00	89.40	0.00
Me2+	0.00	0.68	0.00	0.67
Na	0.00	0.00	0.00	0.00
Sodium Hydroxid	50.00	0.00	0.00	0.00
Sulfuric Acid	0.00	2.61	0.00	2.59
Water	50.00	96.57	0.00	95.63
Zn2+	0.00	0.11	0.00	0.00
ZnS (s)	0.00	0.00	0.00	0.17
Component Flowrates (kg/h)				
Carb. Dioxide	0.00	0.00	4.84	0.00
Cu2+	0.00	0.17	0.00	0.00
CuS (s)	0.00	0.17	0.00	0.43
H+	0.00	0.17	0.00	4.48

H2CO3	0.00	38.12	0.00	1168.36
H2S (aq)	0.00	0.00	0.00	9.52
Hydr. Sulfide	0.00	0.00	40.84	0.00
Me2+	0.00	847.39	0.00	847.39
Na	0.00	0.00	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	0.00	3252.78	0.00	3252.78
Water	0.00	120462.41	0.00	120134.14
Zn2+	0.00	139.75	0.00	0.22
ZnS (s)	0.00	0.00	0.00	208.02
TOTAL (kg/h)	0.00	124740.97	45.68	125625.35
TOTAL (L/h)	0.00	123026.03	32008.15	123569.07
Stream Name	**S-122**	**S-102**	**S-131**	**S-126**
Source	**CuS precipitati**	**CuS precipitati**	**P-1**	**P-1**
Destination	**P-1**	**P-3**	**MeS precipitati**	**Aeration**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	24.98	25.00	24.98	24.98
Pressure (bar)	1.01	1.01	1.01	1.01
Density (g/L)	1.54	1013.99	1.54	1.54
Total Enthalpy (kW-h)	31.06	3571.43	28.23	2.82
Specific Enthalpy (kcal/kg)	92.59	24.53	92.59	92.59
Heat Capacity (kcal/kg-°C)	0.22	0.98	0.22	0.22
Weight Percent (mass)				
Carb. Dioxide	42.89	0.00	42.89	42.89
Cu2+	0.00	0.00	0.00	0.00
CuS (s)	0.00	0.01	0.00	0.00
H+	0.00	0.00	0.00	0.00
H2CO3	0.00	0.03	0.00	0.00
Hydr. Sulfide	57.11	0.00	57.11	57.11
Me2+	0.00	0.68	0.00	0.00
Sulfuric Acid	0.00	2.61	0.00	0.00
Water	0.00	96.56	0.00	0.00
Zn2+	0.00	0.11	0.00	0.00
Component Flowrates (kg/h)				
Carb. Dioxide	123.77	0.00	112.52	11.25
Cu2+	0.00	0.18	0.00	0.00
CuS (s)	0.00	8.26	0.00	0.00
H+	0.00	0.17	0.00	0.00
H2CO3	0.00	38.29	0.00	0.00
Hydr. Sulfide	164.83	0.00	149.84	14.98
Me2+	0.00	851.09	0.00	0.00

Sulfuric Acid	0.00	3267.00	0.00	0.00
Water	0.00	120988.88	0.00	0.00
Zn2+	0.00	140.36	0.00	0.00
TOTAL (kg/h)	288.60	125294.23	262.36	26.24
TOTAL (L/h)	187110.86	123565.40	170100.78	17010.08
Stream Name	**S-129**	**ZnS**	**S-130**	**S-108**
Source	**P-4**	**P-4**	**P-15**	**P-3**
Destination	**P-15**	**OUTPUT**	**MeS precipitati**	**P-14**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	25.00	25.00
Pressure (bar)	1.01	1.01	1.04	1.01
Density (g/L)	1015.41	1026.61	1015.41	1013.94
Total Enthalpy (kW-h)	3166.94	392.21	3166.98	3555.84
Specific Enthalpy (kcal/kg)	24.41	24.14	24.41	24.53
Heat Capacity (kcal/kg-°C)	0.97	0.96	0.97	0.98
Weight Percent (mass)				
Cu2+	0.00	0.00	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.00
H+	0.00	0.00	0.00	0.00
H2CO3	0.93	0.92	0.93	0.03
H2S (aq)	0.01	0.01	0.01	0.00
Me2+	0.68	0.67	0.68	0.68
Na	0.00	0.00	0.00	0.00
Sulfuric Acid	2.59	2.56	2.59	2.61
Water	95.78	94.39	95.78	96.57
Zn2+	0.00	0.00	0.00	0.11
ZnS (s)	0.00	1.46	0.00	0.00
Component Flowrates (kg/h)				
Cu2+	0.00	0.00	0.00	0.17
CuS (s)	0.01	0.42	0.01	0.17
H+	3.99	0.49	3.99	0.17
H2CO3	1040.02	128.34	1040.02	38.12
H2S (aq)	8.48	1.05	8.48	0.00
Me2+	754.30	93.08	754.30	847.39
Na	0.00	0.00	0.00	0.00
Sulfuric Acid	2895.48	357.30	2895.48	3252.78
Water	106937.92	13196.22	106937.92	120462.41
Zn2+	0.20	0.02	0.20	139.75
ZnS (s)	4.16	203.86	4.16	0.00
TOTAL (kg/h)	111644.56	13980.79	111644.56	124740.97
TOTAL (L/h)	109950.65	13618.42	109950.65	123026.03

Stream Name	**CuS**	**S-112**	**COD waste/products**	**S-104**
Source	**P-3**	**MeOH removal**	**MeOH removal**	**P-11**
Destination	**OUTPUT**	**P-11**	**OUTPUT**	**P-12**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	25.00	25.70
Pressure (bar)	1.01	1.01	1.01	1.01
Density (g/L)	1025.74	998.92	789.61	998.65
Total Enthalpy (kW-h)	15.60	4429.92	0.00	3642.08
Specific Enthalpy (kcal/kg)	24.26	24.87	15.22	25.56
Heat Capacity (kcal/kg-°C)	0.97	0.99	0.61	0.99
Weight Percent (mass)				
Biomass	0.00	0.02	0.00	0.00
Cu2+	0.00	0.00	0.00	0.00
CuS (s)	1.46	0.00	0.00	0.00
H+	0.00	0.03	0.00	0.03
H2CO3	0.03	1.80	0.00	1.80
H2S (aq)	0.00	0.26	0.00	0.26
Me2+	0.67	0.00	0.00	0.00
MeS (s)	0.00	0.00	0.00	0.00
Methanol	0.00	0.00	100.00	0.00
NH4	0.00	0.00	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	2.57	0.00	0.00	0.00
Water	95.16	97.89	0.00	97.91
Zn2+	0.11	0.00	0.00	0.00
ZnS (s)	0.00	0.00	0.00	0.00
Component Flowrates (kg/h)				
Biomass	0.00	33.81	0.00	0.00
Cu2+	0.00	0.00	0.00	0.00
CuS (s)	8.09	0.00	0.00	0.00
H+	0.00	42.03	0.00	33.63
H2CO3	0.17	2758.89	0.00	2207.58
H2S (aq)	0.00	397.01	0.00	317.67
Me2+	3.70	0.00	0.00	0.00
MeS (s)	0.00	0.47	0.00	0.37
Methanol	0.00	0.00	0.00	0.00
NH4	0.00	0.27	0.00	0.22
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	14.22	0.00	0.00	0.00
Water	526.47	150011.76	0.00	120034.60

Zn2+	0.61	0.00	0.00	0.00
ZnS (s)	0.00	0.00	0.00	0.00
TOTAL (kg/h)	553.26	153244.24	0.00	122594.07
TOTAL (L/h)	539.37	153409.40	0.00	122760.01

Stream Name	**Biomass discard**	**S-118**	**S-113**	**S-109**
Source	**P-11**	**P-12**	**P-12**	**MeS precipitati**
Destination	**OUTPUT**	**MeS precipitati**	**Aeration**	**P-7**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.70	25.70	25.70	25.00
Pressure (bar)	1.01	1.01	1.01	1.01
Density (g/L)	998.70	998.65	998.65	1007.20
Total Enthalpy (kW-h)	910.57	3589.86	52.22	6673.01
Specific Enthalpy (kcal/kg)	25.56	25.56	25.56	24.71
Heat Capacity (kcal/kg-°C)	0.99	0.99	0.99	0.98
Weight Percent (mass)				
Biomass	0.11	0.00	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.00
H+	0.03	0.03	0.03	0.03
H2CO3	1.80	1.80	1.80	1.13
H2S (aq)	0.26	0.26	0.26	0.07
MeS (s)	0.00	0.00	0.00	0.50
NH4	0.00	0.00	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	0.00	0.00	0.00	1.25
Water	97.80	97.91	97.91	97.02
ZnS (s)	0.00	0.00	0.00	0.00
Component Flowrates (kg/h)				
Biomass	33.81	0.00	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.01
H+	8.40	33.15	0.48	63.05
H2CO3	551.32	2175.93	31.65	2637.08
H2S (aq)	79.33	313.12	4.55	156.51
MeS (s)	0.09	0.37	0.01	1166.70
NH4	0.05	0.22	0.00	0.22
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	0.00	0.00	0.00	2895.48
Water	29977.16	118313.67	1720.93	225419.72
ZnS (s)	0.00	0.00	0.00	4.46
TOTAL (kg/h)	30650.16	120836.44	1757.63	232343.22

TOTAL (L/h)	30689.99	121000.00	1760.01	230682.94
Stream Name	**S-111**	**MeS**	**S-134**	**General MeS**
Source	**P-7**	**P-7**	**P-2**	**P-2**
Destination	**P-2**	**OUTPUT**	**P-16**	**OUTPUT**
Stream Properties				
Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.00	25.00	25.00
Pressure (bar)	1.01	1.01	1.01	1.01
Density (g/L)	1003.50	1014.66	1003.38	1014.66
Total Enthalpy (kW-h)	4459.42	2213.60	4415.14	44.27
Specific Enthalpy (kcal/kg)	24.80	24.53	24.80	24.53
Heat Capacity (kcal/kg-°C)	0.99	0.98	0.99	0.98
Weight Percent (mass)				
CuS (s)	0.00	0.00	0.00	0.00
H+	0.03	0.03	0.03	0.03
H2CO3	1.14	1.12	1.14	1.12
H2S (aq)	0.07	0.07	0.07	0.07
MeS (s)	0.02	1.47	0.00	1.47
NH4	0.00	0.00	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	1.25	1.23	1.25	1.23
Water	97.50	96.07	97.51	96.07
ZnS (s)	0.00	0.01	0.00	0.01
Component Flowrates (kg/h)				
CuS (s)	0.00	0.01	0.00	0.00
H+	42.19	20.86	41.77	0.42
H2CO3	1764.53	872.55	1747.08	17.45
H2S (aq)	104.73	51.79	103.69	1.04
MeS (s)	23.33	1143.36	0.47	22.87
NH4	0.14	0.07	0.14	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfuric Acid	1937.43	958.05	1918.27	19.16
Water	150833.26	74586.46	149341.53	1491.73
ZnS (s)	0.09	4.37	0.00	0.09
TOTAL (kg/h)	154705.70	77637.53	153152.95	1552.75
TOTAL (L/h)	154166.82	76516.12	152636.50	1530.32
Stream Name	**Air**	**S-124**	**S-114**	**Effluent water**
Source	**INPUT**	**Aeration**	**Aeration**	**Ultra filtratio**
Destination	**Aeration**	**OUTPUT**	**Ultra filtratio**	**OUTPUT**
Stream Properties				

Activity (U/ml)	0.00	0.00	0.00	0.00
Temperature (°C)	25.00	25.54	25.54	27.03
Pressure (bar)	1.01	1.01	1.01	1.01
Density (g/L)	1.18	1.21	1004.14	998.87
Total Enthalpy (kW-h)	0.70	1.46	52.45	54.37
Specific Enthalpy (kcal/kg)	6.05	12.34	25.33	26.92
Heat Capacity (kcal/kg-°C)	0.24	0.24	0.99	0.99
Weight Percent (mass)				
Carb. Dioxide	0.00	11.02	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.00
H+	0.00	0.00	0.03	0.03
H2CO3	0.00	0.00	1.78	1.79
MeS (s)	0.00	0.00	0.00	0.00
NH4	0.00	0.00	0.00	0.00
Nitrogen	76.71	75.15	0.00	0.00
Oxygen	23.29	13.83	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfur	0.00	0.00	1.03	0.00
Water	0.00	0.00	97.16	98.18
ZnS (s)	0.00	0.00	0.00	0.00
Component Flowrates (kg/h)				
Carb. Dioxide	0.00	11.25	0.00	0.00
CuS (s)	0.00	0.00	0.00	0.00
H+	0.00	0.00	0.48	0.48
H2CO3	0.00	0.00	31.65	31.19
MeS (s)	0.00	0.00	0.01	0.00
NH4	0.00	0.00	0.00	0.00
Nitrogen	76.71	76.71	0.00	0.00
Oxygen	23.29	14.12	0.00	0.00
Sodium Hydroxid	0.00	0.00	0.00	0.00
Sulfur	0.00	0.00	18.38	0.00
Water	0.00	0.00	1731.26	1706.26
ZnS (s)	0.00	0.00	0.00	0.00
TOTAL (kg/h)	100.00	102.08	1781.79	1737.94
TOTAL (L/h)	84801.70	84196.06	1774.44	1739.91
Stream Name	**Retentate**			
Source	**Ultra filtratio**			
Destination	**OUTPUT**			
Stream Properties				
Activity (U/ml)	0.00			
Temperature (°C)	27.03			
Pressure (bar)	1.01			

Density (g/L)	1234.98
Total Enthalpy (kW-h)	1.13
Specific Enthalpy (kcal/kg)	22.11
Heat Capacity (kcal/kg-°C)	0.82
Weight Percent (mass)	
CuS (s)	0.00
H+	0.02
H2CO3	1.04
MeS (s)	0.01
NH4	0.00
Sulfur	41.92
Water	57.01
ZnS (s)	0.00
Component Flowrates (kg/h)	
CuS (s)	0.00
H+	0.01
H2CO3	0.46
MeS (s)	0.01
NH4	0.00
Sulfur	18.38
Water	25.00
ZnS (s)	0.00
TOTAL (kg/h)	43.85
TOTAL (L/h)	35.51

4. OVERALL COMPONENT BALANCE (kg/yr)

COMPONENT	**IN**	**OUT**	**IN-OUT**
Biomass	0	267753	- 267753
Carb. Dioxide	0	89115	- 89115
Cu2+	44849	6	44843
CuS (s)	0	67468	- 67468
H+	76859	242769	- 165910
H2CO3	0	12684000	- 12684000
H2S (aq)	0	1054971	- 1054971
Me2+	6740631	766539	5974093
MeS (s)	0	9237334	- 9237334
Methanol	6966586	6	6966580
Na	0	4	- 4
NH4	39327	1034	38293
Nitrogen	607557	607557	0
Oxygen	184443	111798	72645
Sodium Hydroxid	7	0	7
Sulfur	0	145777	- 145777

Sulfuric Acid	25874640	10681960	15192680
Water	958320007	962370979	- 4050972
Zn2+	1111651	5032	1106619
ZnS (s)	0	1649841	- 1649841
TOTAL	999966558	999983943	17384

Overall Error: 0.002%

Appendix A.4 – SuperPro “Equipment report”

Equipment Report *for Case 1- MeOH opt- 0.67 RISE PARTNER data*

1. EQUIPMENT SUMMARY (2018 prices)

Name	**Type**	**Units**	**Standby/ Staggered**	**Size (Capacity)**	**Unit**	**Material of Construction**	**Purchase Cost ($/Unit)**
R-101	Stirred Reactor	18	0/0	96,09	m3	SS316	1196000
R-102	Stirred Reactor	18	0/0	95,69	m3	SS316	1194000
FR-101	Fermentor	6	0/0	301496,09	L	SS316	2564000
R-103	Stirred Reactor	33	0/0	97,98	m3	SS316	1205000
FSP-101	Flow Splitter	1	0/0	803,02	kg/h	CS	0
IPCL-101	IP Clarifier	1	0/0	5,45	m2	CS	56000
IPCL-102	IP Clarifier	1	0/0	6,60	m2	CS	57000
IPCL-103	IP Clarifier	1	0/0	12,33	m2	CS	60000
MF-102	Microfilter	77	0/0	79,69	m2	SS316	124000
FSP-102	Flow Splitter	1	0/0	122594,07	kg/h	CS	0
PM-101	Centrifugal Pump	1	0/0	0,14	kW	SS316	9000
PM-102	Centrifugal Pump	1	0/0	0,14	kW	SS316	9000
PM-103	Centrifugal Pump	1	0/0	0,13	kW	SS316	9000
PM-104	Centrifugal Pump	1	0/0	0,18	kW	SS316	9000
G-101	Centrifugal Compressor	1	0/0	70,05	kW	CS	81000
RO-101	Reverse Osmosis Filter	2	0/0	47,10	m2	SS316	15000
R-105	Stirred Reactor	1	0/0	1955,56	L	SS316	539000
PM-105	Centrifugal Pump	1	0/0	0,00	kW	SS316	9000
FSP-103	Flow Splitter	1	0/0	288,60	kg/h	CS	0
GBX-101	Generic Box	1	0/0	153244,24	kg/h	CS	0
IPCL-104	IP Clarifier	1	0/0	8,32	m2	CS	58000

2. ITEMIZED EQUIPMENT LIST

R-101 (Stirred Reactor)		
Equipment size was calculated		
Number of Units	18,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,30	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Stirred Reactor)	1196000,00	$/unit
Max Volume	100,00	m3
Min Working/Vessel Volume	0,00	%
Max Working/Vessel Volume	80,00	%
Volume	96,09	m3
Height	9,14	m
Design Pressure	1,52	bar
Vessel is constructed according to ASME standards		
Diameter	3,66	m
R-102 (Stirred Reactor)		
Equipment size was calculated		
Number of Units	18,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,30	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Stirred Reactor)	1194000,00	$/unit
Max Volume	100,00	m3
Min Working/Vessel Volume	0,00	%
Max Working/Vessel Volume	80,00	%
Volume	95,69	m3
Height	9,13	m
Design Pressure	1,52	bar
Vessel is constructed according to ASME standards		
Diameter	3,65	m
FR-101 (Fermentor)		
Equipment size was calculated		

Number of Units	6,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,30	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Fermentor)	2564000,00	$/unit
Max Volume	350000,00	L
Min Working/Vessel Volume	0,00	%
Max Working/Vessel Volume	90,00	%
Volume	301496,09	L
Height	15,12	m
Design Pressure	1,52	bar
Vessel is constructed according to ASME standards		
Diameter	5,04	m
R-103 (Stirred Reactor)		
Equipment size was calculated		
Number of Units	33,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,30	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Stirred Reactor)	1205000,00	$/unit
Max Volume	100,00	m3
Min Working/Vessel Volume	0,00	%
Max Working/Vessel Volume	80,00	%
Volume	97,98	m3
Height	9,20	m
Design Pressure	1,52	bar
Vessel is constructed according to ASME standards		
Diameter	3,68	m
FSP-101 (Flow Splitter)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	

Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for Flow Splitter)	0,00	$/unit
Rated Throughput	803,02	kg/h
IPCL-101 (IP Clarifier)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,20	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for IP Clarifier)	56000,00	$/unit
Flow Type = Down-Flow		
Installation is above ground		
No of Plates Per Unit	19,00	
Plate Inclination	55,00	degrees
Plate Spacing	10,00	cm
Plate Width	0,50	m
Plate Length	1,00	m
Horizontal Area (all plates)	5,45	m2
IPCL-102 (IP Clarifier)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,20	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for IP Clarifier)	57000,00	$/unit
Flow Type = Down-Flow		
Installation is above ground		

No of Plates Per Unit	23,00	
Plate Inclination	55,00	degrees
Plate Spacing	10,00	cm
Plate Width	0,50	m
Plate Length	1,00	m
Horizontal Area (all plates)	6,60	m2
IPCL-103 (IP Clarifier)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,20	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for IP Clarifier)	60000,00	$/unit
Flow Type = Down-Flow		
Installation is above ground		
No of Plates Per Unit	43,00	
Plate Inclination	55,00	degrees
Plate Spacing	10,00	cm
Plate Width	0,50	m
Plate Length	1,00	m
Horizontal Area (all plates)	12,33	m2
MF-102 (Microfilter)		
Equipment size was calculated		
Number of Units	77,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Microfilter)	124000,00	$/unit
Unit Cost of Consumable: Dft Membrane	400,00	$/m2
Disposal Cost of Consumable: Dft Membrane	0,00	$/m2
Number of Available Cartridge Slots	1,00	
FSP-102 (Flow Splitter)		
Equipment size was calculated		

Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for Flow Splitter)	0,00	$/unit
Rated Throughput	122594,07	kg/h
PM-101 (Centrifugal Pump)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Centrifugal Pump)	8500,00	$/unit
Power	0,14	kW
PM-102 (Centrifugal Pump)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Centrifugal Pump)	8500,00	$/unit
Power	0,14	kW
PM-103 (Centrifugal Pump)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	

Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Centrifugal Pump)	8500,00	$/unit
Power	0,13	kW
PM-104 (Centrifugal Pump)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Centrifugal Pump)	8500,00	$/unit
Power	0,18	kW
G-101 (Centrifugal Compressor)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for Centrifugal Compressor)	81000,00	$/unit
Power	70,05	kW
RO-101 (Reverse Osmosis Filter)		
Equipment size was calculated		
Number of Units	2,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,40	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h

Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Reverse Osmosis Filter)	15000,00	$/unit
Unit Cost of Consumable: UF Membrane (Biotech)	981,11	$/m2
Disposal Cost of Consumable: UF Membrane (Biotech)	0,00	$/m2
Number of Available Cartridge Slots	1,00	
R-105 (Stirred Reactor)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,30	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Stirred Reactor)	539000,00	$/unit
Max Volume	40000,00	L
Min Working/Vessel Volume	0,00	%
Max Working/Vessel Volume	90,00	%
Volume	1955,56	L
Height	2,50	m
Design Pressure	1,52	bar
Vessel is constructed according to ASME standards		
Diameter	1,00	m
PM-105 (Centrifugal Pump)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		SS316
Purchase Cost (system model for Centrifugal Pump)	8500,00	$/unit
Power	0,00	kW
FSP-103 (Flow Splitter)		

Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for Flow Splitter)	0,00	$/unit
Rated Throughput	288,60	kg/h
GBX-101 (Generic Box)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,50	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for Generic Box)	0,00	$/unit
Rated Throughput	153244,24	kg/h
IPCL-104 (IP Clarifier)		
Equipment size was calculated		
Number of Units	1,00	
Number of Standby Units	0,00	
Number of Staggered Units	0,00	
Installation Factor	0,20	
Maintenance Factor	0,10	
Cost Allocation Factor	1,00	
Usage Rate	100,00	$/equipment-h
Availability Rate	100,00	$/h
Material of Construction		CS
Purchase Cost (system model for IP Clarifier)	58000,00	$/unit
Flow Type = Down-Flow		
Installation is above ground		
No of Plates Per Unit	29,00	
Plate Inclination	55,00	degrees
Plate Spacing	10,00	cm
Plate Width	0,50	m

Plate Length			1,00	m	
Horizontal Area (all plates)			8,32	m2	

3. CIP SKID LIST

No CIP skids are present in the flowsheet.

4. SIP PANEL LIST

No SIP panels are present in the flowsheet.

5. EQUIPMENT CONSUMABLES

MF-102 (Microfilter)

Name	**Consumption Rate**	**Amount per Use**		**Replac. Frequency**	
Dft Membrane	N/A	6136,38	m2	1000,00	hrs

RO-101 (Reverse Osmosis Filter)

Name	**Consumption Rate**	**Amount per Use**		**Replac. Frequency**	
UF Membrane (Biotech)	N/A	94,19	m2	1,50	yrs

Appendix A.5 – Matlab code, case 1 scenario 2

Below the script defining the model is stated.

```
function [ dy ] = molL( t,y,par)

%Volumetric flows, based on the SuperPro numbers from case 1 opt.

%OBS! literature and SuperPro data has been used in combination.

%% STATE VARIABLES
%==========================================================================
==
%BIOREACTOR
S_Sred = y(1);           %Reduced sulfur, total or
S_MeOH = y(2);           %MeOH
S_SO4 = y(3);            %Sulfate
X_mSRB = y(4);           %MeOH biomass
S_HS = y(5);             %Dissolved H2S in HS form
S_IC = y(6);             %Inorganic carbon in liquid phase
S_HCO3 = y(7);           %Dissolved CO2 in HCO3 form
S_H2S_g = y(8);          %H2S in gas phase
S_CO2_g = y(9);          %CO2 in gas phase

% TANK 1
S_HS15 = y(10);          %Dissolved H2S in HS form pH 1.5
S_HCO315 = y(11);        %Dissolved CO2 in HCO3 form pH 1.5
S_Cu = y(12);            %Produced CuS

% TANK 2
S_HS45 = y(13);          %Dissolved H2S in HS form pH 4.5
S_HCO345 = y(14);        %Dissolved CO2 in HCO3 form pH 1.5
S_Zn = y(15);            %Produced ZnS

% TANK 3
S_HS72 = y(16);          %Dissolved H2S in HS form pH 7.2
S_HCO372 = y(17);        %Dissolved CO2 in HCO3 form pH 7.2
S_Me = y(18);            %Produced MeS

%%
===========================================================================
==
% Flow rates and relationships (continuous mode, connection between tanks)
%==========================================================================
==

% VOLUMETRIC FLOWS
qp1 = 4.5e-03*par.qflow; %Product outflow from precipitation tank 1 [L/h]
qp2 = 1.1e-01*par.qflow; %Product outflow from precipitation tank 2 [L/h]
qp3 = 3.3e-01*par.qflow; %Product outflow from precipitation tank 3 [L/h]

qin1 = par.qflow;                  %Inflow to precip. tank 1 [L/h]
qin2 = qin1 - qp1;                 %Inflow to precip. tank 2 [L/h]
qH2O_in3 =  121000;                %Recycling to precip. tank 3 [L/h]
                                   According to SuperPro script.
qin3 = qin2 - qp2 + qH2O_in3;      %Inflow to precip. tank 3 [L/h]
q_bin = qin3 - qp3 + par.qs_vol;         % Total flow rate into the
                                         bioreactor [L/h]
```

```
q_bout = q_bin - qH2O_in3;                % Outflow from the bioreactor
[L/h]

%% BIOREACTOR
%===========================================================================
==

%Inhibition terms (bioreactor)
%===========================================================================
==

S_H = 10^-(par.pHbio); % (mol/L)

%===========================================================================
% High and low pH inhibition
%===========================================================================
% %MeOH utilizing bacteria
% if (par.pH_LL_mSRB < par.pHbio < par.pH_UL_mSRB)
% I_pH_mSRB = 1;
% else
% I_pH_mSRB = ((1 + 2 * 10^(0.5 * (par.pH_LL_mSRB - par.pH_UL_mSRB)))/(1 +
10^(par.pHbio - par.pH_UL_mSRB) + 10^(par.pH_LL_mSRB - par.pHbio)));
%Inhibition term from pH with both high and low inhibition
% end

%===========================================================================
==
% Low pH inhibition
%===========================================================================
==

if(par.pHbio < par.pH_UL_mSRB) %Inhibition term for mSRB
I_pH_mSRB = exp(- 3 * ((par.pHbio - par.pH_UL_mSRB)/(par.pH_UL_mSRB -
par.pH_LL_mSRB))^2);
else
 I_pH_mSRB = 1;
end

%===========================================================================
==
%H2S inhibition (different suggested terms)
%===========================================================================
==

%Non-competetive inhibition (Provad, fungerar)
%I_H2S = 1/(1 + (S_Sred/par.KI(1)));

%Inhibition for high H2S concentration
%
I_H2S = exp(-(S_Sred/0.60056 * par.KI(1))^2);

%===========================================================================
==
% Rates for biomass production, substrate consumption and bacterial decay
%===========================================================================
==
```

```
r_MeOH = (par.umax_MeOH(2)/(par.Y_mSO4/0.75)) * S_MeOH/(par.Ks_MeOH +
S_MeOH) * S_SO4/(par.Ks_SO4 + S_SO4)...
    * X_mSRB * I_pH_mSRB * I_H2S;     %Rate for biomass production for
mSRB.
                                       [mol/L*h]

r_SO4 = r_MeOH*0.75;    %Rate for biomass production for mSRB, based on
                        stoichiometry of the SO4 reduction balance.
                  [mol/L*h]

r_dec_mSRB = par.k_dec_mSRB * X_mSRB;        %Rate of decay of mSRB
[mol/L*h]

%===========================================================================
==
% Acid base reaction equilibrium - BIOREACTOR
%===========================================================================
==
S_H2S = S_Sred-S_HS;                        % H2S = [S--] - [HS-]
[mol/L]
S_CO2 = S_IC-S_HCO3;

rho_A_HS = par.K_ab_H2S * (S_HS* (S_H + par.K_a_H2S) - par.K_a_H2S *
S_Sred);
                                          %Dissociation to HS-
                                    [mol^3/L^3*h]

rho_A_HCO3 = par.K_ab_CO2 * (S_HCO3 * (S_H+ par.K_a_CO2) - par.K_a_CO2 *
S_IC);                                   %Dissociation to HCO3-
[mol^3/L^3*h]

%===========================================================================
==
% Mass transport for gases - bioreactor
%===========================================================================
==
p_H2S_gas = S_H2S_g*par.R*par.T ;         %Partial pressure of H2S [atm]
p_CO2_gas = S_CO2_g*par.R*par.T;          %Partial pressure of CO2 [atm]

q_gas = (par.N2g/(1-(p_H2S_gas/1)-(p_CO2_gas/1)));          %[mol/L*atm]

S_prim_H2S = p_H2S_gas/par.H_H2S; %Henry's law, maximum solubility of H2S
in
                                   liquid [mol/L]

S_prim_CO2 = p_CO2_gas/par.H_CO2; %Henry's law, maximum solubiliy of CO2 in
                                   liquid [mol/L]

r_H2St = par.kLa_H2S *(S_H2S-S_prim_H2S); %Mass transport for H2S gas,
                                           from liquid to gas. [mol/L*h]
r_CO2t = par.kLa_CO2 *(S_CO2-S_prim_CO2); %Mass transport for CO2 gas,
                                           from liquid to gas. [mol/L*h]

%===========================================================================
==
% Dynamic mass balances - bioreactor
%===========================================================================
==
```

```
%Biomass
dX_mSRB_dt = q_bin/par.Vliq_bio*(par.X_mSRB_in - X_mSRB)...
    + par.Y_mSO4 * r_SO4 - r_dec_mSRB;
                                    %Biomass production/consumption
                              [mol/L*h]

%Substrate (electron donor) uptake
dS_MeOH_dt = q_bin/par.Vliq_bio*(par.S_MeOH_in - S_MeOH) - r_MeOH;
                                    %Methanol consumption [mol/L*h]

dS_SO4_dt = q_bin/par.Vliq_bio*(par.S_SO4_in - S_SO4) - r_SO4;
                                    %SO4 consumption [mol/L*h]


%Product formation (inorganic carbon)
dS_IC_dt = q_bin/par.Vliq_bio * (par.S_IC_in - S_IC) - r_CO2t;
                                                %Inorganic carbon [mol/L*h]

%Acid-base
d_S_HS_dt = -rho_A_HS;           %Dissociation of H2S to HS- [mol^3/L^3*h]
d_S_HCO3_dt = -rho_A_HCO3;       %Dissociation of CO2 tp HCO3-
[mol^3/L^3*h]

%Gas phase
dS_H2S_g_dt = 0 -
(q_gas/par.V_gas)*S_H2S_g+r_H2St*(par.Vliq_bio/par.V_gas);
%Gas phase for H2S with compensation for volume difference [mol/L*h]

dS_CO2_g_dt = 0 -
(q_gas/par.V_gas)*S_CO2_g+r_CO2t*(par.Vliq_bio/par.V_gas);
%Gas phase for CO2 with compensation for volume difference [mol/L*h]

%%
% PRECIPITATION REACTIONS
%=============================================================================
==

%% TANK 1

% Acid base expressions
S_H15 = (10^-par.pH15);                              % [H+] at pH 1.5
[mol/L]
S_Hin1 = S_H15 + S_H/3;                              % [mol/L]

S_Sred1 = S_Sred/3;

S_H2S15 = S_Sred1-S_HS15;          % H2S = [S--] - [HS-] at pH 1.5 [mol/L]
S_CO215 = S_IC/3-S_HCO315;         % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St15 = par.kLa_H2S * (S_prim_H2S-S_H2S15); % [mol/L*h]

% Acid base equilibrium rate expression
rho_A_HS15 = par.K_ab_H2S * (S_HS15 * (S_Hin1 + par.K_a_H2S) - par.K_a_H2S*
S_Sred1);
            %Dissociation to HS- in the first precipitation tank [mol/L*h]

rho_A_HCO315 = par.K_ab_CO2 * (S_HCO315 * (S_Hin1 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
```

```
            %Dissociation to HCO3- in the first precipitation tank
            [mol/L*h]

% Dissociation rate in precipitation tanks
d_S_HS_dt15 = -rho_A_HS15;          %Dissociation of H2S to HS- in
                                     precipitation tank 1 [mol^3/L^3*h]
d_S_HCO3_dt15 = -rho_A_HCO315;      %Dissociation of CO2 to HCO3- in
                                    precipitation tank 1 [mol^3/L^3*h]

% [Cu2+] conditioned >0
% Precipitation mass balance and rate, conditioned to be >0

if S_Cu >= 0
    S_Cu = par.Cu_in - S_HS15;        % Concentration Cu [mol/L]
    rho_CuS = par.k_Cu*(S_Cu+S_HS15); % Precipitation rate equation
[mol/L*h]
    d_Cu_dt = qin1/par.Vliq_tank1*(par.Cu_in - S_Cu) - rho_CuS * par.YCuS;
    %According to Tokuda et al. (2008) [mol/L*h]% [mol/L]
else
    S_Cu = 0;                                   % Concentration Cu [mol/L]
    d_Cu_dt = 0;
    rho_CuS = par.k_Cu*(S_Cu+S_Sred1);          % Precipitation rate
equation [mol/L*h]
end


%% TANK 2
% Acid base expressions
S_H45 = (10^-par.pH45);                         % [H+] at pH 4.5 [mol/L]
S_Hin2 = qin1/qin2*S_H15 + S_H45 + S_H/3;       % [mol/L]

S_Sred2 = S_Sred/3; % [mol/L]

S_H2S45 = S_Sred2-S_HS45;           % H2S = [S--] - [HS-] at pH 4.5 [mol/L]
S_CO245 = S_IC/3-S_HCO345;          % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St45 = par.kLa_H2S * (S_H2S45-S_prim_H2S);                   % [mol/L*h]

% Acid base equilibrium rate expression
rho_A_HS45 = par.K_ab_H2S * (S_HS45 * (S_Hin2 + par.K_a_H2S) - par.K_a_H2S
* S_Sred2);
      %Dissociation to HS- in the second precipitation tank   [mol^3/L^3*h]

rho_A_HCO345 = par.K_ab_CO2 * (S_HCO345 * (S_Hin2 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
      %Dissociation to HCO3- in the second precipitation tank
[mol^3/L^3*h]

% Dissociation rate in precipitation tanks
d_S_HS_dt45 = -rho_A_HS45;                  %Dissociation of H2S to HS- in
                                             precipitation tank 2 [mol/L*h]
d_S_HCO3_dt45 = -rho_A_HCO345;              %Dissociation of CO2 to HCO3- in
                                             precipitation tank 2 [mol/L*h]

% [Zn2+] is conditioned to be >=0 mol/L
% Precipitation mass balances, conditioned to be >0

if S_Zn >= 0
```

```
    S_Zn = par.Zn_in - S_HS45;              % Concentration Zn [mol/L]
    rho_ZnS = par.k_Zn*(S_Zn+S_HS45);       % Precipitation rate equation
                                              [mol/L*h]
    d_Zn_dt = qin2/par.Vliq_tank2*(par.Zn_in - S_Zn)- rho_ZnS * par.YZnS;
    %According to Tokuda et al. (2008) [mol/L*h]
else
    S_Zn = 0;                               % Concentration Zn [mol/L]
    rho_ZnS = par.k_Zn*(S_Zn+S_Sred2);      % Precipitation rate equation
                                              [mol/L*h]
    d_Zn_dt = 0;
end

%% TANK 3
% Acid base expressions
S_H72 = (10^-par.pH72);                           % [H+] at pH 7.2 ([mol/L]
S_Hin3 = qin2/qin3*S_H45 + S_H/3 + S_H72;         % [mol/L]

S_Sred3 = S_Sred/3; % [mol/L]

S_H2S72 = S_Sred3-S_HS72;                     % H2S = [S--] - [HS-] at pH 7.2
                                              [mol/L]
S_CO272 = S_IC/3-S_HCO372;                    % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St72 = par.kLa_H2S * (S_H2S72-S_prim_H2S);                    % [mol/L*h]

% Acid base equilibrium rate expression
rho_A_HS72 = par.K_ab_H2S * (S_HS72 * (S_Hin3 + par.K_a_H2S) - par.K_a_H2S
* S_Sred3);
            % Dissociation to HS- in the third precipitation tank.
      [mol/L*h]

rho_A_HCO372 = par.K_ab_CO2 * (S_HCO372 * (S_Hin3 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
            %Dissociation to HCO3- in the third precipitation tank.
      [mol/L*h]

% Dissociation rate in precipitation tanks

d_S_HS_dt72 = -rho_A_HS72;
            %Dissociation of H2S to HS- in precipitation tank 3.
             [mol^3/L^3*h]
d_S_HCO3_dt72 = -rho_A_HCO372;
            %Dissociation of CO2 to HCO3- in precipitation tank 3.
             [mol^3/L^3*h]



% [Me2+] is conditioned to be >=0 mol/L
% Precipitation mass balances, conditioned to be >0

if S_Me >= 0
    S_Me = par.Me_in - S_HS72;                    % Concentration Me [mol/L]
    rho_MeS = par.k_Zn*(S_Me+S_HS72);             % Precipitation rate
equation
                                                    [mol/L*h]
    d_Me_dt = qin3/par.Vliq_tank3*(par.Me_in - S_Me) - rho_MeS * par.YMeS;
    %MeS precipitation is based on ZnS because it is more abundant.
    [mol/L*h]
    %According to Tokuda et al. (2008)
```

```
else
    S_Me = 0;                                       % Concentration Me [mol/L]
    rho_MeS = par.k_Zn*(S_Me+S_Sred3);              % Precipitation rate
equation
                                                      [mol/L*h]
    d_Me_dt = 0;
end

% Product formation with respect to the precipitation
if S_Sred >=0
    dS_S_red_dt = q_bin/par.Vliq_bio*(par.S_red_in - S_Sred) + (1-
par.Y_mSO4) * r_SO4 - r_H2St...
    - rho_CuS * par.YCuS - rho_ZnS * par.YZnS - rho_MeS * par.YMeS;
%Reduced sulfur = [S--]+[HS-]+[H2S] [mol/L*h]
else
      dS_S_red_dt = q_bin/par.Vliq_bio*(par.S_red_in - S_Sred) + (1-
par.Y_mSO4) * r_SO4 - r_H2St;
end
%===========================================================================
==

dy =
[dS_S_red_dt;dS_MeOH_dt;dS_SO4_dt;dX_mSRB_dt;d_S_HS_dt;dS_IC_dt;d_S_HCO3_dt
;dS_H2S_g_dt;dS_CO2_g_dt;...

d_S_HS_dt15;d_S_HCO3_dt15;d_Cu_dt;d_S_HS_dt45;d_S_HCO3_dt45;d_Zn_dt;d_S_HS_
dt72;d_S_HCO3_dt72;d_Me_dt];

end
function [par] = molLpar
```

Below is the .m-file containing the process parameters.

```
%OBS! Somewhat modified data. Tidigare litteratur data används tillsammans
med SuperPro data.

%===========================================================================
==
% FLOW PARAMETERS - Overall process
%===========================================================================
==
par.qflow = 123547.3322;                  %VOLUMETRIC BASE FLOW INTO THE
                                           PROCESS (IE. LEACHATE FLOW)[L/h]
par.qs_vol = 1126.5494;                   % VOLUMETRIC BASE FLOW OF
SUBSTRATE
                                           [L/h]
par.q_g_vol = 506.8165e03;                % VOLUMETRIC gas flow from the
                                           bioreactor [L/h]


par.q_in = 6766673.6064/par.qflow;        %Flow rate of leachate into the
                                           precipitation [mol/L]
par.qs = 37354.55/par.qs_vol;             %Flow rate of substrate into the
                                           bioreactor  [mol/L]

par.qp = 0.001*par.q_in;
par.q_g = 20717.2/par.q_g_vol;            %Flow rate gas phase out from the
                                           bioreactor [mol/L]
```

```
%===========================================================================
==
% Coefficients for: MeOH degrading SRB: mSRB // Acetate degrading SRB: aSRB
%===========================================================================
==
par.umax_MeOH = [0.013 0.612];      %Maximum specific growth rate of eSRB,
                                     0.013 [h^-1](Nagpal)
                                     0.612 (d^-1) (Barrera)
par.Y_mSO4 = 0.5/24;            %Yield for biomass on sulfate, mSRB, YSO4X

                        (Nagpal)[gprotein/molSO4/96.6gSO4/mol]=[g/g]
par.Y_MmSRB = 0.5*par.Y_mSO4;   %(Nagpal)  Yield of mSRB biomass on MeOH
par.Ks_MeOH = 0.0045;           %Ks for MeOH is approximated to be equal to
                                 that of EtOH [mol/l] (Nagpal)
par.Ks_SO4 = 0.0085;            %Ks for SO4 [mol/l] (Nagpal) 0.297*10^-3
                                 [mol/l] (Barrera)

par.umax_Ac = 0.063;             %Maximum specific growth rate of
aSRB,0.063
                                 [h^-1](Moosa)
par.Y_AaSRB = 0.58;              %Yield for Acetate on biomass, YACX,
                                 [g bacteria/g acetate] (Moosa)
par.Ks_Ac = 0.0035;              %Ks for Acetate, Bara provat värde 24*10^-
3

                                 [mol/l] (Barrera)

%===========================================================================
==
% Coefficients for: BIOREACTOR INGREDIENT FEEDS/FLOWS
%===========================================================================
==
par.X_mSRB_in = 0.00000002*par.q_in;          %Inflow of mSRB biomass
[mol/L]
par.X_aSRB_in = 0.00000002*par.q_in;          %Inflow of aSRB biomass
[mol/L]
par.S_MeOH_in = 27452.07570/153763;           %Substrate MeOH concentration
                                               inflow [mol/L] from SuperPro
                                          -

                                               RISE PARTNER + CASE 1
par.S_Ac_in = 0/153763;              %CASE 1 AND 4: Acetate concentration
                                      inflow [mol/L] 27452.08/par.qs_vol
par.S_SO4_in = 19559.59/153763;      %Sulfate concentration inflow [mol/L]
                                      from SuperPro  - RISE PARTNER + CASE1

par.Y_aSO4 = 0.5/24;                 %(Nagpal) Yield for sulfate on
biomass,

                                      aSRB [gprotein/molSO4/96.6gSO4/mol]
par.S_red_in = 0.00000000001*par.q_in*2;        %Reduced sulfur
concentration

                                                 inflow [mol/L]
par.S_IC_in = 0.00000000001*par.q_in*2;         %Inorganic carbon
                                                 concentration inflow
                                          [mol/L]

par.k_dec_mSRB = 0.00;                 %Decay coefficient for MeOH SRB [h^-
1]
par.k_dec_aSRB = 0.00;                 %Decay coefficient for acetate SRB.
                                       (Moosa et al. 2002) [h^-1]
```

```
%=========================================================================
==
% Mass transfer gas-liquid parameters: BIOREACTOR
%=========================================================================
==
par.kLa_H2S = 10;                  %kLa mass transfer coefficient for H2S
                                   [h^-1] (Nagpal)
par.kLa_CO2 = 10;                  %kLa mass transfer coefficient for CO2
                                    [h^-1]
par.H_H2S = 10.70;                 %Henrys constant for H2S [atmL/mol]
10.70;
                                   (Nagpal)
par.H_CO2 = 25.88;                 %Henrys constant for CO2 [atmL/mol]
25.88;
                                   (Nagpal)
par.T = 298;                       %Temperature [K]
par.R = 0.082057338;               %Gas constant [L atm K^-1 mol^-1]
par.V_liq = 343288;                %Liquid volume [L] (from SuperPro)
par.C_gas = 0.0409;                %Gas concentration in outgoing flow from
                                    the reactor [mol/L]

par.P_tot = 1;                     %Total pressure from all gases [atm]
par.N2g = 435244;                  %Stripping gas, Flow of N2 [L/h]

par.density_N2 = 1.2151;           %Density for N2[g/L]
par.mol_N2 =28;                    %Molar weight for N2[g/mol]
%=========================================================================
==
% BIOREACTOR: Acid-base coefficients
%=========================================================================
==
par.K_ab_H2S = 10^15;              %Acid base kinetic parameter for H2S
par.K_a_H2S = 1.075 * 10^-7;       %Acid-base equilibirum coefficient for
H2S

par.K_a_CO2 = 4.97 * 10^-7;        %Acid-base equilibirum coefficient for
                                    HCO3
par.K_ab_CO2 = 10^12;              %Acid base kinetic parameter for HCO3

%=========================================================================
==
% Inhibition parameters - BIOREACTOR
%=========================================================================
==
par.pH_LL_mSRB = 6;               %Lower pH limit where the group of
                                   organisms (eSRB) are 50% inhibited
par.pH_UL_mSRB = 7.5;             %Upper pH limit where the group of
                                   organisms (eSRB) are 50% inhibited
par.pH_LL_aSRB = 6;               %Lower pH limit where the group of
                                   organisms (aSRB) are 50% inhibited
par.pH_UL_aSRB = 7.5;             %Upper pH limit where the group of
                                   organisms (aSRB) are 50% inhibited
par.pHbio = 7;                    %pH for the pH inhibition term

par.KI = [5.13*10^-3 0.003875 1.54];      %KI for the H2S inhibition term
                                           0.003875 mol/l (Kaksonen)
                                           1.54 mol/l (Nagpal)
                                           5.13*10^-3 mol/l (Barrera)
```

```
%========================================================================
==
% PRECIPITATION: Acid/base parameters
%========================================================================
==
par.pH15 = 1.5;                  %pH 1.5 in the first precipitation tank
par.pH45 = 4.5;                  %pH 4.5 in the second precipitation tank
par.pH72 = 7.2;                  %pH 7.2 in the third precipitation tank

%========================================================================
==
% PRECIPITATION: Precipitation rate constants + conversion
yields/coefficients
%========================================================================
==
par.k_Cu = 6.71e-04*3600;        % precipitation rate constant for CuS
                                   precipitation. [h^-1]
                                   Tokuda et al.(2008)
par.k_Zn = 8.18e-04*3600;        % precipitation rate constant for ZnS
                                   precipitation, also used for MeS
                                   precipitation. [h^-1]
                                   Tokuda et al. (2008)

par.YCuS = 0.966;                % Conversion achieved for the CuS
reaction
par.YZnS = 0.96;                 % Conversion achieved for the ZnS
reaction
par.YMeS = 1;                    % Conversion achieved for the MeS
reaction

%========================================================================
==
% PRECIPITATION: Molar flows of [Cu2+]in [Zn2+]in [Me2+]in (from RISE
PARTNER +)
%========================================================================
==

par.Cu_in = 89.11/(par.qflow);             % Ingoing Cu2+(mol/L)
par.Zn_in = 2146.834/(par.qflow);          % Ingoing Zn2+(mol/L)
par.Me_in = 14500.70366/(par.qflow);       % Ingoing Me2+(mol/L)

%========================================================================
==
% PRECIPITATION: Tank/process/unit operation parameters
%========================================================================
==
par.Vliq_bio = (301496.09+305095.98)/2*6;    % Bioreactor/fermenter average
                                               size currently in SuperPro
                                           (L)
par.Vliq_tank1 = 96.13e+03*18;               % Precipitation tank 1: 96.13
m3

                                               currently in SuperPro =
                                               96.13e+03 L
par.Vliq_tank2 = 95.89e+03*18;               % Precipitation tank 2: 95.89
m3

                                               currently in SuperPro =
                                               95.89e+033 L
par.Vliq_tank3 = 97.47e+03*33;               % Precipitation tank 3: 97.47
m3

                                               currently in SuperPro =
```

```
                                                    97.47e+03 L

par.V_gas = par.Vliq_bio*0.17;                    % Gas volume [L] (from
SuperPro)

save('par','par')                                 % save par as par in a .mat
file
end
```

Below is the ODE solver, used to solve and graphically display the results of the previously stated model.

```
% ODE solver, outputs and subplots for the output.
close all
clear all

Data = molLpar;                                 %Load indata from parameter file
load par;

Data = Ppardata;                                %Load data from literature
load D;
%...........................................................................
% Time
%...........................................................................

tspan = linspace(0, 1000, 100); % [h]

%===========================================================================
% Start values
%===========================================================================
%BIOREACTOR
S_Sred0 = par.S_red_in;                   %H2S liquid concentration [mol/L]
S_MeOH0 = par.S_MeOH_in;                  %MeOH concentration out from the
                                           bioreactor  [mol/L]
S_SO40 = par.S_SO4_in;                    %SO4 concentration out from the
                                           bioreactor [mol/L]
X_mSRB0 = 0.000001;                       %MeOH biomass [mol/L]
S_HS0 = 0;                                %Dissovled H2S in HS form [mol/L]
S_IC0 = 0;                                %Inorganic carbon in liquid phase
S_HCO3 = 0;                               %Ion state of CO2 and HCO3
S_H2S_g0 = 0.0001;                        %H2S concentration gas phase
S_CO2_g0 = 0.0001;                        %CO2 in gas phase

% TANK 1
S_HS150 = 0;                             %Dissolved H2S in HS form pH 1.5
S_HCO3150 = 0;                           %Dissolved CO2 in HCO3 form pH 1.5
SCu0 = par.Cu_in;                        %Outgoing Cu [mol/L]

% TANK 2
S_HS450 = 0;                             %Dissolved H2S in HS form pH 4.5
S_HCO3450 = 0;                           %Dissolved CO2 in HCO3 form pH 4.5
SZn0 = par.Zn_in;                        %Outgoing Zn [mol/L]

% TANK 3
S_HS720 = 0;                             %Dissolved H2S in HS form pH 7.2
S_HCO3720 = 0;                           %Dissolved CO2 in HCO3 form pH 7.2
SMe0 = par.Me_in;                        %Outgoing Me [mol/L]

y0 = [S_Sred0 S_MeOH0 S_SO40 X_mSRB0  S_HS0 S_IC0 S_HCO3 S_H2S_g0 ...
```

```
  S_CO2_g0 S_HS150 S_HCO3150 SCu0 S_HS450 S_HCO3450 SZn0  S_HS720 S_HCO3720
SMe0 ]; %

[t,y] = ode15s(@molL2,tspan,y0,[],par);

%=============================================================================
% Outputs
%=============================================================================
%BIOREACTOR
R.Reducedsulfur = y(:,1);                          %Total amount of reduced
sulfur
                                                    out from the bioreactor
R.Methanol =  y(:,2);                              %MeOH left
R.Sulfate = y(:,3);                                %SO4 left
R.Mbiomass = y(:,4);                               %EtOH biomass
R.DisSulfide = y(:,5);                             %Dissolved H2S
R.ICarbonliquid = y(:,6);                          %Inorganic carbon in liquid
                                                    phase
R.Carbonate = y(:,7);                              %Dissociated CO2
R.Sulfidegas = y(:,8);                             %H2S gas phase
R.Carbondioxidegas = y(:,9);                       %CO2 gas phase

% TANK 1
R.DisSulfide15 = y(:,10);                          %Dissolved H2S
R.Carbonate15 = y(:,11);                           %Dissociated CO2
R.Copper = y(:,12);                                %Cu left



% TANK 2
R.DisSulfide45 = y(:,13);                          %Dissolved H2S
R.Carbonate45 = y(:,14);                           %Dissociated CO2
R.Zinc =   y(:,15);                                %Zn left

% TANK 3
R.DisSulfide72 = y(:,16);                          %Dissolved H2S
R.Carbonate72 = y(:,17);                           %Dissociated CO2
R.Metal = y(:,18);                                 %Me left

R.tspan = linspace(0, 1000, 100);                  %Time [h]
R.t=t;
save ('R','R')

%% Plots over the results

a.fig1 = figure(1);
a.fig1 = subplot(2,2,1);
a.fig1 = plot(R.tspan,R.Reducedsulfur);
a.ax1 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax1 = ylabel('Reduced sulfur-liquid [mol/L]');
a.tit1 = title('Reduced sulfur-liquid');

a.ax1.FontName = 'Times New Roman';
a.ax1.FontSize = 20;
a.yax1.FontName = 'Times New Roman';
a.yax1.FontSize = 20;
a.tit1.FontName = 'Times New Roman';
a.tit1.FontSize = 20;
```

```
a.fig1 = subplot(2,2,2);
a.fig1 = plot(R.tspan,R.Methanol);
a.ax2 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax2 = ylabel('Methanol [mol/L]');
a.tit2 = title('Methanol concentration');

a.ax2.FontName = 'Times New Roman';
a.ax2.FontSize = 20;
a.yax2.FontName = 'Times New Roman';
a.yax2.FontSize = 20;
a.tit2.FontName = 'Times New Roman';
a.tit2.FontSize = 20;

a.fig1 = subplot(2,2,3);
a.fig1 = plot(R.tspan,R.Sulfate);
a.ax3 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax3 = ylabel('Sulfate [mol/L]');
a.tit3 = title('Sulfate concentration');

a.ax3.FontName = 'Times New Roman';
a.ax3.FontSize = 20;
a.yax3.FontName = 'Times New Roman';
a.yax3.FontSize = 20;
a.tit3.FontName = 'Times New Roman';
a.tit3.FontSize = 20;

a.fig1 = subplot(2,2,4);
a.fig1 = plot(R.tspan,R.Mbiomass);
a.ax4 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax4 = ylabel('Biomass [mol/L]');
a.tit4 = title('Biomass on Methanol and Sulfate');

a.ax4.FontName = 'Times New Roman';
a.ax4.FontSize = 20;
a.yax4.FontName = 'Times New Roman';
a.yax4.FontSize = 20;
a.tit4.FontName = 'Times New Roman';
a.tit4.FontSize = 20;

%%
b.fig2 = figure(2);
b.fig2 = subplot(3,2,1);
b.fig2 = plot(R.tspan,R.Sulfidegas);
b.ax1 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax1 = ylabel('H_2S-gas [mol/l]');
b.tit1 = title('H_2S-gas phase');

b.ax1.FontName = 'Times New Roman';
b.ax1.FontSize = 20;
b.yax1.FontName = 'Times New Roman';
b.yax1.FontSize = 20;
b.tit1.FontName = 'Times New Roman';
b.tit1.FontSize = 20;

b.fig2 = subplot(3,2,2);
b.fig2 = plot(R.tspan,R.Carbondioxidegas);
```

```
b.ax2 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax2 = ylabel('CO_2 [mol/l]');
b.tit2 = title('CO_2-gas phase');

b.ax2.FontName = 'Times New Roman';
b.ax2.FontSize = 20;
b.yax2.FontName = 'Times New Roman';
b.yax2.FontSize = 20;
b.tit2.FontName = 'Times New Roman';
b.tit2.FontSize = 20;

b.fig2 = subplot(3,2,3);
b.fig2 = plot(R.tspan,R.ICarbonliquid);
b.ax3 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax3 = ylabel('Inorganic Carbon-liquid [mol/l]');
b.tit3 = title('Inorganic Carbon-liquid');

b.ax3.FontName = 'Times New Roman';
b.ax3.FontSize = 20;
b.yax3.FontName = 'Times New Roman';
b.yax3.FontSize = 20;
b.tit3.FontName = 'Times New Roman';
b.tit3.FontSize = 20;

b.fig2 = subplot(3,2,4);
b.fig2 = plot(R.tspan,R.DisSulfide);
b.ax4 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax4 = ylabel('HS^-[mol/l]');
b.tit4 = title('Concentration HS^-');

b.ax4.FontName = 'Times New Roman';
b.ax4.FontSize = 20;
b.yax4.FontName = 'Times New Roman';
b.yax4.FontSize = 20;
b.tit4.FontName = 'Times New Roman';
b.tit4.FontSize = 20;

b.fig2 = subplot(3,2,5);
b.fig2 = plot(R.tspan,R.Carbonate);
b.ax5 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax5 = ylabel('HCO_3^- [mol/l]');
b.tit5 = title('Concentration HCO_3^-');

b.ax5.FontName = 'Times New Roman';
b.ax5.FontSize = 20;
b.yax5.FontName = 'Times New Roman';
b.yax5.FontSize = 20;
b.tit5.FontName = 'Times New Roman';
b.tit5.FontSize = 20;

subplot(3,2,6);
plot(R.tspan,R.Reducedsulfur,'*');
hold on
plot(R.tspan,R.Sulfidegas);
b.ax6 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
```

```
b.yax6 = ylabel('Concentration [mol/l]');
b.tit6 = title('Reduced sulfur-liquid and H_2S-gas');
b.leg6 = legend('Reduced sulfur-liquid','H_2S-gas');

b.ax6.FontName = 'Times New Roman';
b.ax6.FontSize = 20;
b.yax6.FontName = 'Times New Roman';
b.yax6.FontSize = 20;
b.tit6.FontName = 'Times New Roman';
b.tit6.FontSize = 20;
b.leg6.FontName = 'Times New Roman';
b.leg6.FontSize = 20;

%%
c.fig3 = figure(3)
c.fig3 = subplot(2,2,1)
c.fig3 = plot(R.tspan,R.Copper,'LineWidth',2)
c.ax1 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax1 = ylabel('Cu^(2+) [mol/L]')
c.tit1 = title('Concentration Cu^2+')
hold on
grid
hold off

c.ax1.FontName = 'Times New Roman';
c.ax1.FontSize = 20;
c.yax1.FontName = 'Times New Roman';
c.yax1.FontSize = 20;
c.tit1.FontName = 'Times New Roman';
c.tit1.FontSize = 20;

c.fig3 = subplot(2,2,2)
c.fig3 = plot(R.tspan,R.Zinc,'LineWidth',2)
c.ax2 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax2 = ylabel('Zn [mol/L]')
c.tit2 = title('Concentration Zn^2+')
hold on
grid
hold off

c.ax2.FontName = 'Times New Roman';
c.ax2.FontSize = 20;
c.yax2.FontName = 'Times New Roman';
c.yax2.FontSize = 20;
c.tit2.FontName = 'Times New Roman';
c.tit2.FontSize = 20;

c.fig3 = subplot(2,2,3)
c.fig3 = plot(R.tspan,R.Metal,'LineWidth',2)
c.ax3 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax3 = ylabel('Me [mol/L]')
c.tit3 = title('Concentration Me^2+')
hold on
grid
hold off

c.ax3.FontName = 'Times New Roman';
```

```
c.ax3.FontSize = 20;
c.yax3.FontName = 'Times New Roman';
c.yax3.FontSize = 20;
c.tit3.FontName = 'Times New Roman';
c.tit3.FontSize = 20;
```

Appendix A.6 – Matlab code, case 3 scenario 2

Below the script defining the process model is stated.

```
function [ dy ] = molL3( t,y,par)

%Based on using both MeOH and Ac- as substrate, the same as for case 3 in
%SuperPro.

%OBS! literature and SuperPro data has been used in combination.
%% STATE VARIABLES
%==========================================================================
==
%BIOREACTOR
S_Sred = y(1);             %Reduced sulfur, total or
S_MeOH = y(2);             %MeOH
S_SO4 = y(3);              %Sulfate
X_mSRB = y(4);             %MeOH biomass
S_Ac = y(5);               %Acetate
X_aSRB = y(6);             %Acetate biomass
S_HS = y(7);               %Dissolved H2S in HS form
S_IC = y(8);               %Inorganic carbon in liquid phase
S_HCO3 = y(9);             %Dissolved CO2 in HCO3 form
S_H2S_g = y(10);           %H2S in gas phase
S_CO2_g = y(11);           %CO2 in gas phase

% TANK 1
S_HS15 = y(12);            %Dissolved H2S in HS form pH 1.5
S_HCO315 = y(13);          %Dissolved CO2 in HCO3 form pH 1.5
S_Cu = y(14);              %Produced CuS

% TANK 2
S_HS45 = y(15);            %Dissolved H2S in HS form pH 4.5
S_HCO345 = y(16);          %Dissolved CO2 in HCO3 form pH 1.5
S_Zn = y(17);              %Produced ZnS

% TANK 3
S_HS72 = y(18);            %Dissolved H2S in HS form pH 7.2
S_HCO372 = y(19);          %Dissolved CO2 in HCO3 form pH 7.2
S_Me = y(20);              %Produced MeS

%%
===========================================================================
==
% Flow rates and relationships (continuous mode, connection between tanks)
%==========================================================================
==

% VOLUMETRIC FLOWS
qp1 = 4.5e-03*par.qflow; %Product outflow from precipitation tank 1 [L/h]
qp2 = 1.1e-01*par.qflow; %Product outflow from precipitation tank 2 [L/h]
qp3 = 3.3e-01*par.qflow; %Product outflow from precipitation tank 3 [L/h]

qin1 = par.qflow;        %Inflow to precip. tank 1 [L/h]
qin2 = qin1 - qp1;       %Inflow to precip. tank 2 [L/h]
qH2O_in3 = 121000;       %Recycling to precip. tank 3 [L/h] According to
                          SuperPro script. (Theoretically/according to flow
                        relationships: qin2-qp2)
qin3 = qin2 - qp2 + qH2O_in3;             %Inflow to precip. tank 3 [L/h]
```

```
q_bin = qin3 - qp3 + par.qs_vol;  % Total flow rate into the bioreactor
[L/h]

q_bout = q_bin - qH2O_in3;        % Outflow from the bioreactor [L/h]

%% BIOREACTOR
%=========================================================================
==
%Inhibition terms (bioreactor)
%=========================================================================
==

S_H = 10^-(par.pHbio);                                         % [mol/L]

%=========================================================================
% High and low pH inhibition
%=========================================================================
% %MeOH utilizing bacteria
% if (par.pH_LL_mSRB < par.pHbio < par.pH_UL_mSRB)
% I_pH_mSRB = 1;
% else
% I_pH_mSRB = ((1 + 2 * 10^(0.5 * (par.pH_LL_mSRB - par.pH_UL_mSRB)))/(1 +
10^(par.pHbio - par.pH_UL_mSRB) + 10^(par.pH_LL_mSRB - par.pHbio)));
%Inhibition term from pH with both high and low inhibition
% end
% %
% % Acetate utilizing bacteria
% if (par.pH_LL_aSRB < par.pH < par.pH_UL_aSRB)       %Ändra par.pH till pH
nÃ¤r Acid-base fungerar.
% I_pH_aSRB = 1;
% else
% I_pH_aSRB = ((1 + 2 * 10^(0.5 * (par.pH_LL_aSRB - par.pH_UL_aSRB)))/(1 +
10^(par.pH - par.pH_UL_aSRB) + 10^(par.pH_LL_aSRB - par.pH))); %Inhibition
term from pH with both high and low inhibition
% end

%=========================================================================
==
% Low pH inhibition
%=========================================================================
==
if(par.pHbio < par.pH_UL_mSRB) %Inhibition term for mSRB
I_pH_mSRB = exp(- 3 * ((par.pHbio - par.pH_UL_mSRB)/(par.pH_UL_mSRB -
par.pH_LL_mSRB))^2);
else
 I_pH_mSRB = 1;
end
%
if(par.pHbio < par.pH_UL_aSRB) %Inhibition term for aSRB
I_pH_aSRB = exp(-3 * ((par.pHbio - par.pH_UL_aSRB)/(par.pH_UL_aSRB -
par.pH_LL_aSRB))^2);
else
I_pH_aSRB = 1;
end

%=========================================================================
==
%H2S inhibition (different suggested terms)
```

```
%============================================================================
==

%Non-competetive inhibition (Provad, fungerar)
%I_H2S = 1/(1 + (S_Sred/par.KI(1)));

%Inhibition for high H2S concentration (Provad och fungerar)
%
I_H2S = exp(-(S_Sred/0.60056 * par.KI(1))^2);         % [(mol/L)/(mol/L)]

%============================================================================
==
% Rates for biomass production, substrate consumption and bacterial decay
%============================================================================
==
%r_MeOH and r_Ac are separate because they're separate parts of the
metabolism and are conusmed separately.

r_MeOH = (par.umax_MeOH(2)/(par.Y_mSO4/0.75)) * S_MeOH/(par.Ks_MeOH +
S_MeOH) * S_SO4/(par.Ks_SO4 + S_SO4)...
    * X_mSRB * I_pH_mSRB * I_H2S;     %Rate for biomass production for
mSRB.
                                      [mol/L*h] 0.75 (or 3/4) because of
                                      reaction stoichiometry

r_SO4 = r_MeOH*0.75;                  %Rate for biomass production for
mSRB,
                                      based on stoichiometry of the SO4
                                      reduction balance. [mol/L*h]

r_dec_mSRB = par.k_dec_mSRB * X_mSRB; %Rate of decay of mSRB [mol/L*h]

r_Ac = (par.umax_Ac/par.Y_AaSRB) * S_Ac/(par.Ks_Ac + S_Ac) *
S_SO4/(par.Ks_SO4 + S_SO4)...
        * X_aSRB * I_pH_aSRB * I_H2S;               %Rate for biomass
production for aSRB. [mol/L*h] 1:1 because of reaction stoichiometry

r_dec_aSRB = par.k_dec_aSRB * X_aSRB; %Rate of decay of aSRB

%============================================================================
==
% Acid base reaction equilibrium - BIOREACTOR
%============================================================================
==
S_H2S = S_Sred-S_HS;          % H2S = [S--] - [HS-]     [mol/L]
S_CO2 = S_IC-S_HCO3;

rho_A_HS = par.K_ab_H2S * (S_HS* (S_H + par.K_a_H2S) - par.K_a_H2S *
S_Sred);
%Dissociation to HS-  [mol^3/L^3*h]

rho_A_HCO3 = par.K_ab_CO2 * (S_HCO3 * (S_H+ par.K_a_CO2) - par.K_a_CO2 *
S_IC);
%Dissociation to HCO3- [mol^3/L^3*h]

%============================================================================
==
% Mass transport for gases - bioreactor
```

```
%=========================================================================
==
p_H2S_gas = S_H2S_g*par.R*par.T ;   %Partial pressure of H2S [atm]
p_CO2_gas = S_CO2_g*par.R*par.T;    %Partial pressure of CO2 [atm]

q_gas = (par.N2g/(1-(p_H2S_gas/1)-(p_CO2_gas/1))); % mol/L*atm

S_prim_H2S = p_H2S_gas/par.H_H2S;
                  %Henry's law, maximum solubility of H2S in liquid [mol/L]

S_prim_CO2 = p_CO2_gas/par.H_CO2;
                  %Henry's law, maximum solubiliy of CO2 in liquid [mol/L]

r_H2St = par.kLa_H2S *(S_H2S-S_prim_H2S); %Mass transport for H2S gas,
                                          %from liquid to gas. [mol/L*h]
r_CO2t = par.kLa_CO2 *(S_CO2-S_prim_CO2); %Mass transport for CO2 gas,
                                          %from liquid to gas. [mol/L*h]

%=========================================================================
==
% Dynamic mass balances - bioreactor
%=========================================================================
==


%Biomass
dX_mSRB_dt = q_bin/par.Vliq_bio*(par.X_mSRB_in - X_mSRB) + par.Y_mSO4 *
r_SO4 - r_dec_mSRB;                                % MeOH biomass change
[mol/L*h]
dX_aSRB_dt = q_bin/par.Vliq_bio*(par.X_aSRB_in - X_aSRB) + par.Y_AaSRB *
r_Ac - r_dec_aSRB;                                 %Ac- biomass change
[mol/L*h]

%Substrate (electron donor) uptake
dS_MeOH_dt = q_bin/par.Vliq_bio * (par.S_MeOH_in - S_MeOH) - r_MeOH;
                                          %Methanol concentration [mol/L*h]
dS_Ac_dt = q_bin/par.Vliq_bio * (par.S_Ac_in - S_Ac) - r_Ac;
                                          %Acetate concentration [mol/L*h]
dS_SO4_dt = q_bin/par.Vliq_bio*(par.S_SO4_in - S_SO4) - r_SO4 - r_Ac;

%Product formation
dS_IC_dt = q_bin/par.Vliq_bio* (par.S_IC_in - S_IC)  - r_CO2t + (1 -
par.Y_AaSRB) * r_Ac;
                                          %Inorganic carbon [mol/L*h]


%Acid-base
d_S_HS_dt = -rho_A_HS;         %Dissociation of H2S to HS-  [mol^3/L^3*h]
d_S_HCO3_dt = -rho_A_HCO3;     %Dissociation of CO2 tp HCO3- [mol^3/L^3*h]

%Gas phase
dS_H2S_g_dt = 0 -
(q_gas/par.V_gas)*S_H2S_g+r_H2St*(par.Vliq_bio/par.V_gas);
%Gas phase for H2S with compensation for volume difference [mol/L*h]
dS_CO2_g_dt = 0 -
(q_gas/par.V_gas)*S_CO2_g+r_CO2t*(par.Vliq_bio/par.V_gas);
%Gas phase for CO2 with compensation for volume difference [mol/L*h]

%%
% PRECIPITATION REACTIONS
```

```
%=============================================================================
==

%% TANK 1
% Acid base expressions
S_H15 = (10^-par.pH15); % [H+] at pH 1.5 [mol/L]
S_Hin1 = S_H15 + S_H/3;  % [mol/L]

S_Sred1 = S_Sred/3;

S_H2S15 = S_Sred1-S_HS15;          % H2S = [S--] - [HS-] at pH 1.5 [mol/L]
S_CO215 = S_IC/3-S_HCO315;         % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St15 = par.kLa_H2S * (S_prim_H2S-S_H2S15); % [mol/L*h]

% Acid base equilibrium rate expression
rho_A_HS15 = par.K_ab_H2S * (S_HS15 * (S_Hin1 + par.K_a_H2S) - par.K_a_H2S*
S_Sred1);
            %Dissociation to HS- in the first precipitation tank [mol/L*h]
rho_A_HCO315 = par.K_ab_CO2 * (S_HCO315 * (S_Hin1 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
            %Dissociation to HCO3- in the first precipitation tank
            [mol/L*h]

% Dissociation rate in precipitation tanks
d_S_HS_dt15 = -rho_A_HS15;
            %Dissociation of H2S to HS- in precipitation tank 1
      [mol^3/L^3*h]
d_S_HCO3_dt15 = -rho_A_HCO315;
            %Dissociation of CO2 to HCO3- in precipitation tank 1
             [mol^3/L^3*h]

if S_Cu >= 0
    S_Cu = par.Cu_in - S_HS15;        % Concentration Cu [mol/L]
    rho_CuS = par.k_Cu*(S_Cu+S_HS15); % Precipitation rate equation
[mol/L*h]
    d_Cu_dt = qin1/par.Vliq_tank1*(par.Cu_in - S_Cu) - rho_CuS * par.YCuS;
    %According to Tokuda et al. (2008) [mol/L*h]% [mol/L]
else
    S_Cu = 0;                         % Concentration Cu [mol/L]
    d_Cu_dt = 0;
    rho_CuS = par.k_Cu*(S_Cu+S_Sred1); % Precipitation rate equation
[mol/L*h]
end


%% TANK 2
% Acid base expressions
S_H45 = (10^-par.pH45);                      % [H+] at pH 4.5 [mol/L]
S_Hin2 = qin1/qin2*S_H15 + S_H45 + S_H/3;    % [mol/L]

S_Sred2 = S_Sred/3;                          % [mol/L]

S_H2S45 = S_Sred2-S_HS45;          % H2S = [S--] - [HS-] at pH 4.5 [mol/L]
S_CO245 = S_IC/3-S_HCO345;         % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St45 = par.kLa_H2S * (S_H2S45-S_prim_H2S); % [mol/L*h]
```

```
% Acid base equilibrium rate expression
rho_A_HS45 = par.K_ab_H2S * (S_HS45 * (S_Hin2 + par.K_a_H2S) - par.K_a_H2S
* S_Sred2);
      %Dissociation to HS- in the second precipitation tank   [mol^3/L^3*h]
rho_A_HCO345 = par.K_ab_CO2 * (S_HCO345 * (S_Hin2 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
      %Dissociation to HCO3- in the second precipitation tank
[mol^3/L^3*h]

% Dissociation rate in precipitation tanks
d_S_HS_dt45 = -rho_A_HS45;
      %Dissociation of H2S to HS- in precipitation tank 2 [mol/L*h]
d_S_HCO3_dt45 = -rho_A_HCO345;
      %Dissociation of CO2 to HCO3- in precipitation tank 2 [mol/L*h]

if S_Zn >= 0
    S_Zn = par.Zn_in - S_HS45; % concentration Zn [mol/L]
    rho_ZnS = par.k_Zn*(S_Zn+S_HS45); % Precipitation rate equation
[mol/L*h]
    d_Zn_dt = qin2/par.Vliq_tank2*(par.Zn_in - S_Zn)- rho_ZnS * par.YZnS;
    %According to Tokuda et al. (2008) [mol/L*h]
else
    S_Zn = 0;                               % Concentration Zn [mol/L]
    rho_ZnS = par.k_Zn*(S_Zn+S_Sred2);      % Precipitation rate equation
                                              [mol/L*h]
    d_Zn_dt = 0;
end

%% TANK 3
% Acid base expressions
S_H72 = (10^-par.pH72);                     % [H+] at pH 7.2 ([mol/L]
S_Hin3 = qin2/qin3*S_H45 + S_H/3 + S_H72; % [mol/L]

S_Sred3 = S_Sred/3;                         % [mol/L]

S_H2S72 = S_Sred3-S_HS72;            % H2S = [S--] - [HS-] at pH 7.2 [mol/L]
S_CO272 = S_IC/3-S_HCO372;           % [mol/L]

%Mass transport for H2S gas, from liquid to gas.
r_H2St72 = par.kLa_H2S * (S_H2S72-S_prim_H2S);      % [mol/L*h]

% Acid base equilibrium rate expression
rho_A_HS72 = par.K_ab_H2S * (S_HS72 * (S_Hin3 + par.K_a_H2S) - par.K_a_H2S
* S_Sred3);
            %Dissociation to HS- in the third precipitation tank. [mol/L*h]
rho_A_HCO372 = par.K_ab_CO2 * (S_HCO372 * (S_Hin3 + par.K_a_CO2) -
par.K_a_CO2 * S_IC/3);
            %Dissociation to HCO3- in the third precipitation tank.
      [mol/L*h]

% Dissociation rate in precipitation tanks

d_S_HS_dt72 = -rho_A_HS72;
            %Dissociation of H2S to HS- in precipitation tank 3
            [mol^3/L^3*h]
d_S_HCO3_dt72 = -rho_A_HCO372;
            %Dissociation of CO2 to HCO3- in precipitation tank 3
            [mol^3/L^3*h]
```

```
if S_Me >= 0
    S_Me = par.Me_in - S_HS72;                 % Concentration Me [mol/L]
    rho_MeS = par.k_Zn*(S_Me+S_HS72);          % Precipitation rate
equation
                                                 [mol/L*h]
    d_Me_dt = qin3/par.Vliq_tank3*(par.Me_in - S_Me) - rho_MeS * par.YMeS;
    %MeS precipitation is based on ZnS because it is more abundant.
[mol/L*h]
                                                                    %
According to Tokuda et al. (2008)
else
    S_Me = 0;                                   % Concentration Me [mol/L]
    rho_MeS = par.k_Zn*(S_Me+S_Sred3);          % Precipitation rate
equation
                                                  [mol/L*h]
    d_Me_dt = 0;
end

% Product formation with respect to the precipitation
if S_Sred >=0
    dS_S_red_dt = q_bin/par.Vliq_bio*(par.S_red_in - S_Sred) + (1-
par.Y_mSO4) * r_SO4 - r_H2St...
    + (1 - par.Y_AaSRB)  * r_Ac - rho_CuS * par.YCuS - rho_ZnS * par.YZnS -
rho_MeS * par.YMeS;
%Reduced sulfur = [S--]+[HS-]+[H2S] [mol/L*h] %Reduced sulfur = [S--]+[HS-
]+[H2S] [mol/L*h] *par.Y_aSO4
else
    dS_S_red_dt = q_bin/par.Vliq_bio*(par.S_red_in - S_Sred) + (1-
par.Y_mSO4) * r_SO4 - r_H2St...
    + (1 - par.Y_AaSRB)* r_Ac;
end

%============================================================================
==

dy = [dS_S_red_dt;dS_MeOH_dt;dS_SO4_dt;dX_mSRB_dt; dS_Ac_dt;dX_aSRB_dt;
d_S_HS_dt;dS_IC_dt;d_S_HCO3_dt;dS_H2S_g_dt;dS_CO2_g_dt;...

d_S_HS_dt15;d_S_HCO3_dt15;d_Cu_dt;d_S_HS_dt45;d_S_HCO3_dt45;d_Zn_dt;d_S_HS_
dt72;d_S_HCO3_dt72;d_Me_dt];

end
```

Below the .m-file containing the process parameters are stated.

```
function [par] = molL3par


%OBS! Somewhat modified data. Tidigare litteratur data används tillsammans
med SuperPro data.

%============================================================================
====================
% FLOW PARAMETERS - Overall process
%============================================================================
====================
par.qflow = 123547.3322;                   % VOLUMETRIC BASE FLOW INTO THE
                                             PROCESS (IE. LEACHATE FLOW)[L/h]
```

```
par.qs_vol = 1673.9493;                   % VOLUMETRIC BASE FLOW OF
SUBSTRATE
                                            [L/h]
par.q_g_vol = 506.8165e03;                % VOLUMETRIC gas flow from the
                                            bioreactor [L/h]

par.q_in = 6766673.6064/par.qflow;         %Total Flow rate of leachate into
                                           the precipitation [mol/L]
par.qs = 37354.55/par.qs_vol;             %Total Flow rate of substrate into
                                           the bioreactor  [mol/L]
par.qp = 0.001*par.q_in;
par.q_g = 20717.2/par.q_g_vol;            %Total Flow rate gas phase out
from
                                           the bioreactor [mol/L]

%===========================================================================
====================
% Coefficients for: MeOH degrading SRB: mSRB // Acetate degrading SRB: aSRB
%===========================================================================
====================
par.umax_MeOH = [0.013 0.612];             %Maximum specific growth rate of
                                            eSRB, 0.013 [h^-1](Nagpal)
                                            0.612 (d^-1) (Barrera)
par.Y_mSO4 = 0.5/24;                       %Yield for biomass on sulfate,
                                            mSRB, YSO4X (Nagpal)
                                           [gprotein/molSO4/96.6gSO4/mol] =
                                           [g/g]
par.Y_MmSRB = 0.5*par.Y_mSO4;      %(Nagpal) Yield of mSRB biomass on MeOH
par.Ks_MeOH = 0.0045;              %Ks for MeOH is approximated to be equal
to
                                    that of EtOH [mol/l] (Nagpal)
par.Ks_SO4 = 0.0085;               %Ks for SO4 [mol/l] (Nagpal) 0.297*10^-3
                                    mol/l (Barrera)

par.umax_Ac = 0.063;               %Maximum specific growth rate of aSRB,
                                    0.063 [h^-1](Moosa)
par.Y_AaSRB = 0.58;                %Yield for Acetate on biomass, YACX,
                                    [g bacteria/g acetate] (Moosa)
par.Ks_Ac = 0.0035;                %Ks for Acetate, Bara provat värde
24*10^-3
                                    mol/l (Barrera)

%===========================================================================
====================
% Coefficients for: BIOREACTOR INGREDIENT FEEDS/FLOWS
%===========================================================================
====================
par.X_mSRB_in = 0.00000002*par.q_in;         %Inflow of mSRB biomass [mol/L]
par.X_aSRB_in = 0.00000002*par.q_in;         %Inflow of aSRB biomass [mol/L]

par.S_MeOH_in = 11432/(1673.9493+152423.2011);
%Substrate MeOH concentration inflow [mol/L] from SuperPro - RISE PARTNER +
CASE 1 opt
par.S_Ac_in = 11432/(1673.9493+152423.2011);
%Substrate Ac- concentration inflow [mol/L] from SuperPro - RISE PARTNER +
CASE 3 opt
par.S_SO4_in = 19559.59/(1673.9493+152423.2011);
%Sulfate concentration inflow [mol/L] from SuperPro  - RISE PARTNER + CASE1
```

```
par.Y_aSO4 = 0.5/24;                 %(Nagpal) Yield for sulfate on
biomass,
                                       aSRB [gprotein/molSO4/96.6gSO4/mol]
par.S_red_in = 0.00000000001*par.q_in*2;     %Reduced sulfur concentration
                                              inflow [mol/L]
par.S_IC_in = 0.00000000001*par.q_in*2;      %Inorganic carbon
concentration
                                              inflow [mol/L]

par.k_dec_mSRB = 0.00;                       %Decay coefficient for MeOH
SRB
                                              [h^-1]
par.k_dec_aSRB = 0.00;                       %Decay coefficient for acetate
                                              SRB. (Moosa et al. 2002) [h^-
                                          1]

%============================================================================
======================
% Mass transfer gas-liquid parameters: BIOREACTOR
%============================================================================
======================
par.kLa_H2S = 10;                   %kLa mass transfer coefficient for H2S
                                     [h^-1] (Nagpal)
par.kLa_CO2 = 10;                   %kLa mass transfer coefficient for CO2
                                     [h^-1]
par.H_H2S = 10.70;                  %Henrys constant for H2S [atmL/mol]
10.70;
                                     (Nagpal)
par.H_CO2 = 25.88;                  %Henrys constant for CO2 [atmL/mol]
25.88;
                                     (Nagpal)
par.T = 298;                        %Temperature [K]
par.R = 0.082057338;                %Gas constant [L atm K^-1 mol^-1]
par.V_liq = 343288;                 %Liquid volume [L] (from SuperPro)
par.C_gas = 0.0409;                 %Gas concentration in outgoing flow from
                                     the reactor [mol/L]

par.P_tot = 1;                      %Total pressure from all gases [atm]
par.N2g = 435244;                   %Stripping gas, Flow of N2 [L/h]

par.density_N2 = 1.2151;            %Density for N2[g/L]
par.mol_N2 =28;                     %Molar weight for N2[g/mol]
%============================================================================
======================
% BIOREACTOR: Acid-base coefficients
%============================================================================
=====================
par.K_ab_H2S = 10^15;               %Acid base kinetic parameter for H2S
par.K_a_H2S = 1.075 * 10^-7;        %Acid-base equilibirum coefficient for
H2S

par.K_a_CO2 = 4.97 * 10^-7;         %Acid-base equilibirum coefficient for
                                     HCO3
par.K_ab_CO2 = 10^12;               %Acid base kinetic parameter for HCO3

%============================================================================
======================
% Inhibition parameters - BIOREACTOR
%============================================================================
======================
```

```
par.pH_LL_mSRB = 6;               %Lower pH limit where the group of
                                   organisms (eSRB) are 50% inhibited
par.pH_UL_mSRB = 7.5;             %Upper pH limit where the group of
                                   organisms (eSRB) are 50% inhibited
par.pH_LL_aSRB = 6;               %Lower pH limit where the group of
                                   organisms (aSRB) are 50% inhibited
par.pH_UL_aSRB = 7.5;             %Upper pH limit where the group of
                                   organisms (aSRB) are 50% inhibited
par.pHbio = 7;                    %pH for the pH inhibition term

par.KI = [5.13*10^-3 0.003875 1.54];     %KI for the H2S inhibition term
                                          0.003875 mol/l (Kaksonen)
                                          1.54 mol/l (Nagpal)
                                          5.13*10^-3 mol/l (Barrera)

%===========================================================================
==
% PRECIPITATION: Acid/base parameters
%===========================================================================
==

par.pH15 = 1.5;                   %pH 1.5 in the first precipitation tank
par.pH45 = 4.5;                   %pH 4.5 in the second precipitation tank
par.pH72 = 7.2;                   %pH 7.2 in the third precipitation tank

%===========================================================================
==
% PRECIPITATION: Precipitation rate constants + conversion
yields/coefficients
%===========================================================================
==
par.k_Cu = 6.71e-04*3600;         % precipitation rate constant for CuS
                                    precipitation. [h^-1]
                                    Tokuda et al.(2008)
par.k_Zn = 8.18e-04*3600;         % precipitation rate constant for ZnS
                                    precipitation, also used for MeS
                                    precipitation. [h^-1]
                                    Tokuda et al.(2008)

par.YCuS = 0.966;                 % Conversion achieved for the CuS
reaction
par.YZnS = 0.96;                  % Conversion achieved for the ZnS
reaction
par.YMeS = 1;                     % Conversion achieved for the MeS
reaction

%===========================================================================
==
% PRECIPITATION: Molar flows of [Cu2+]in [Zn2+]in [Me2+]in (from RISE
PARTNER +)
%===========================================================================
==

par.Cu_in = 89.11/(par.qflow);              % Ingoing Cu2+(mol/L)
par.Zn_in = 2146.834/(par.qflow);           % Ingoing Zn2+(mol/L)
par.Me_in = 14500.70366/(par.qflow);        % Ingoing Me2+(mol/L)

%===========================================================================
==
% PRECIPITATION: Tank/process/unit operation parameters
```

```
%==========================================================================
==
par.Vliq_bio = (301496.09+305095.98)/2*6;    % Bioreactor/fermenter average
                                               size currently in SuperPro
                                          (L)
par.Vliq_tank1 = 96.13e+03*18;               % Precipitation tank 1: 96.13
m3

                                               currently in SuperPro =
                                               96.13e+03 L
par.Vliq_tank2 = 95.89e+03*18;               % Precipitation tank 2: 95.89
m3

                                               currently in SuperPro =
                                               95.89e+033 L
par.Vliq_tank3 = 97.47e+03*33;               % Precipitation tank 3: 97.47
m3

                                               currently in SuperPro =
                                               97.47e+03 L


par.V_gas = par.Vliq_bio*0.17;               % Gas volume [L] (from
SuperPro)

save('par','par')                            % save par as par in a .mat
file
end
```

Below the ODE solver is stated. It was applied to solve the ODE's as well as graphically display the results.

```
% ODE solver, outputs and subplots for the output.
close all
clear all

Data = molL3par;          %Load indata from parameter file
load par;

Data = Ppardata;         %Load data from literature
load D;
%.........................................................................
% Time
%.........................................................................

tspan = linspace(0, 100, 100); % [h]

%==========================================================================
% Start values
%==========================================================================
%BIOREACTOR
S_Sred0 = par.S_red_in;              %H2S liquid concentration [mol/L]
S_MeOH0 = par.S_MeOH_in;             %MeOH concentration out from the
                                      bioreactor [mol/L]
S_SO40 = par.S_SO4_in;               %SO4 concentration out from the
                                      bioreactor [mol/L]
X_mSRB0 = 0.000001;                  %MeOH biomass [mol/L]
S_Ac0 = par.S_Ac_in;                 %Acetate concentration
X_aSRB0 = 0.000001;                  %Acetate biomass
S_HS0 = 0;                           %Dissovled H2S in HS form [mol/L]
S_IC0 = 0;                           %Inorganic carbon in liquid phase
S_HCO3 = 0;                          %Ion state of CO2 and HCO3
S_H2S_g0 = 0.0001;                   %H2S concentration gas phase
```

```
S_CO2_g0 = 0.0001;                    %CO2 in gas phase

% TANK 1
S_HS150 = 0;                         %Dissolved H2S in HS form pH 1.5
S_HCO3150 = 0;                       %Dissolved CO2 in HCO3 form pH 1.5
SCu0 = par.Cu_in;                    %Outgoing Cu [mol/L]

% TANK 2
S_HS450 = 0;                         %Dissolved H2S in HS form pH 4.5
S_HCO3450 = 0;                       %Dissolved CO2 in HCO3 form pH 4.5
SZn0 = par.Zn_in;                    %Outgoing Zn [mol/L]

% TANK 3
S_HS720 = 0;                         %Dissolved H2S in HS form pH 7.2
S_HCO3720 = 0;                       %Dissolved CO2 in HCO3 form pH 7.2
SMe0 = par.Me_in;                    %Outgoing Me [mol/L]

y0 = [S_Sred0 S_MeOH0 S_SO40 X_mSRB0 S_Ac0 X_aSRB0 S_HS0 S_IC0 S_HCO3
S_H2S_g0 ...
  S_CO2_g0 S_HS150 S_HCO3150 SCu0 S_HS450 S_HCO3450 SZn0  S_HS720 S_HCO3720
SMe0 ];

[t,y] = ode15s(@molL3,tspan,y0,[],par);

%==========================================================================
% Outputs
%==========================================================================
%BIOREACTOR
R.Reducedsulfur = y(:,1);                  %Total amount of reduced sulfur
out
R.Methanol =  y(:,2);                      %MeOH left
R.Sulfate = y(:,3);                        %SO4 left
R.Mbiomass = y(:,4);                       %EtOH biomass
R.Acetate = y(:,5);                        %Acetate concentration
R.Abiomass = y(:,6);                       %Acetate biomass
R.DisSulfide = y(:,7);                     %Dissolved H2S
R.ICarbonliquid = y(:,8);                  %Inorganic carbon in liquid phase
R.Carbonate = y(:,9);                      %Dissociated CO2
R.Sulfidegas = y(:,10);                    %H2S gas phase
R.Carbondioxidegas = y(:,11);              %CO2 gas phase

% TANK 1
R.DisSulfide15 = y(:,12);                  %Dissolved H2S
R.Carbonate15 = y(:,13);                   %Dissociated CO2
R.Copper = y(:,14);                        %Cu left


% TANK 2
R.DisSulfide45 = y(:,15);                  %Dissolved H2S
R.Carbonate45 = y(:,16);                   %Dissociated CO2
R.Zinc =  y(:,17);                         %Zn left

% TANK 3
R.DisSulfide72 = y(:,18);                  %Dissolved H2S
R.Carbonate72 = y(:,19);                   %Dissociated CO2
R.Metal = y(:,20);                         %Me left

R.tspan = linspace(0, 100, 100);           %Time [h]
R.t=t;
```

```
save ('R','R')
%%
a.fig1 = figure(1);
a.fig1 = subplot(3,2,1);
a.fig1 = plot(R.tspan,R.Reducedsulfur);
a.ax1 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax1 = ylabel('Reduced sulfur-liquid [mol/L]');
a.tit1 = title('Reduced sulfur-liquid');

a.ax1.FontName = 'Times New Roman';
a.ax1.FontSize = 20;
a.yax1.FontName = 'Times New Roman';
a.yax1.FontSize = 20;
a.tit1.FontName = 'Times New Roman';
a.tit1.FontSize = 20;

a.fig1 = subplot(3,2,2);
a.fig1 = plot(R.tspan,R.Methanol);
a.ax2 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax2 = ylabel('Methanol [mol/L]');
a.tit2 = title('Methanol concentration');

a.ax2.FontName = 'Times New Roman';
a.ax2.FontSize = 20;
a.yax2.FontName = 'Times New Roman';
a.yax2.FontSize = 20;
a.tit2.FontName = 'Times New Roman';
a.tit2.FontSize = 20;

a.fig1 = subplot(3,2,3);
a.fig1 = plot(R.tspan,R.Sulfate);
a.ax3 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax3 = ylabel('Sulfate [mol/L]');
a.tit3 = title('Sulfate concentration');

a.ax3.FontName = 'Times New Roman';
a.ax3.FontSize = 20;
a.yax3.FontName = 'Times New Roman';
a.yax3.FontSize = 20;
a.tit3.FontName = 'Times New Roman';
a.tit3.FontSize = 20;

a.fig1 = subplot(3,2,4);
a.fig1 = plot(R.tspan,R.Mbiomass);
a.ax4 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax4 = ylabel('Biomass [mol/L]');
a.tit4 = title('Biomass on Methanol and Sulfate');

a.ax4.FontName = 'Times New Roman';
a.ax4.FontSize = 20;
a.yax4.FontName = 'Times New Roman';
a.yax4.FontSize = 20;
a.tit4.FontName = 'Times New Roman';
a.tit4.FontSize = 20;

a.fig1 = subplot(3,2,5);
```

```
a.fig1 = plot(R.tspan,R.Acetate);
a.ax5 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax5 = ylabel('Acetate [mol/L]');
a.tit5 = title('Acetate concentration');

a.ax5.FontName = 'Times New Roman';
a.ax5.FontSize = 20;
a.yax5.FontName = 'Times New Roman';
a.yax5.FontSize = 20;
a.tit5.FontName = 'Times New Roman';
a.tit5.FontSize = 20;

a.fig1 = subplot(3,2,6);
a.fig1 = plot(R.tspan,R.Abiomass);
a.ax6 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
a.yax6 = ylabel('Biomass [mol/L]');
a.tit6 = title('Biomass on Acetate and Sulfate');

a.ax6.FontName = 'Times New Roman';
a.ax6.FontSize = 20;
a.yax6.FontName = 'Times New Roman';
a.yax6.FontSize = 20;
a.tit6.FontName = 'Times New Roman';
a.tit6.FontSize = 20;


%%
b.fig2 = figure(2);
b.fig2 = subplot(3,2,1);
b.fig2 = plot(R.tspan,R.Sulfidegas);
b.ax1 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax1 = ylabel('H_2S-gas [mol/l]');
b.tit1 = title('H_2S-gas phase');

b.ax1.FontName = 'Times New Roman';
b.ax1.FontSize = 20;
b.yax1.FontName = 'Times New Roman';
b.yax1.FontSize = 20;
b.tit1.FontName = 'Times New Roman';
b.tit1.FontSize = 20;

b.fig2 = subplot(3,2,2);
b.fig2 = plot(R.tspan,R.Carbondioxidegas);
b.ax2 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax2 = ylabel('CO_2 (g) [mol/l]');
b.tit2 = title('CO_2-gas phase');

b.ax2.FontName = 'Times New Roman';
b.ax2.FontSize = 20;
b.yax2.FontName = 'Times New Roman';
b.yax2.FontSize = 20;
b.tit2.FontName = 'Times New Roman';
b.tit2.FontSize = 20;

b.fig2 = subplot(3,2,3);
```

```
b.fig2 = plot(R.tspan,R.ICarbonliquid);
b.ax3 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax3 = ylabel('Inorganic Carbon-liquid [mol/l]');
b.tit3 = title('Inorganic Carbon-liquid');

b.ax3.FontName = 'Times New Roman';
b.ax3.FontSize = 20;
b.yax3.FontName = 'Times New Roman';
b.yax3.FontSize = 20;
b.tit3.FontName = 'Times New Roman';
b.tit3.FontSize = 20;

b.fig2 = subplot(3,2,4);
b.fig2 = plot(R.tspan,R.DisSulfide);
b.ax4 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax4 = ylabel('HS^-[mol/l]');
b.tit4 = title('Concentration HS^-');

b.ax4.FontName = 'Times New Roman';
b.ax4.FontSize = 20;
b.yax4.FontName = 'Times New Roman';
b.yax4.FontSize = 20;
b.tit4.FontName = 'Times New Roman';
b.tit4.FontSize = 20;

b.fig2 = subplot(3,2,5);
b.fig2 = plot(R.tspan,R.Carbonate);
b.ax5 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax5 = ylabel('HCO_3^- [mol/l]');
b.tit5 = title('Concentration HCO_3^-');

b.ax5.FontName = 'Times New Roman';
b.ax5.FontSize = 20;
b.yax5.FontName = 'Times New Roman';
b.yax5.FontSize = 20;
b.tit5.FontName = 'Times New Roman';
b.tit5.FontSize = 20;

subplot(3,2,6);
plot(R.tspan,R.Reducedsulfur,'*');
hold on
plot(R.tspan,R.Sulfidegas);
b.ax6 = xlabel('Time [h]');
set(gca,'FontSize',20,'FontName','Times New Roman');
b.yax6 = ylabel('Concentration [mol/l]');
b.tit6 = title('Reduced sulfur-liquid and H_2S-gas');
%b.leg6 = legend('Reduced sulfur-liquid','H_2S-gas');

b.ax6.FontName = 'Times New Roman';
b.ax6.FontSize = 20;
b.yax6.FontName = 'Times New Roman';
b.yax6.FontSize = 20;
b.tit6.FontName = 'Times New Roman';
b.tit6.FontSize = 20;
b.leg6.FontName = 'Times New Roman';
%b.leg6.FontSize = 20;
```

```
%%
c.fig3 = figure(3)
c.fig3 = subplot(2,2,1)
c.fig3 = plot(R.tspan,R.Copper,'LineWidth',2)
c.ax1 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax1 = ylabel('Cu^(2+) [mol/L]')
c.tit1 = title('Concentration Cu^2+')
hold on
grid
hold off

c.ax1.FontName = 'Times New Roman';
c.ax1.FontSize = 20;
c.yax1.FontName = 'Times New Roman';
c.yax1.FontSize = 20;
c.tit1.FontName = 'Times New Roman';
c.tit1.FontSize = 20;

c.fig3 = subplot(2,2,2)
c.fig3 = plot(R.tspan,R.Zinc,'LineWidth',2)
c.ax2 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax2 = ylabel('Zn [mol/L]')
c.tit2 = title('Concentration Zn^2+')
hold on
grid
hold off

c.ax2.FontName = 'Times New Roman';
c.ax2.FontSize = 20;
c.yax2.FontName = 'Times New Roman';
c.yax2.FontSize = 20;
c.tit2.FontName = 'Times New Roman';
c.tit2.FontSize = 20;

c.fig3 = subplot(2,2,3)
c.fig3 = plot(R.tspan,R.Metal,'LineWidth',2)
c.ax3 = xlabel('Time [h]')
set(gca,'FontSize',20,'FontName','Times New Roman');
c.yax3 = ylabel('Me [mol/L]')
c.tit3 = title('Concentration Me^2+')
hold on
grid
hold off

c.ax3.FontName = 'Times New Roman';
c.ax3.FontSize = 20;
c.yax3.FontName = 'Times New Roman';
c.yax3.FontSize = 20;
c.tit3.FontName = 'Times New Roman';
c.tit3.FontSize = 20;
```