Framework for a Mixed-Culture Biofilm Model to Describe Oxidized Nitrogen, Sulfur, and Selenium Removal in a Biofilm Reactor

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ABSTRACT

Biofilm reactors are an emerging environmental biotechnology used for the removal of selenium from industrial wastewater. Bioreactor models for selenium removal that are based on the mechanistic description of biofilms are few in number, but a dynamic model that is based on mechanistic principles (chemical and biochemical) and is capable of describing selenium removal is needed for efficient and more rigorous process design and optimization. This paper summarizes variables, processes, and rate expressions for both chemical and biochemical conversions inside a mixed-culture biofilm containing facultative heterotrophs, sulfate reducing bacteria, and autotrophic denitrifiers. Chemical and biochemical processes in the biofilm model are based on theoretical considerations. Chemical processes include acid-base equilibria reactions primarily to describe substrate. The proposed mathematical description of our multispecies selenium removal biofilm is based on the general one-dimensional model of Wanner and Gujer (1985) and Wanner and Reichert (1995). It is assumed that ions do not undergo chemical or biological transformation, but the ions do not maintain a constant concentration profile across the one-dimensional biofilm. State variables, processes, and kinetic expressions in the biofilm model are structured using (Peterson) matrix notation. The framework for a mechanistic mathematical model describing a dynamic one-dimensional biofilm has been developed and presented.

KEYWORDS
Biofilm, SRB, sulfur reducing bacteria, model, reactor, industrial, wastewater, denitrification
INTRODUCTION
Selenium is widely distributed throughout the environment, and may be found in soils and natural waters. Naturally occurring elemental selenium may be redistributed from these various geological sources into aquatic, sedimentary, atmospheric, and terrestrial compartments (Chapman et al. 2010). Trace amounts of selenium are required to sustain biological processes in ecosystems. Despite its nutritional value, selenium concentrations only slightly greater than those that are required by microorganisms as a micronutrient can be hazardous to the environment and human health. Therefore, selenium can be a serious environmental concern, and is typically regulated in drinking water by environmental protection agencies.

Selenium contamination originates from several sources including mining operations, mineral processing, abandoned mine sites, petroleum processing, and agricultural run-off. Mining coal, for example, creates large waste rock volumes. When exposed to air and water, the release of naturally occurring selenium from the waste rock is accelerated. A review (conducted by CH2M HILL, Inc.) of available environmental biotechnologies for selenium removal from waters concluded that the successful management of selenium-contaminated wastewater to achieve bulk-liquid concentrations in the range 1 to 5 µg/L is dependent on the industry from which the wastewater is discharged. Furthermore, there is a general lack of demonstrated full-scale technologies that are capable of cost-effectively removing selenium from industrial wastewaters.

Regulatory limits related to the discharge of selenium from various industrial processes are becoming increasingly stringent. Therefore, the removal of selenium from contaminated waters presents a significant challenge to various industrial and environmental entities. Selenium contamination may be of a low-level, high volume nature which makes most selenium-laden wastewater treatment options expensive. Therefore, refined design criteria and a mechanistic model capable of optimizing the design of biological selenium removal processes that have been demonstrated effective are warranted.

This paper describes state variables, processes, and transformation-rate expressions for modeling nitrate, sulfate, selenate, and selenite removal from wastewater using a submerged completely mixed biofilm reactor. Pollutant transformation and biomass death, decay, and regeneration is modeled with a series of biochemical processes that occur in aerobic, anoxic, and/or anaerobic environments. The Peterson Matrix, a standard biological process model notation, has been used to present the model. The variables, processes, kinetic expressions and supporting biokinetic parameters presented or discussed in this manuscript are a pre-cursor to the development of a dynamic mixed-culture one-dimensional biofilm model. Ultimately, the dynamic one-dimensional biofilm model may be used for the design and optimize of a biofilm reactor for reduction of the aforementioned compounds. The biofilm (reactor) model is introduced in this paper to offer perspective on the model components described. The biofilm (reactor) model will be presented elsewhere.

A BIOFILM MODEL TO DESCRIBE SELENIUM REMOVAL
The selenium biofilm model describes the simultaneous diffusion, chemical and biochemical conversion of multiple substrates and ions, respectively, in a mixed-culture one-dimensional biofilm. The mathematical description of this biofilm model is based on the general dynamic one-dimensional biofilm model of Wanner and Gujer (1985) and Wanner and Reichert (1995).
Describing the platform for executing simulations is outside the scope of this paper. The biofilm model is structured in (Peterson) matrix notation. Two matrices exists describing (1) chemical and (2) biochemical processes, (state) variables, stoichiometry, and rate expressions. Appendix A contains a biological process, (state) variable, stoichiometric, and rate matrix.

State Variables
State variables are categorized into soluble (S) and particulate (X) matter similar to the Activated Sludge Models (ASMs) (see Henze et al. 2000). Table 1 lists state variables used in the biofilm model. Variables (and processes) related to EPS production and transformations are not included in the biofilm, but the precipitation of selenium and sulfur by SRB is modeled.

Inorganic Carbon: Carbon Dioxide, Bicarbonate, Carbonate
Ions are explicitly accounted for in the biofilm model. The net molar sum of ions at any point inside the biofilm are assumed to equal zero, and the net molar flux of ions entering and leaving a biofilm layer is assumed zero to maintain overall electroneutrality (Robinson and Stokes 1959). Three different inorganic carbon species are considered: carbon dioxide (SCO2), bicarbonate (SICO3), and carbonate (SCO3). Carbon dioxide and bicarbonate are used as a carbon and energy source by chemoaerotrophic organisms (Goldman 1999). The three inorganic carbon species arise from their acid-base equilibria. Carbon dioxide is the predominant inorganic carbon species when the pH is below 6.3, bicarbonate is predominant when pH is in the range 6.3 and 10.3, and carbonate is predominant when the pH is greater than 10.3 (Snoeyink and Jenkins 1990).

Nitrogenous Compounds: Ammonia, Ammonium, Nitrate, and Nitrogen Gas
Four nitrogenous compounds are considered: ammonia (SNH3), the ammonium ion (SNH4), the nitrate ion (SN03), and soluble nitrogen gas (SN2). Ammonia is used in this model only as a macronutrient. Ammonia is the primary (macronutrient) nitrogen source for the autotrophic denitrifying organisms. However, in the absence of ammonia, the autotrophic denitrifiers are capable of assimilative denitrification whereby nitrate is transformed to make nitrogen available (to the autotrophic denitrifiers) as the macronutrient nitrogen. In addition, under anoxic conditions nitrate is reacted upon by the autotrophic denitrifying organisms where it is ultimately transformed into nitrogen gas. Finally, ammonia is released after the death, decay and lysis of microbial matter. The ammonia ion results from pH-dependent acid-base equilibrium of dissolved ammonia. The ammonium ion is predominant at pH values below nine while ammonia is predominant at greater pH values. The nitrate ion is used as the electron acceptor by (non-methanol degrading) denitrifying heterotrophs.

Hydrogen Ion, Cations, and Anions
The hydrogen ion (SH) is included in the selenium biofilm model to calculate pH. The pH is calculated algebraically from a later described charge balance. Cations (Scat) and anions (San) are non-reactive, but are considered in the model for charge compensation in the event data describing cations such as sodium (Na+) and magnesium (Mg2+) or anions such as chloride (Cl-) and sulfate (SO42-) are available.

Sulfate Reducing Bacteria
Sulfate reducing bacteria are a phylogenetically and physiologically diverse bacterial group which is characterized by their capacity to conserve energy for growth by linking the oxidation of different substrates to the dissimilatory reduction of sulfate to sulfide. Six types of sulfate
reducing bacteria and are considered in the selenium model including propionate-degrading SRB ($X_{SRB,P}$), acetotrophic SRB ($X_{SRB,A}$), and hydrogenotrophic SRB ($X_{SRB,H}$).

Selenium-based substrates are described here because sulfate reducing bacteria are primarily responsible for the biochemical transformations of selenium-based soluble substrates such as selenate ($S_{SeO_4^{2-}}$) and selenite ($S_{SeO_3^{2-}}$). Finally, the insoluble material elemental selenium ($Se^0$) is considered.

Selenium exists in both soluble and particulate forms, and as a variety of compounds. In addition, selenium speciation varies and is dependent on environmental conditions, both chemical and physical. The species can be grouped into four major categories that include (1) inorganic selenium, (2) volatile and methylated selenium, (3) protein and amino-acid selenium, and (4) non-protein amino acids and biochemical intermediates. Selenium typically occurs in the environment in one of four redox states, namely Se(VI), Se(IV), Se(0), and Se(-II). Se(VI) and Se(IV) occur as the oxyanions selenate ($SeO_4^{2-}$) and selenite ($SeO_3^{2-}$). These oxyanions are commonly encountered in aerobic environments, while under anaerobic conditions the insoluble, elemental selenium ($Se^0$) appears to predominate (Hockin and Gadd 2006). Selenium is assimilated by micro-organisms as selenide ($Se^{2-}$), while a wide variety of micro-organisms, including sulfate reducing bacteria (SRB), can liberate volatile, methylated species (Michalke et al. 2000). Reduction to elemental selenium is more widespread amongst micro-organisms and some bacteria are able to carry out dissimilatory reduction of selenium oxyanions (Lovley 1993). No evidence exists that SRB can grow by dissimilatory selenium reduction, but the potential of SRB to carry out reductive selenium oxyanion transformations has been demonstrated by Tucker et al. (1998), amongst others.

**Other Soluble and Particulate State Variables**

The state variables for oxygen ($S_{O_2}$), phosphorus ($S_{PO4}$), fermentable substrates ($S_{F}$), fermentation products (acetate) ($S_{A}$), soluble inerts ($S_{I}$), non-diffusible slowly biodegradable substrate ($X_{S}$), heterotrophic biomass ($X_{H}$), chemoautotrophic biomass (or nitrifiers) ($X_{A}$), total suspended solids ($X_{TSS}$), and particulate inerts ($X_{I}$) are analogous with state variables used in Activated Sludge Model No. 2d (Henze et al. 2000). The reader is referred to Henze et al. (2000) for a definition and explanation of these well-known model variables.

The aerobic and anoxic decay product state variable ($X_{daa}$) was introduced by Johnson et al. (2008). This variable accounts for the separation of microbial decay products from the inert particulates ($X_{I}$) into a separate particulate decay product for aerobic and anoxic growth of microorganisms.
<table>
<thead>
<tr>
<th>Index i</th>
<th>Variable</th>
<th>Definition</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Soluble Components</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>$S_{CO2}$</td>
<td>Carbon Dioxide (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>2</td>
<td>$S_{HCO3}$</td>
<td>Bicarbonate (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>3</td>
<td>$S_{CO3}$</td>
<td>Carbonate (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>4</td>
<td>$S_{NH4}$</td>
<td>Ammonium (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>5*</td>
<td>$S_{NH3}$</td>
<td>Ammonia</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>6</td>
<td>$S_{NO3}$</td>
<td>Nitrate (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>7</td>
<td>$S_{N2}$</td>
<td>Nitrogen Gas</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>8</td>
<td>$S_H$</td>
<td>Hydrogen (ion)</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>9*</td>
<td>$S_{an}$</td>
<td>Anions</td>
<td>mol m$^{-3}$</td>
</tr>
<tr>
<td>10*</td>
<td>$S_{cat}$</td>
<td>Cations</td>
<td>mol m$^{-3}$</td>
</tr>
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<td>11</td>
<td>$S_{O2}$</td>
<td>Dissolved Oxygen</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>12</td>
<td>$S_{PO4}$</td>
<td>Ortho-Phosphorus</td>
<td>(mol or g COD) m$^{-3}$</td>
</tr>
<tr>
<td>13</td>
<td>$S_{F}$</td>
<td>Fermentable Substrate</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>14</td>
<td>$S_A$</td>
<td>Fermentation Products, Volatile Fatty Acids (Acetate)</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>15</td>
<td>$S_I$</td>
<td>Inert Organic Compounds</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>16</td>
<td>$S_{SeO4}$</td>
<td>Selenate</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>17</td>
<td>$S_{SeO3}$</td>
<td>Selenite</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>18</td>
<td>$S_{Se}$</td>
<td>Elemental Selenium</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>19</td>
<td>$S_{Se2-}$</td>
<td>Selenide</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>20</td>
<td>$S_{SO4}$</td>
<td>Sulfate</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>21</td>
<td>$S_{H2S}$</td>
<td>Hydrogen sulfide</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td><strong>Particulate Components</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>22</td>
<td>$X_S$</td>
<td>Slowly Biodegradable Substrates</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>23</td>
<td>$X_H$</td>
<td>Facultative Heterotrophic Biomass</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>24</td>
<td>$X_A$</td>
<td>Autotrophic Nitrifying Biomass</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>25</td>
<td>$X_{TSS}$</td>
<td>Total Suspended Solids</td>
<td>g TSS m$^{-3}$</td>
</tr>
<tr>
<td>26</td>
<td>$X_I$</td>
<td>Inert Organic Compounds</td>
<td>g COD m$^{-3}$</td>
</tr>
<tr>
<td>27</td>
<td>$X_{daa}$</td>
<td>Aerobic/Anoxic Decay Products</td>
<td>g COD m$^{-3}$</td>
</tr>
</tbody>
</table>

Note: * affixed to index 5, 9, and 10 indicate that the state variable is not directly used in the biological process model. Rather, these state variables are used in the chemical process simulation, which determines substrate speciation resulting from environmental conditions.
Processes
In order to capture the essential mechanisms driving conversion processes by selenium removing biofilms, the biofilm model takes into account biochemical and chemical conversion processes.

Biochemical Processes
Table 2 lists descriptions of each biochemical process simulated in the biofilm model. Table 3 is the biochemical process, stoichiometric, and transformation rate-expression (Peterson) matrix. Biochemical transformation rate expressions will be listed in the technical presentation.

The processes describing facultative heterotrophic organism activity are similar to the processes in Activated Sludge Model No. 2d. Relevant processes consider the biochemical transformation of organic matter and nitrogenous compounds. Influent non-diffusible slowly biodegradable organic matter (X_S) is attached onto the biofilm surface and is either (1) hydrolyzed under aerobic, anoxic, or anaerobic conditions or (2) exits the biofilm matrix with detached biofilm fragments. The slowly biodegradable organic matter may be hydrolyzed into fermentable substrate (S_F), which can be fermented under anaerobic conditions into volatile fatty acids (S_A). Soluble fermentable substrates and products may exist in the influent wastewater stream. The fermentable substrates and fermentation products can be consumed for the growth of facultative heterotrophic organisms under anoxic conditions with nitrate as the electron acceptor. Nitrate reduction produces nitrogen gas (S_N2). For simplicity, it is assumed that autotrophic denitrifying organisms convert nitrate as the electron acceptor and use a sulfurous compound (which usually results from the biochemical transformation of hydrogen sulfide) as the electron donor for energy production.

Ammonia (S_NH3) and phosphorus (S.PO4) are used for cell synthesis. Death-decay-regeneration of the biomasses produces ammonia and phosphorus. Additionally, death-decay-regeneration results in two distinct fractions of particulates. One fraction enters with the influent wastewater while the other fraction originates from cellular decay products. The inert particles in the influent wastewater stream include material such as cellulose fibers and hair, but very few microbial decay products. Johnson et al. (2008) hypothesized that the decay products resulting from (non-methanol degrading) heterotrophic organisms grown under aerobic and anoxic conditions (X_daa) are actually slowly biodegradable (i.e., X_S) under anaerobic conditions due to the fact that the decay products primarily consist of cell wall material.

The anaerobic conversion processes by sulfate reducing bacteria are described with the process model of Poinapen and Ekama (2010)
<table>
<thead>
<tr>
<th>Index i</th>
<th>Process Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydrolysis</td>
<td></td>
</tr>
<tr>
<td>1.</td>
<td>Aerobic</td>
</tr>
<tr>
<td>2.</td>
<td>Anoxic</td>
</tr>
<tr>
<td>3.</td>
<td>Anaerobic</td>
</tr>
<tr>
<td>Sulfur Reducing Bacteria: X_{SRB}</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>Growth of Propionate-Degrading SRB</td>
</tr>
<tr>
<td>5.</td>
<td>Endogenous Decay of Propionate-Degrading SRB</td>
</tr>
<tr>
<td>6.</td>
<td>Growth of Acetotrophic SRB</td>
</tr>
<tr>
<td>7.</td>
<td>Endogenous Decay of Acetotrophic SRB</td>
</tr>
<tr>
<td>8.</td>
<td>Growth of Hydrogenotrophic SRB</td>
</tr>
<tr>
<td>9.</td>
<td>Endogenous Decay of Hydrogenotrophic SRB</td>
</tr>
<tr>
<td>10.</td>
<td>Selenate Uptake by SRB</td>
</tr>
<tr>
<td>11.</td>
<td>Selenite Uptake by SRB</td>
</tr>
<tr>
<td>Heterotrophic Organisms: X_{H}</td>
<td></td>
</tr>
<tr>
<td>12.</td>
<td>Growth on Fermentable Substrate, S_{F}</td>
</tr>
<tr>
<td>13.</td>
<td>Growth on Fermentation Products, S_{A}</td>
</tr>
<tr>
<td>14.</td>
<td>Denitrification with Fermentable Substances, S_{F}</td>
</tr>
<tr>
<td>15.</td>
<td>Denitrification with Fermentation Products, S_{A}</td>
</tr>
<tr>
<td>16.</td>
<td>Selenate Reduction with Fermentation Products, S_{F}</td>
</tr>
<tr>
<td>17.</td>
<td>Selenate Reduction with Fermentation Products, S_{A}</td>
</tr>
<tr>
<td>18.</td>
<td>Selenite Reduction with Fermentation Products, S_{F}</td>
</tr>
<tr>
<td>19.</td>
<td>Selenite Reduction with Fermentation Products, S_{A}</td>
</tr>
<tr>
<td>20.</td>
<td>Fermentation</td>
</tr>
<tr>
<td>21.</td>
<td>Lysis of X_{H}</td>
</tr>
<tr>
<td>Chemoautotrophic (Denitrifying) Organism: X_{A}</td>
<td></td>
</tr>
<tr>
<td>22.</td>
<td>Anaerobic conversion of hydrogen sulfide to sulfate, S_{H2S}</td>
</tr>
<tr>
<td>22.</td>
<td>Denitrification with Sulfate, S_{SO4}</td>
</tr>
<tr>
<td>23.</td>
<td>Lysis of X_{A}</td>
</tr>
<tr>
<td>Microbial Decay Products: X_{d33}</td>
<td></td>
</tr>
<tr>
<td>24.</td>
<td>Anaerobic Hydrolysis of Decay Products</td>
</tr>
</tbody>
</table>
Biochemical Rate Expressions

Biochemical rate expressions will be listed in the technical presentation. Due to the complexity of this model, rate expressions were still under development at the time this paper was written. Explicitly describing the kinetic expressions is an integral component of this paper’s intent, namely to create model framework that can be contemplated and thoroughly reviewed prior to integrating the process model with the dynamic one-dimensional biofilm model.

Chemical Processes

Chemical conversion processes in the selenium biofilm model include acid-base equilibria in order to calculate biofilm pH profiles, speciation, and to determine inorganic carbon availability for the growth of autotrophic denitrifying and sulfur reducing bacteria. Table 5 (after Wolf et al. 2007) is the chemical processes, stoichiometric, and rate-expression matrix. The charge balance used to calculate the concentration of protons was also presented by Wolf et al. (2007). The equations are summarized below. Equation (3) is the charge balance.

\[
S_{H} + S_{NH_4} + S_{\text{cat}} - S_{HCO_3} - S_{NO_3} - 2 \cdot S_{CO_3} - S_{OH} - S_{an} - S_{SO_4^{2-}} = 0 \tag{3}
\]

Equation (3) can be rearranged as Equation (4).

\[
\left( S_{NH_4} + S_{\text{cat}} - S_{HCO_3} - S_{NO_3} - 2 \cdot S_{CO_3} - S_{an} - S_{SO_4^{2-}} \right) = \sum_i S_{i,\text{ion}} \tag{4}
\]

The pH is calculated with Equation (5).

\[
S_{OH} = \frac{K_{a,H_2O}}{S_{H}} \tag{5}
\]

Substituting Equations (4) and (5) into Equation (3), and rearranging results in Equation (6) for the calculation of pH.

\[
S_{H} = 0.5 \cdot \left( \sqrt{\sum_i S_{i,\text{ion}}}^2 + 4 \cdot K_{a,H_2O} - \sum_i S_{i,\text{ion}} \right) \tag{6}
\]
The technical presentation will contain a table similar to Table 3 that describes sulfide system chemistry. The chemical model will describe methodology for the dissociation of hydrogen sulfide to sulfate, and will account for the collective impact that carbon-sulfide chemistry will have on pH, and, therefore, substrate speciation (e.g., ammonia/ammonium).

**Parameter Definitions**

Parameter values for autotrophic denitrifiers and sulfate reducing bacteria do not exist with the support of a substantial data base. Therefore, related biokinetic parameter values will be developed using a combination of laboratory testing and parameter estimation by electron counts. This information will require evaluation/calibration efforts using the functional dynamic one-dimensional biofilm model. Therefore, these parameter values and methodology for establishing the values will be presented at a later time. Establishing this information within the confines of this manuscript is outside the scope of this investigation. Parameter values for facultative heterotrophic biomass may be estimated as the values used for ASM 2d (Henze et al. 2000). However, defining parameter values is outside the scope of the present paper. The development of conversion factors and values are the subject of ongoing work.

**DISCUSSION**

There is a paucity of mechanically-based process design models for biofilm reactors meant to remove selenium from industrial wastewaters. The information presented in this paper develops framework for a more rigorous and mechanistic design model to describe biofilm reactors for the treatment of selenium-laden industrial wastewater.
Biofilm (Reactors) System for Selenium Removals – Practical Applications

Selenium can be a serious environmental concern. Selenium is required in trace amounts to sustain a majority of organisms. However, despite its nutritional purposes, concentrations only slightly greater than the required amount can be hazardous. Selenium contamination originates from several sources including mining operations, mineral processing, abandoned mine sites, petroleum processing, and agricultural run-off (EPA 2001). Contamination may be of a low-level, high volume nature which makes most treatment options expensive. Therefore, refined design criteria and a mechanistic model capable of optimizing the design of biological selenium removal processes are warranted. Figure 1 illustrates process flow diagrams of biofilm reactors applied for the biological reduction of oxidized nitrogen, sulfur, and selenium, namely the fluidized bed biofilm reactor and granular carbon based media filer.

Future Work

Additional research is required to properly define parameter values for the autotrophic denitrifying biomass and sulfate reducing bacteria component of this biofilm model. Future work includes the development of an additional chemical model that accounts for sulfide system chemistry and its impact on pH (and therefore substrate speciation). Once this framework has been properly reviewed, it will be incorporated with a dynamic one-dimensional biofilm model. The biofilm model will require verification. It is the recommendation of the authors that pilot- and full-scale systems be thoroughly investigated to (1) further develop basic design criteria, and (2) generate the in-depth information that is required to evaluate and verify mechanistic design models such as this biofilm (reactor model).

CONCLUSIONS

Biofilm reactors are an emerging environmental biotechnology used for the removal of selenium from industrial wastewater. Bioreactor models for selenium removal that are based on the mechanistic description of biofilms are few in number, but a dynamic model that is based on mechanistic principles (chemical and biochemical) and is capable of describing selenium removal is needed for efficient and more rigorous process design and optimization. This paper summarizes variables, processes, and rate expressions for both chemical and biochemical conversions inside a mixed-culture biofilm containing facultative heterotrophs, sulfate reducing bacteria, and autotrophic denitrifiers. Chemical and biochemical processes in the biofilm model are based on theoretical considerations. Chemical processes include acid-base equilibria reactions primarily to describe substrate. The proposed mathematical description of our multispecies selenium removal biofilm is based on the general one-dimensional model of Wanner and Gujer (1985) and Wanner and Reichert (1995). It is assumed that ions do not undergo chemical or biological transformation, but the ions do not maintain a constant concentration profile across the one-dimensional biofilm. State variables, processes, and kinetic expressions in the biofilm model are structured using (Peterson) matrix notation. The framework for a mechanistic mathematical model describing a dynamic one-dimensional biofilm has been developed and presented.
REFERENCES


### APPENDIX A: PROCESS, STOICHIOMETRIC, AND KINETIC MATRIX

| Index | \( S_{CO} \) | \( S_{NO3} \) | \( S_{NH3} \) | \( S_{PO4} \) | \( S_{F} \) | \( S_{A} \) | \( S_{Se} \) | \( X_{S} \) | \( X_{H} \) | \( X_{A} \) | \( X_{TSS} \) | \( X_{Im} \) | \( X_{SRB,P} \) | \( X_{SRB,A} \) | \( X_{SRB,H} \) | Rate |
|-------|---------------|---------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------------|-------|
| 1.    | 1  | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 2.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 3.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 4.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 5.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 6.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 7.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 8.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 9.    | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 10.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 11.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 12.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 13.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 14.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 15.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 16.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 17.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 18.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 19.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 20.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 21.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 22.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 23.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 24.   | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |

- Actually consists of three state variables: selenate (\( SeO_4^{2-} \)), selenite (\( SeO_3^2- \)), and elemental selenium (\( Se^{0} \))
- Two additional columns for the state variables hydrogen sulfide (\( H_2S \)) and sulfate (\( SO_4^{2-} \))