Supporting Information

for

Immobilization of Selenite via Two Parallel Pathways during *In-situ* Bioremediation

Youneng Tang[*, §, **], Charles J. Werth[*, †], Robert A. Sanford[‡], Rajveer Singh[*], Kyle Michelson[*], Masaru Nobu[*], Wen-Tso Liu[*], Albert J. Valocchi[*]

* Department of Civil and Environmental Engineering, University of Illinois at Urbana-Champaign

† Department of Civil, Architectural, and Environmental Engineering, University of Texas at Austin

‡ Department of Geology, University of Illinois at Urbana-Champaign

§ Department of Civil and Environmental Engineering, FAMU-FSU College of Engineering, Florida State University

** Corresponding author: 2525 Pottsdamer Street, Building A, Suite A130 Tallahassee, FL 32310; (850)4106119; ytang2@fsu.edu

The supporting information contains 1 table (Table S1. Reaction rates in the model), 4 figures (Figure S1. Biomass distribution in the micro-fluid flow cell after inoculation; Figure S2. Representative EDS spectra of the two solid chemical species; Figure S3. Simulated concentration of dissolved chemical species on the 40^{th} day; Figure S4. Distribution of reaction products at the end of the mixing zone on the 40^{th} day of flow cell operation), and a section for the mathematical model (Initial conditions, boundary conditions, and numerical solutions).

Table S1. Reaction rates in the model

Reaction (j)	Coefficient of species i, n, and m in reaction j ($\eta_{i,j}, \eta_{n,j}, \eta_{m,j}$)									Reaction rate (r_j)
	Biomass species n		Solid chemical species m		Dissolved chemical species i					
	SRB (n = 1)	SeRB (n = 2)	Se_nS_{8-n} (m = 1)	Se^0 (m = 2)	$CH_3CH_2COO^-$ (i = 1)	SeO_3^{2-} (i = 2)	SO_4^{2-} (i = 3)	HS^- (i = 4)	CH_3COO^- (i = 5)	
j = 1	Y_1				-1		-0.75	0.75	1	$k_{\max,1,1}\varepsilon_1 X \frac{C_1}{K_1+C_1}\frac{C_3}{K_3+C_3}$
j = 2	Y_2						-1	1	-1	$k_{\max,5,2}\varepsilon_1 X \frac{C_5}{K_5+C_5}\frac{C_3}{K_3+C_3}$
j = 3		Y_3		1.5	-1	-1.5			1	$k_{\max,1,3}\varepsilon_2 X \frac{C_1}{K_1+C_1}\frac{C_2}{K_2+C_2}$
j = 4		Y_4		2		-2			-1	$k_{\max,5,4}\varepsilon_2 X \frac{C_5}{K_5+C_5}\frac{C_2}{K_2+C_2}$
j = 5			3/8			-1		-2		$k_{SeS}C_2C_4$
summed rate	$\boldsymbol{R_{X,n}} = \sum_j \eta_{n,j} \times r_j$		$\boldsymbol{R_{S,m}} = \sum_j \eta_{m,j} \times r_j$			$\boldsymbol{R_{D,i}} = \sum_j \eta_{i,j} \times r_j$				

Note: Parameters used in this table are defined in Tables 1 and 2.

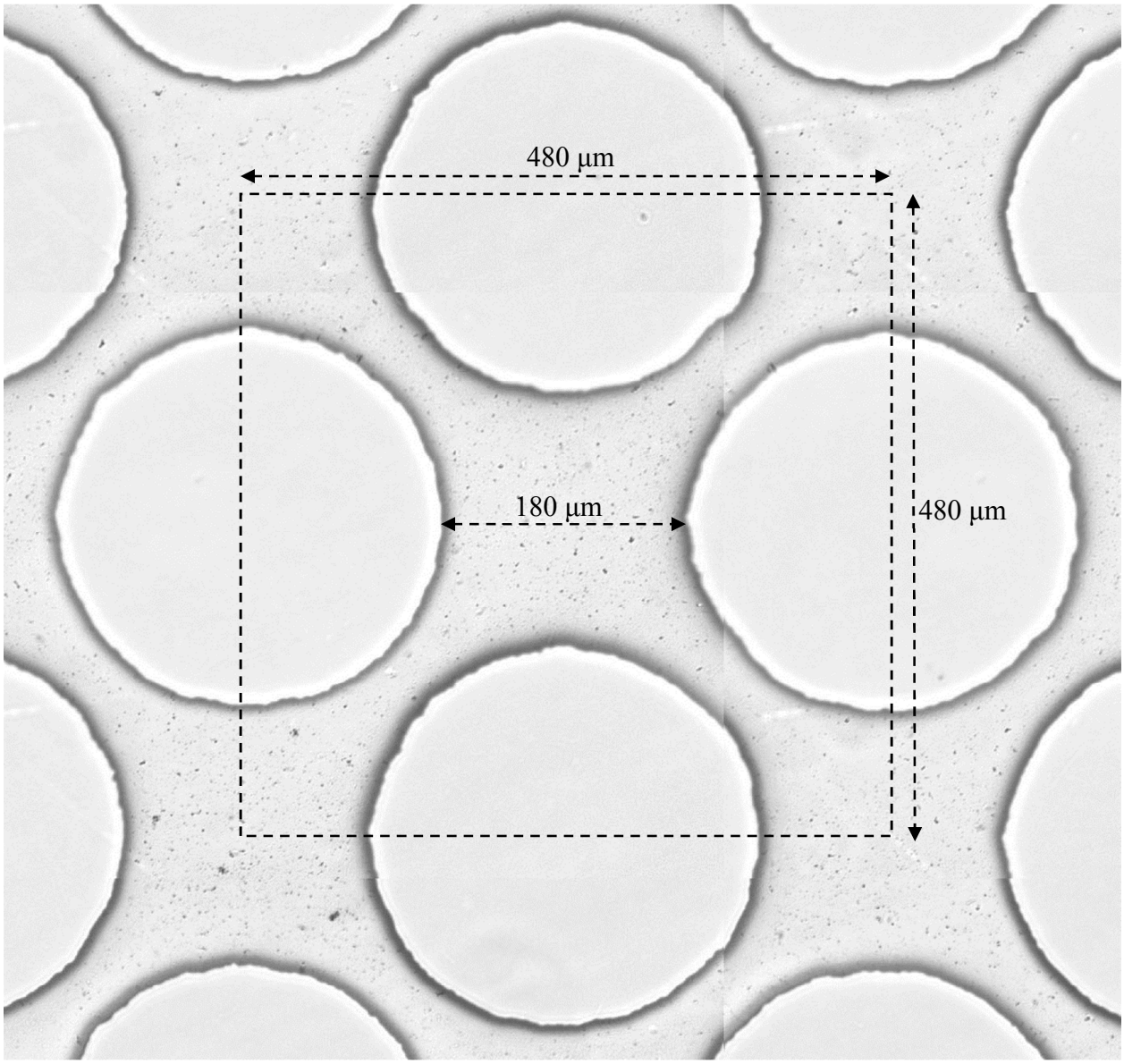
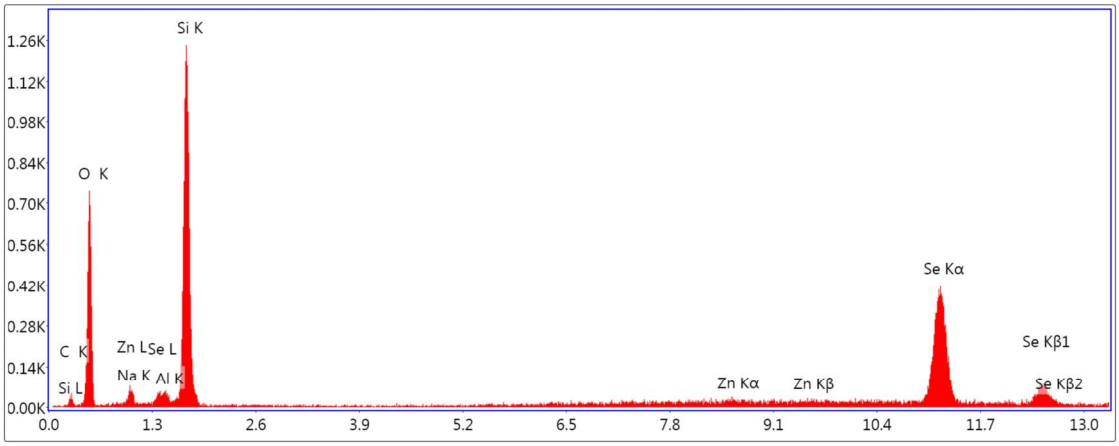
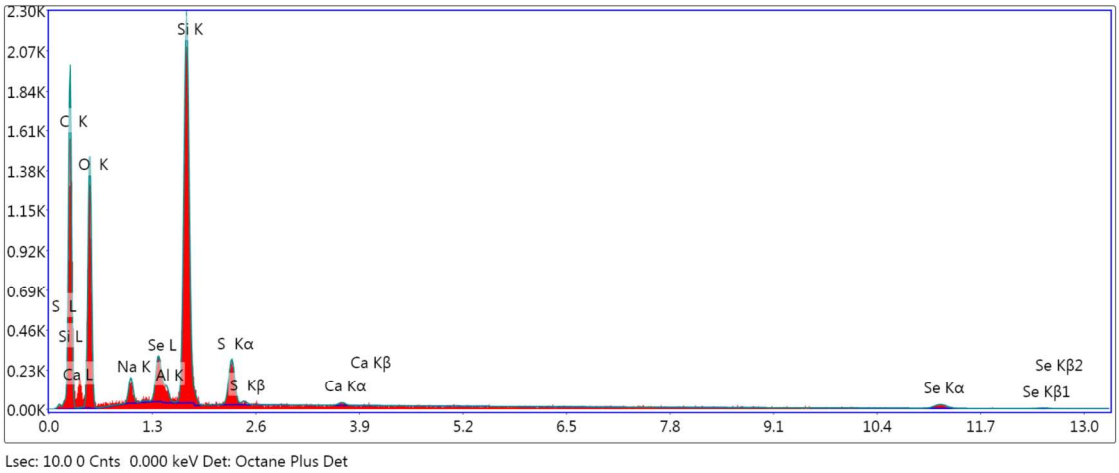


Figure S1. Biomass distribution in the micro-fluid flow cell after inoculation. The square represents a unit grid cell grid for numerical solution.



a) EDS spectrum for the elemental selenium particles in Figure 7a



b) EDS spectrum for the crystal of selenium sulfides in Figure 7b. Atomic ratio: Se:S = 0.55.

Figure S2. Representative EDS spectra of the two solid chemical species. The C and O peaks correspond to biomass; the Si peak corresponds to the bottom of the flow cell; and the Na, Al, and Ca peaks correspond to the Pyrex cover of the flow cell.

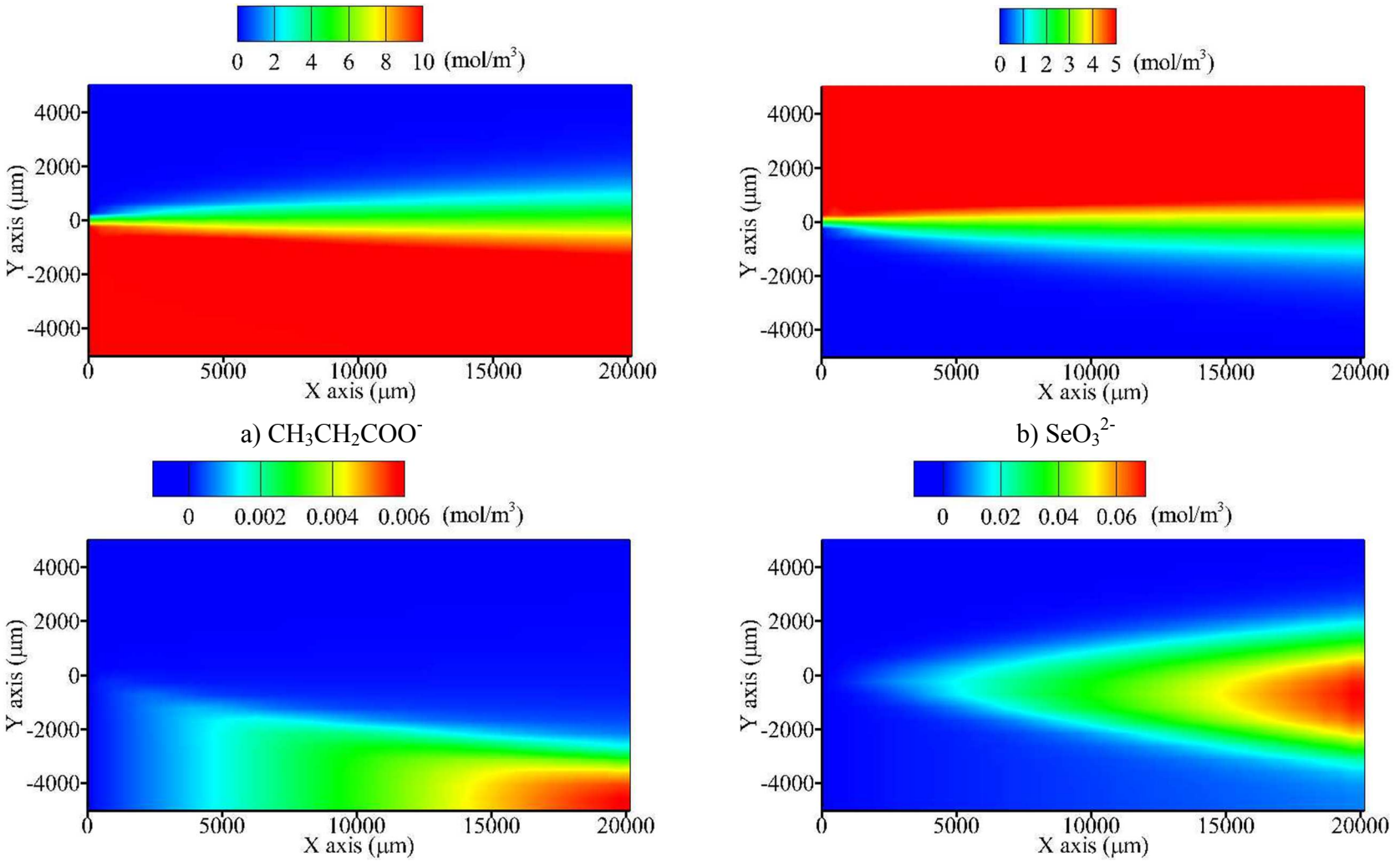


Figure S3. Simulated concentration of dissolved chemical species on the 40th day. The simulated SO_4^{2-} concentration is ~0.1 mol/L everywhere in the flow cell.

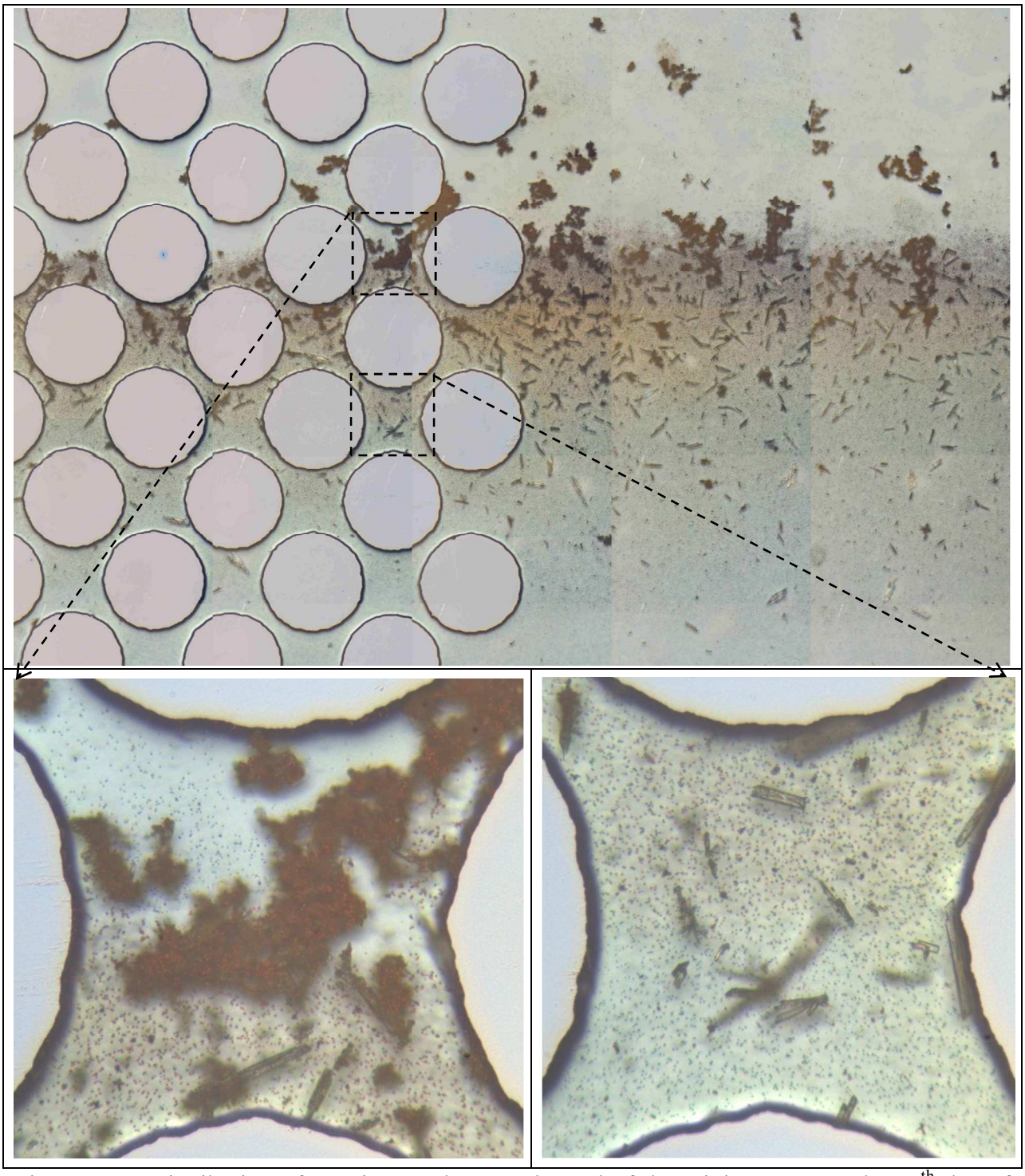


Figure S4. Distribution of reaction products at the end of the mixing zone on the 40th day of flow cell operation. a) Color image of the pore area; b) Representative reaction products on the selenite-rich side of the mixing zone, dominated by red particles; c) Representative reaction products on the propionate-rich side of the mixing zone, dominated by long crystals.

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Initial conditions, boundary conditions, and numerical solutions

Initial conditions

The initial concentrations for all dissolved and solid chemical species are zero, and the initial concentrations of biomass species (after inoculation and enrichment) are estimated by observing that there are approximately 500 biofilms in each unit grid cell (480 μm × 480 μm × 20 μm (flow cell thickness) in Figure S1), each biofilm has dimensions of 3 μm × 3 μm × 3 μm. We assume that the initial volume fraction of either biomass species is 50%. Therefore, the initial biomass concentration for both species is: porosity × (3.0 μm × 3.0 μm × 3.0 μm) × 500 × (42 × 21)/(20.0 μm × (1.0×10^4) μm × (2.0×10^4) μm) × X × 50%, in which, (42 × 21) is the number of unit grid cells in the porous media of the flow cell and (20.0 μm × (1.0×10^4) μm × (2.0×10^4) μm) are the dimensions of the porous media.

Boundary conditions

The concentrations of dissolved chemical species at inlets A and B are constant: $C_{D,1} = 0$, $C_{D,2} = C_{in,2}$, $C_{D,3} = C_{in,3}$, $C_{D,4} = 0$, $C_{D,5} = 0$ for inlet A and $C_{D,1} = C_{in,1}$, $C_{D,2} = 0$, $C_{D,3} = C_{in,3}$, $C_{D,4} = 0$, $C_{D,5} = 0$ for inlet B. The concentration gradient of dissolved chemical species at the outlet boundary and the two side walls is zero: $\frac{\partial C_{D,i}}{\partial n} = 0$, where n is the unit normal direction of the boundary.

Numerical solutions

This problem includes two dynamic processes, which occur at very different time scales: biofilm development on the order of hours or days; chemical species transport and reaction on the order of seconds or minutes.[1] Therefore, equations for these two processes can be decoupled and solved sequentially. We used two time steps: 0.25 day for biofilm development (Equation 7) and 5.0×10^{-8} day for chemical species transport and reaction (Equation 6 and 8). When we solved Equations 6 and 8 using a simple explicit-in-time finite difference method during a 0.25-day period, we assumed that the biofilm was "frozen", meaning its concentration and composition was fixed. We updated the biofilm information after each 0.25-day period by solving Equation 7 using the finite difference method. The grid size is 480 μm × 480 μm, which are the dimensions of a unit grid cell for the porous media. These numerical strategies were widely used in previous similar work,[1, 2] and were confirmed by numerical tests such as grid and time-step refinement exercises.

REFERENCES

(1) Picioreanu, C.; van Loosdrecht, M. C. M.; Heijnen, J. J. Effect of diffusive and convective substrate transport on biofilm structure formation: A two-dimensional modeling study. *Biotechnol. Bioeng.*, **2000**, 69(5), 504-515.

(2) Tang, Y.; Valocchi, A.; Werth, C. A hybrid pore-scale and continuum-scale model for solute diffusion, reaction and biofilm development in porous media. *Water Resour. Res.*, **2014**, submitted.