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MODELING THE ROLE OF ACTIVE BIOMASS ON FATE AND TRANSPORT OF A HEAVY METAL IN THE PRESENCE OF ROOT EXUDATES

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ABSTRACT

The influence of active biomass in immobilizing heavy metals in the soil rhizosphere is investigated through mechanistic models. The movement of water in the soil is modeled using Richards equation. An advection-dispersion equation, with a sink term for metal uptake by biomass, is used for modeling the fate and transport of lead. This sink term represents the nonlinear kinetics of metal adsorption to the biomass that is partitioned into mobile and stationary fractions within the soil. Transport of the mobile biomass fraction is modeled by an advection-dispersion equation, with a source term that is based on Monod growth kinetics, and a linear sink term for endogenous decay. The movement of metal in association with mobile biomass is also included as a transport mechanism for lead. Root exudates serve as carbon substrate for the biomass growth, and their transport is modeled in a similar way as that of the biomass. A hypothetical one-dimensional vertical soil column containing metal, biomass and a carbon substrate is used for analyzing lead movement. Model simulations demonstrate the influence of water content, growth rate of biomass, partitioning coefficient of biomass between soil and aqueous phase, and partitioning coefficient of metal between biomass and aqueous phase of the soil on fate and transport of lead. The extent of immobilization of lead in soil is found to be dependent on the growth of biomass, which in turn depends on the availability of root exudates in the rhizosphere. Apart from analyzing different scenarios, such a model can be used for designing future experiments.

Key words: heavy metals, rhizosphere, mathematical modeling, kinetics, biomass

INTRODUCTION

Heavy metal contamination of ground water poses a serious environmental concern. Common sources of contamination of heavy metals include smelter emissions, chat piles or mine tailings, active mining areas, and industrial sites. Remediation of metal-contaminated sites can be achieved by first immobilizing heavy metals in soil, thus preventing them from reaching the water table. Pierzynski et al. (1994) and Lambert et al. (1997) discuss different ways by which metals can be immobilized in soils. They suggest soil amendments, such as phosphates, for this purpose. Certain plant species called hyper-accumulators can concentrate significant amounts of metals in their root-shoot system. The root exudates released by plants can bind with metals and increase dissolution from their parent minerals in soils. The different processes by which metals can be distributed among soil constituents are illustrated in Figure 1.

The United States Environmental Protection Agency issued criteria that would restrict the bioavailable forms of metals in streams (Hall et al., 1992). Thus, it is important to understand how microbial activity in soils influence the leaching of metals into ground water. For instance, the ability of algae and bacteria to sequester heavy metals from dilute aqueous solutions and accumulate them in their cell structure was recognized by Ting et al. (1989). This has potential for heavy metal immobilization (Baath, 1989). The physico-chemical mechanisms involved in biosorption are adsorption to the cell and

ion exchange. If living biomass is involved, metal immobilization occurs due to active metabolic uptake also (Gadd and White, 1993). It is believed that algae are the most tolerant among the various species that can immobilize metals, followed by fungi, bacteria, and actinomycetes (Hiroki, 1992). Lead and cadmium have been effectively segregated from aqueous solutions by the dried biomass of brown marine algae. Volesky and Holan (1995) conducted an extensive review of the literature relating to heavy metal adsorption to biomass. They emphasized the need to understand the fundamental mechanisms for metal binding by specific strains of fungi, algae, and bacteria. A model that can predict the simultaneous sorption of Cd, Cu, and Zn by protonated *Sargassum fluitans* biomass was developed using the theory of multi-metal ion exchange (Schiewer and Volesky, 1996). An equilibrium isotherm model was used to describe the adsorption of cadmium by a certain filamentous cyano bacterium, *Tolypothrix tenius* (Inthorn et al., 1996). Sorption of Cu, Zn, and Ni by a filamentous bacterium, *Thiothrix Strain A1*, was modeled using Freundlich and biphasic Freundlich isotherms by Shuttleworth and Unz (1993). The authors have shown that metal uptake by the biomass is reduced by strong chelating agents EDTA (Ethylene diamine tetraacetic acid) and NTA (Ni-nitrilo triacetic acid). The primary mechanism of metal sorption was ion exchange. Metal uptake increased with cell growth.

Volesky and Holan (1995) support the use of selected microorganisms in the stabilization of heavy metal contaminated sites. For instance, lead is strongly accumulated by certain algal species such as *Chlorella vulgaris* that is usually present in organic acid-rich environments in soil, fermentation processes, and seaweeds of marine environments. A study by Trevors et al. (1986) has shown cadmium accumulation by algae through a dual kinetic process. Table 1 presents information on adsorption potential of different classes of biomass.

Modeling metal uptake by biomass is important for gaining insight into the influence of various physical and biochemical processes on the fate and transport of heavy metals. Transport of substrate and biomass also needs to be included while modeling metal movement in the presence of biomass. Corapcioglu and Haridas (1984) have considered the processes of advection, dispersion, growth, and Brownian motion of the biomass. A kinetic model was used to describe the bacterial deposition. The authors later extended the model to incorporate the movement of the growth-limiting substrate (Corapcioglu and Haridas, 1985). Changes in porosity due to bacterial growth are also incorporated in their formulation. Taylor and Jaffe (1990) have considered the reduction in porosity, accompanying changes in permeability, and dispersivity due to the growth of biomass in their transport model for biomass and substrate. Mass transfer between the water and biofilm phases was also included in this study. The model was calibrated and validated with experimental data. Most previous studies in the literature have dealt with movement of either metal or biomass. Though there is some experimental evidence of heavy metal adsorption to biomass in soils, attempts at modeling metal transport in soils in the presence of mobile biomass have not been reported to the best of our knowledge.

Influence of organic substrates on metal transport has been confirmed by several researchers. Waters (1992) studied the effect of these substrates (succinic and formic acids) on leaching of Zn from a heavy metal contaminated soil found near Galena, Kansas. It was observed that columns receiving formic acid released less Zn when compared to control columns with no formic acid. Microbial degradation of formic acid may have resulted in enhanced microbial activity. It was concluded that Zn was microbially immobilized in soil resulting in reduced concentrations in the leachate. He (1997) investigated the influence of citric and salicylic acids on the transport of Zn and Pb. Complete degradation of the substrate was observed in all the columns receiving citric acid. Carbon dioxide, microbial biomass, and acetate were the main products of citrate degradation. These columns showed less Zn and Pb in the leachate, and more Zn and Pb in carbonate and organic fractions of the soil. Large quantities of carbon dioxide and microbial biomass produced during citrate decomposition significantly enhanced metal retention in soil columns through biosorption and metal-carbonate precipitation. Availability of these acids in field soils depends on their transport characteristics within the soil. Hence, it is important to consider the fate and transport of both the substrate and biomass when evaluating metal leaching through soil.

In this study, the movement of heavy metal in a soil column, maintained under unsaturated conditions, is modeled by considering metal uptake by biomass. The biomass is partitioned into mobile and immobile fractions. The mobile fraction of the biomass undergoes transport similar to that of the heavy metal. Biomass grows in the presence of substrate, the concentration of which is modeled similar to that of biomass. Monod kinetics are assumed for modeling the growth of the biomass. Trevors et al. (1986) observed that cadmium uptake by algae was a dual kinetic process. Ting et al. (1989) used such a conceptual model to quantify sorption of the metal to biomass in dilute aqueous systems. Two distinct mechanisms of biochemical uptake are conceptualized. Firstly, certain organic chemicals such as polysaccharides, amino and phosphoryl groups, and acetamido groups present on the surface of microbial cells have a strong tendency to adsorb metals. This process, known as passive uptake, takes place very rapidly and hence is modeled as an equilibrium reaction. In this case, metal attached to the biomass surface is assumed to be in equilibrium with the metal in the soil solution. Secondly, the metal on the cell surface subsequently moves into the interior of the cell by a transport process that can only be performed by live cells. This process, known as active uptake, is modeled as a kinetic process.

MATHEMATICAL MODELING

The entire model is comprised of three parts. In the first part, Richards equation is solved to simulate the movement of water in the unsaturated zone of the column. In the second part, an advection-dispersion equation is solved to obtain the concentrations of biomass, substrate, and metal throughout the column. In the last part, the sink/source terms in the transport equations, such as growth/decay of biomass and uptake of metal by active biomass are modeled by incorporating micro-

bial kinetics. The concentrations of biomass, substrate, and metal are, therefore, influenced by growth/decay of biomass and metal adsorption and uptake by biomass.

Water Movement in Unsaturated Zone

The flow model is formulated using Richards equation for unsaturated movement of water in one dimension as in Govindaraju and Kavvas (1993). The equations governing vertical water movement are

$$v(z) = -K\left(\frac{\partial \Psi}{\partial Z} - 1\right) \tag{1}$$

$$\frac{\partial \theta}{\partial t} = \frac{\partial v(z)}{\partial z} \tag{2}$$

where q is the water content by volume (m^3/m^3); v(z) is the Darcian water flux; (m/hr) in the z direction; K is the hydraulic conductivity (m/hr); Y(<0) is the soil matric potential (m); z is the vertical coordinate direction taken positive downward; and t is time (hr). The two equations can be combined to yield Richards equation, in terms of q as

$$\frac{\partial \theta}{\partial t} = \frac{\partial}{\partial z}\left[K(\theta)\left(\frac{\partial \psi}{\partial z} - 1\right)\right] \tag{3}$$

where the K(q) and Y (q) are represented by Brooks and Corey (1964) functional forms;

$$\frac{(\theta_S - \theta)}{\theta - \theta_0} = \left(\frac{\Psi}{\Psi_b}\right)^\lambda \tag{4}$$

and

$$\frac{K(\Psi)}{K_{Sat}} = \frac{\Psi_b^\eta}{\Psi^\eta} \tag{5}$$

Here, q_S is the saturated water content; q_0 is the residual water content; Y_b is the air entry pressure head; K_{Sat} is the saturated hydraulic conductivity; and l and h are the Brooks-Corey parameters whose values depend on the soil type.

For numerical simulation, the partial differential equation (3) is discretized in time and space using an implicit finite difference procedure. At each time step, a set of non-linear algebraic equations are formulated. These equations are solved using Newton's method to obtain the spatial distribution of

water content along the length of the column. The initial water content distribution and the water content or flux at the top of the column (inlet) have to be provided at all times. The model computes the transient water content distribution and flow field along the length of the column.

Modeling Biomass and Substrate Movement

In this work, biomass is partitioned into mobile and immobile fractions. The immobile fraction gets adsorbed to the soil. A linear relationship between the biomass adsorbed to the soil C_{sb} (mg cell dry weight/kg dry soil) and the biomass in soil solution C_b (mg cell dry weight/l of aqueous phase of soil) is assumed and is given as

$$C_{sb} = K_{db}C_b \tag{6}$$

where K_{db} is the partition coefficient for the biomass (L/kg). Movement of the mobile fraction of the biomass is modeled using an advection-dispersion equation, which is given as

$$\frac{\partial[C_b(\theta + \rho K_{db})]}{\partial t} = \frac{\partial}{\partial z}\left[\theta D_b \frac{\partial C_b}{\partial z}\right] - \frac{\partial}{\partial z}[v(z)C_b] + R_b \tag{7}$$

where ¶ is the bulk density of soil (kg of dry soil/L of total or bulk soil volume); D_b is the dispersion coefficient for the biomass (m^2/hr); and R_b is the source/sink term for the biomass. Total concentration of biomass x (mg cell dry weight/L of total or bulk soil volume) in soil at any time t and at any location z is given as

$$x = C_b(z)[\theta(z) + \rho K_{db}] \tag{8}$$

In equation (8), x is the sum of the mobile and immobile fractions of the biomass.
The sink/source term in equation (7) is given as

$$R_b = \frac{\mu_m x C_s}{K_m + C_s} - K_d x \tag{9}$$

where μ_m is the maximum specific growth rate (hr^{-1}); C_S is the dissolved organic substrate concentration (mg/L); K_m is the half-saturation constant (mg/L); and K_d is the endogenous decay constant (hr^{-1}). The equation for substrate transport is given as

$$\frac{\partial[C_s\theta]}{\partial t} = \frac{\partial}{\partial z}\left[\theta D_s \frac{\partial C_s}{\partial z}\right] - \frac{\partial}{\partial z}[v(z)C_s] + R_s \tag{10}$$

where D_s is the dispersion coeffcient for substrate, and R_s is the sink term for substrate consumption. It is given as

$$R_s = -\frac{\mu_m x C_s}{Y(K_s + C_s)} \tag{11}$$

where Y is the yield, defined as the ratio of the amount of biomass formed to the amount of substrate consumed per unit volume of rhizosphere, and K_s is the half-saturation constant.

Modeling Metal Uptake Kinetics by Microbial Biomass

Empirical models, such as Langmuir and Freundlich isotherms, are conventionally used to quantify metal adsorption to microbial biomass. These models are based on regression of experimental equilibrium data and, as such, may not provide physical insight into the rate processes involved in metal uptake by biomass. A model, that can predict the lead uptake both on the cell surface and the cell interior using cell metabolic transport processes, will adjust to transient changes in the metal concentrations more successfully than would a simpler correlation model based on equilibrium (Swift and Forcinti, 1997).

The kinetic model presented here was developed by Ting et al. (1989) and was based on the observation that metal uptake is a dual process-an initial rapid uptake (passive uptake) followed by a slower process (active uptake). During passive uptake, the metal ions adsorb onto the surface of the cells. The metal concentration in soil solution C (mol/L) is in equilibrium with the adsorbed metal concentration on the cells C_p (mol/mg cell dry weight). A linear relationship is assumed between the two, which is given as

$$C = K_p C_p \tag{12}$$

where K_p (mg/L) is the surface adsorption constant. The metal ion attached to the cell surface subsequently traverses the cell membrane with carrier molecules (E) acting as vehicles of transport as shown in Figure 2. A detailed description of this kind of carrier-mediated mechanism is given by Ting et al. (1989). The active uptake process is kinetically controlled and is given by the following rate expression:

$$\frac{d(x C_a)}{dt} = x R_1 \left[C_p - R_2 C_a \right] \tag{13}$$

Here, R_1 (hr^{-1}) and R_2 (dimensionless) are carrier rate constants; x is the concentration of the microbial cells (mg cell dry weight/L of total or bulk soil volume); and C_a is the intracellular metal concentration (mol of metal/mg cell dry weight).

A certain fraction of total metal in the soil will be attached to the mobile fraction of the biomass. The transport of the intracellular metal with the biomass needs to be modeled, and it is represented as

$$\frac{\partial [xC_a]}{\partial t} = \frac{\partial}{\partial z}\left[\theta D_b \frac{\partial (C_b C_a)}{\partial z}\right] - \frac{\partial}{\partial z}\left[v(z) C_b C_a \right] + R_{ma} \tag{14}$$

where R_{ma} is now a source term representing the amount of active uptake of metal by the biomass, and is given as

$$R_{ma} = xR_1\left[C_p - R_2 C_a \right] \tag{15}$$

Modeling Metal Transport in Presence of Active Biomass

The movement of the metal is governed by advection and dispersion. Adsorption of metal to the biomass is considered as a dual process, as discussed previously. The equation for the movement of metal is given as

$$\frac{\partial\left[C\left(\theta + \frac{x}{K_p}\right) + xC_a\right]}{\partial t} + \rho\frac{\partial C_{sm}}{\partial t} = \frac{\partial}{\partial z}\left[\theta D\frac{\partial C}{\partial z}\right] - \frac{\partial}{\partial z}\left[v(z) C\right]$$
$$+ \frac{\partial}{\partial z}\left[\theta D_b\frac{\partial C_b}{\partial z}\right]\left[\frac{C}{K_p} + C_a\right] - \frac{\partial}{\partial z}\left[v(z) C_b\right]\left[\frac{C}{K_p} + C_a\right] \tag{16}$$

where C is the concentration of the metal in the aqueous phase (mol/l of aqueous phase of soil or soil water) at any spatial location z and at any time t; D is the dispersion coefficient (m^2/hr); K_p is the partitioning coefficient representing the passive uptake by biomass; and C_{sm} is the concentration of the metal adsorbed to the soil (mol of metal /kg dry soil). Adsorption of metal to the soil is modeled using a linear isotherm as

$$C_{sm} = K_f C \tag{17}$$

where K_f (l of aqueous phase/kg dry soil) is the partitioning coefficient of metal between the aqueous phase and soil.

Total metal in the soil column at any time and at any location, C_{tm}, is given as

$$C_{tm} = \theta C + x\left(C_p + C_a\right) + \rho K_f C \tag{18}$$

Numerical Solution Strategy for Transport Equations

The equations described in the preceding section are used to compute concentrations of the metal and biomass as these undergo vertically downward movement in the unsaturated soil. An implicit-finite difference formulation was used to discretize the equations. In order to circumvent the problem of

numerical dispersion, an Eulerian-Lagrangian approach was adopted (Thomson et al., 1984). The solution procedure for the transport component was split into two steps. During the first step, only pure convection was considered. Then, the dispersion effects were included using Eulerian approach. The system of algebraic equations resulting from discretization at each time step was solved by formulating a tridiagonal system.

The solution space for the transport model described above consists of three domains: spatial, biochemical, and temporal (Walter et al., 1994). The advection-dispersion processes describing aqueous-phase transport are spanning over spatial and temporal domains only, and the biochemical equations describing the translocation of the heavy metal are spanning over the biochemical and temporal domains only. Advection-dispersion equations and biochemical equations are decoupled and solved separately. The non-linear behavior of biokinetic equations is confined to the models describing microbial growth and metal adsorption processes. Thus, the solution system at any time step starts with of a physical step in which the advective-dispersive terms of the transport equations are solved, keeping the reaction terms constant. This is followed by a biochemical step in which the metal uptake kinetics and biomass growth kinetics are solved to obtain biomass concentrations, and aqueous and bio-sorbed concentrations of the metal, at each nodal point in the spatial domain.

At each time step, the flow model is solved first yielding the water content and pore water velocity at each spatial node. Transport models for the substrate and biomass are coupled, and these are solved in an iterative fashion. This is followed by the numerical solution of the transport model for heavy metal concentrations. The nodal concentrations are treated as the total metal concentrations at this stage. The biochemical kinetic equations are then solved to partition the total heavy metal into solution phase, and into the part that is associated with the biomass. The model then proceeds to the next time step. The flowchart depicting this methodology is shown in Figure 3.

The method used in this work has been employed by other investigators (Lensing et al. 1994; Walter et al., 1994; Yeh and Tripathi, 1991; and Cederberg et al. 1985). Generally, the accuracy of the numerical solutions improves as the time step is reduced. Several different time steps were investigated and a time step of 0.2 seconds was found to yield accurate results.

RESULTS

Lead and cadmium are chosen to illustrate the metal uptake kinetics by *Chlorella vulgaris*, belonging to the algae family. Adsorption of these metals with biomass growth is demonstrated using several simulation examples. Two sets of examples were presented. The first set illustrates the kinetic uptake of lead and cadmium in a batch system. The purpose of these simulations is to illustrate the dual uptake kinetics (an initial rapid uptake and a subsequent slower uptake) of heavy metals by algae. A second set of simulation examples are provided to describe fate and transport of metal in the presence of biomass and substrate using lead as an indicating example. The parameters obtained in the batch

simulations were utilized in the transport simulations. This second set of simulations helps us understand how kinetic metal uptake of biomass influences metal transport in unsaturated soils.

Simulation Results for Batch Kinetic Studies

In this section, we are primarily interested in finding the amount of lead that adsorbs to biomass under batch conditions, using equations (12) and (13) only. The entire biomass was in suspension, due to the mixing of soil with the aqueous phase. The soil was considered saturated at a water content of 0.43. There was no water movement and metal transport under these conditions.

The surface adsorption constant 'K_p' for lead and cadmium was assumed to be proportional to the fractional surface area of the cells covered by these metals as given by Ting et al. (1991) . The estimated values for these constants are shown in Table 2. A constant biomass growth rate of 0.01 hr^{-1} (Ting et al., 1991) was used in this simulation. The carrier rate constants R_1 and R_2 are functions of temperature and the cell concentrations. These values are again based on the work of Ting et al. (1991) and are also shown in Table 2.

Figure 4 shows the microbial uptake of lead for an initial concentration of lead in solution of 1mg/L. Initial concentration of the biomass (in this case *Chlorella vulgaris*) is 800 mg/L. Rapid uptake of metal takes place under batch conditions during the first 20 hours, followed by a slower rate of uptake. The biomass concentration increases during this process due to growth. Figure 5 shows the corresponding residual metal concentration in the aqueous solution. The concentration decreases by about 50 percent within 40 hours.

Figure 6 shows the uptake kinetics of cadmium. A comparison is made between the predictions using the model of Ting et al. (1992) and the experimental observations (Kurek et al., 1982) for cadmium uptake by bacterial cells (*Serratia marcenes and paracocus species*). K_p was calculated based on fractional surface area of cells covered by cadmium as 4.2 g/L. The carrier rate constants R_1 and R_2 are provided in Table 2 (same as for lead). Initial concentration of the metal in soil solution is 10 mg/L, and the corresponding concentration of the bacterial cells is 2800 mg/L. These values are obtained from Kurek et al. (1982). Model equations (12) and (13) are solved to compute the sorbed concentrations of cadmium with time. The model results are in good agreement with the experimental data. The cadmium adsorption rate is large initially and reduces at later times of the simulation in accordance with the dual process. Also shown is the concentration of the biomass with a constant growth rate of 0.01 hr^{-1} .

Simulation Example for Metal Transport with Growing Biomass

The system chosen for transport simulations is a 2m-long hypothetical soil column with one-dimensional flow in a vertically downward direction. The soil is assumed to have a saturated hydraulic conductivity of 0.05 m/hr and a dispersion coefficient of 0.1 m^2/hr. The Brooks and Corey parameter (h) is taken as 5 and the air entry pressure head is taken as -15 cm of water. Table 2 provides a list of simulation parameters used for the simulations.

Journal of Hazardous Substance Research **9-9**

A constant initial moisture content distribution of 0.1 was chosen within the column. The boundary conditions at the top of the column were a constant water flux of 0.05 m/hr and a constant inlet metal and biomass concentration of 1 mg/L. At the bottom of the column, the concentration gradient is assumed to be zero. For results in Figures 7-13, the substrate concentration, throughout the column length for all time, was set to 40 mg/L. For Figures 14 &15, substrate transport and degradation was studied for a constant concentration of 40 mg/L maintained at the top of the column. The column was initially free of substrate. The concentration gradient for the substrate is assumed to be zero at the bottom.

Lead was chosen as the heavy metal of interest. *Chlorella vulgaris*, belonging to the algae family, was again used as the biomass species. The surface adsorption constant K_p was taken as 3500 mg/l, and the kinetic constants R_1 and R_2 were taken as 0.1 and 0.22 hr^{-1}, respectively. Specific growth rate of the biomass was set at 0.1 hr^{-1}. The simulations were carried out for a 48-hour period.

Figure 7 shows a comparison of the effects of both biosorption and physical adsorption to soil on lead transport after 48 hours of simulation. Equilibrium is assumed for adsorption of biomass to the soil. Effect of physical adsorption of metal to the soil is investigated by using a partitioning coefficient K_f of 50 L/kg, which is typical of soil whose physical properties and adsorption characteristics (in the presence of citric acid) are shown in Tables 3 and 4. The water content in the column has a steep decrease between 1.3 and 1.5 m, indicating the presence of a steep water front . Lead concentrations in the aqueous phase with biomass growing in the column are lower than in the column without growth of biomass. Adsorption of metal to the active biomass causes a reduction in the metal concentration in the aqueous phase. There is a change in the slope of the concentration profiles at around 1.4 m from the top of the column. These trends in the concentration profiles may be attributed to the sharp change in the water content profile at 1.4 m after 48 hours. Aqueous lead concentrations decreased significantly when adsorption of lead to the soil was considered. The concentration profile is retarded by about 1 m when sorption to the soil is taken into account. The lead concentrations are further decreased when the physical adsorption of soil is considered along with the biosorption. The focus of this study is on biosorption of lead. Hence, in most of the subsequent examples, the physical adsorption to soil is intentionally not included. This enables us to illustrate the effects of various biological parameters on lead biosorption and transport.

Figure 8 shows the influence of biomass on the downward movement of lead at different times of simulation for the above example without lead adsorption to soil. All the other parameters were the same as those described above. It is observed that concentrations of the biomass after 48 hours were significantly greater than those after 24 hours. Differences in concentrations between total lead and lead in aqueous phase are due to the adsorption of lead to the biomass. It can be seen that the differences in the concentrations are much greater after 48 hours of simulation than that after 24 hours. This is likely due to increase in the lead biosorption with the growth of the biomass. Sharp changes in the

slope of the concentration profiles at 0.65 m after 24 hours, and at 1.4 m after 48 hours, are due to the sudden changes in the water content profiles at these distances in the column, thus showing the influence of the water content on the movement of lead in unsaturated soils.

Water flux at the top influences the pore water distribution, and accompanying biomass, and metal in the unsaturated zone. Figures 9, 10, 11, and 12 show transient results after 24 hours of simulation. Figure 9 shows changes in the water content and pore water velocities in the column at different inlet water fluxes. Pore water velocities respond to sharp changes in the water content for different inlet water fluxes. Saturated hydraulic conductivity K_s chosen for this study is 0.05 m/hr; that is, gravitational drainage corresponds to 0.05 m/hr superficial velocity or 0.12 m/hr pore water velocity under saturated conditions. Different water fluxes at the inlet boundary allows us to study changes in biomass and lead concentrations.

Figure 10 shows concentration profiles of the total biomass present in both aqueous and sorbed phases of the soil. A constant source of substrate, at a level of 40 mg/l, is provided for the biomass growth. The value of the partition coefficient for the biomass chosen in this example is 30 kg/L. A significant portion of the total biomass is attached to the soil. Total biomass concentration is increasing with the increasing water fluxes fed at the top of the column. As this water flux increases, mass of biomass entering the column also increases. Biomass growth is obviously enhanced with increasing water flux because of the increased water content (see equations (7) and (8)), and increased inlet flux of biomass. Consequently, the growth rate, and therefore the total biomass concentration, increases. Increased growth of biomass may enhance lead uptake by immobilizing it in biomass adsorbed to soil.

Figure 11 shows the concentrations of lead in the aqueous phase. This includes lead that is in solution as well as that attached to the mobile fraction of the biomass. Lead concentrations do not change very much with increasing inlet water flux. In all cases, growth of biomass is sufficient to reduce the metal concentration in the aqueous phase to below 0.001 mmol/L of solution.

Figure 12 shows the effect of biomass partition coefficient on lead movement in the soil column. It can be seen that the concentration of total lead in the aqueous phase decreases with corresponding increase in the partition coefficient. Sharper changes in the concentration profile accompanying the wetting front are observed with lower partition coefficients.

Figure 13 shows the effect of partitioning of the metal between the aqueous phase and the biomass. Biomass is allowed to grow within the column with a constant substrate concentration of 40 mg/L that is uniformly distributed within the column. Simulations are carried out for 24 hours when both metal and biomass are undergoing advection, dispersion, and retardation. Higher coefficients will partition more metal into the aqueous phase according to the definition of the partition coefficient K_p. As shown in Figure 13, aqueous lead concentrations within the column are always greater for higher values of the partition coefficient than for those with lower values of the partition coefficient.

Journal of Hazardous Substance Research   **9-11**

Thus far, the examples have assumed a uniform distribution of the substrate within the column during the entire period of simulation. Often, the substrate concentrations within the column vary due to its transport by advection and diffusion processes. Figure 14 presents the case of metal movement being influenced by the biomass growing on the supplied substrate, the availability of which is limited by advection and dispersion processes. The substrate concentration at the top of the column is always maintained at 40 mg/L, indicating a constant source at the boundary, while the concentration gradient is zero at the bottom of the column. It can be observed that substrate concentrations decrease in the column when biomass is growing. At any distance greater than 0.8m, advection and dispersion processes dominate, and the substrate concentrations decrease with distance. In the region where the biomass grows near the top of the column, there is a difference between the total and aqueous lead concentrations, indicating lead sorption to the growing biomass. This difference is smaller when compared to that shown in Figure 8 with constant substrate concentration of 40 mg/L throughout the column. Lead sorption to biomass is greater with a constant substrate concentration than that with the substrate concentration limited by transport processes. This can be observed more clearly in Figure 15 where total and aqueous lead concentrations are shown after 48 hours of simulation.

Figure 16 shows the effect of physical adsorption for the soil whose characteristics are given in Tables 3 and 4. Biomass characteristics used in this simulation are presented in Table 2. A partition coefficient of 205 L/kg was used when there is no citric acid present along with lead in soil. This coefficient was obtained by fitting the aqueous concentrations against adsorbed concentrations with a linear isotherm. It can be observed that the aqueous lead concentrations are severely retarded when physical adsorption is considered. Physical adsorption of lead is due to the ion exchange with soil and sorption to organic matter.

SUMMARY AND CONCLUSIONS

Certain microbial species such as algae have strong metal-accumulating capabilities. Metal uptake by microbial species is generally a dual process as confirmed by experimental observations of both passive and active uptake. A mathematical model to describe heavy metal adsorption to both soil and microbial biomass has been developed and simulation results show that both adsorption processes retard heavy metal transport in unsaturated soils.

The model is used to estimate the amount of metal retained by the biomass in the vadose zone of soils. A certain fraction of the biomass in the soil will be in the aqueous phase and is likely to facilitate metal transport. Hence, both the transport of biomass and metal transport with the biomass in the aqueous phase of the soil were incorporated into the model. Growth of biomass depends on several factors such as availability of nutrients or substrates, electron acceptors, pH, and temperature. Soluble organic acids present in the rhizosphere serve as good sources of substrates for the growth of microorganisms. However, their availability at deeper locations in soils is dictated by processes of advection and diffusion. Our model includes the transport of these substrates in order to realistically consider

their influence on microbial growth, which in turn affects the transport of metals in soils. Sorption of lead to biomass in the presence of a substrate, that itself undergoes advection and dispersion in soils (see Figure 14), is lower than the idealized case where there is a continuous supply of substrate (see Figure 8).

He (1997) observed enhanced adsorption of Pb and Zn in the presence of citric acid in her saturated soil column experiments. Zn concentration at the top section of the soil column increased by 150 mg/kg when 500 μmole of citric acid was added to the soil at the top of the column. In case of Pb, a significant fraction of the total lead was retained at the top section of the soil column when citric acid was added to the column. Model results presented in Figures 8 and 15 also illustrate the enhanced retention of lead in soil in the presence of substrates, enriching microbial growth. It can also be observed from Figure 15 that the difference between total lead and aqueous lead concentrations is greater at the top of the column, indicating higher retention of lead to soil due to higher substrate concentrations in that region. The model is in qualitative agreement with experimental observations of He (1997). Thus, enhanced adsorption of lead in soil with active biomass can be used in retarding the amount of lead leaching to the groundwater.

During simulations, water flux at the top of the column was increased from a low value to that corresponding to saturation so that the influence of different water contents could be studied. Increased water fluxes did not bring significant changes in the aqueous lead concentrations. Biomass concentration in the column increased with increasing water flux. As a consequence, we would expect an increase in lead immobilization due to enhanced microbial activity. However, an increase in water flux at the top of the column also resulted in a corresponding increase in the facilitated transport of lead with the aqueous biomass. Due to these opposite effects of enhanced microbial activity and increased facilitated movement, lead concentrations in the aqueous phase did not change significantly as the water flux at the top was increased.

The partition coefficient of biomass to the soil depends on the soil characteristics and the type of the biomass. Clays yield higher partition coefficients for the biomass compared to silty loam soils. Increased partitioning of biomass to the soil resulted in increased lead immobilization. This was concluded from the corresponding reduction of lead in the aqueous phase. Also, partitioning of the metal between the aqueous phase and the biomass depends on the type of biomass considered and the type of metal. In our case, lead concentrations in the aqueous phase are sensitive to changes in the partitioning coefficient of lead between the aqueous phase and the biomass (see Figure 13). Thus, factors that enhance biomass growth and adsorption of biomass to soil generally contribute positively to metal immobilization and retardation.

In areas where the metal contamination is extending over several acres of land, vegetation is an economically viable alternative to conventional technologies. One of the beneficial effects of vegetation is to provide enhanced microbial activity in the presence of root exudates. Our results indicate the

Journal of Hazardous Substance Research **9-13**

possibility of reducing lead contamination in shallow soil layers from leaching to the ground water using biomass that can be sustained by vegetation.

ACKNOWLEDGMENTS

REFERENCES

Baath, E., Effects of heavy metals in soil on microbial processes and populations (a review), Water, Air and Soil Pollution, 47, 335-379, 1989.

Brooks, R.H., and A.T. Corey, Hydraulic properties of porous media, Hydrology Paper No.3. Colorado State University, Fort Collins, 1964.

Cederberg, G.A., R.L. Street, and J.O. Leckie,A mass transport and equilibrium chemistry model for multicomponent systems, Water Resources Research, 21(8), 1095-1104, 1985.

Corapcioglu, M.Y., and A. Haridas, Transport and fate of microorganisms in porous media: A theoretical investigation, Journal of Hydrology, 72, 149-169, 1984.

Corapcioglu, M.Y., and A. Haridas, Microbial transport in soils and : A numerical model, Advances in Water Resources, 8, 188-200, 1985.

Gadd, G.M., and C. White, Microbial treatment of metal pollution - a working biotechnology?, Trends in Biotechnology, Bioremediation, 11(8), 353-359, 1993.

Govindaraju, R.S., and M.L. Kavvas, Development of an approximate model for unsaturated flow with root water uptake under rectangular water content profiles assumption, Journal of Hydrology, 146, 321-339, 1993.

Hall, J.C., R.L. Raider, and J.A. Grafton, EPA's heavy metal criteria, strategies for obtaining reasonable limitations, Water Environment & Technology, 60-63, March 1992.

He, YingHong, Adsorption and transport of zinc and lead in soil as affected by organic ligands, PhD Dissertation, Department of Civil Engineering, Kansas State University, Manhattan, Kansas, 1997.

Hiroki, M., Effects of heavy metal contamination on soil microbial population, Soil Science and Plant Nutrition, 38(1), 141-147, 1992.

Inthorn, D., H. Nagase, Y. Isaji, K. Hirata, and K. Miyamoto, Removal of cadmium from aqueous solution by the filamentous cyanobacterium tolypothrix tenius, Journal of Fermentation and Bioengineering, 82(6), 580-584, 1996.

Kurek, E., Janusz Czaban, and Jean-Marc Bollag, Sorption of cadmium by microorganisms in competition with other soil constituents, Applied and Environmetal Microbiology, 48(5), 1011-1015, 1982.

Lambert, M.W., G.M. Pierzynski, L.E. Erickson, and J.L. Schnoor, Remediation of lead, zinc, and cadmium-contaminated soils, Issues in Environmental Science and Technology, 7, 91-102, 1997.

Lensing, H.J., M. Vogt, and B. Herrling, Modeling of biologically mediated redox processes in the subsurface, Journal of Hydrology, 159, 125-143, 1994.

Pierzynski, G.M., J.L. Schnoor, M.K. Banks, J.C. Tracy, L.A. Licht, and L.E. Erickson, Vegetative remediation at Superfund sites, Mining and its environmental impact, in Issues in Environmental Science and Technology, eds. R.E. Hester and R.M. Harrison, Royal Society of Chemistry, 1, 49-69, 1994.

Schiewer, S., and B. Volesky, Modeling multi-metal ion exchange in biosorption, Environmetal Science and Technology, 30, 2921-2927, 1996.

Shuttleworth, K.L., and R.F. Unz, Sorption of heavy metals to the filamentous bacterium thiothrix strain A1, Applied and Environmental Microbiology, 59(5), 1274-1282, May 1993.

Swift, D.T., and D. Forcinti, Accumulation of lead by Anabena cylindrica: mathematical model and an energy dispersive x-ray study, Biotechnology and Bioengineering, 408-418, 55(2), July 20, 1997.

Taylor, S.W., and P.R. Jaffe, Substrate and biomass transport in a porous medium, Water Resources Research, 26(9), 2181-2194, 1990.

Thomson, N.R., J.R. Sykes, and W.C. Lennox, A Lagrangian porous media mass transport model, Wat. Resour. Res., 20(3), 391-399, 1984.

Ting, Y.P., F. Lawson, and I.G. Prince, Uptake of cadmium and zinc by the alga chlorella vulgaris, Biotechnology and Bioengineering, 34, 990, 1989.

Ting, Y.P., F. Lawson, and I.G. Prince, Uptake of cadmium and zinc by the alga chlorella vulgaris: Part II. Multi-ion situation, Biotechnology and Bioengineering, 37, 445-455, 1991.

Trevors, J.T., G.W. Stratton, and G. M. Gadd, Cadmium transport, resistance, and toxicity in bacteria, algae, and fungi, Can. J. Microbiology, 32, 447-464, 1986.

Volesky, B., and Z.R. Holan, Biosorption of heavy metals, Biotechnol. Prog., 11, 235-250, 1995.

Walter, A.L., E.O. Frind, D.W. Blowes, C.J. Ptacek, and J.W. Molson, Modeling of multicomponent reactive transport in 1. Model development and evaluation, Wat Resour. Res., 30(11), 3137-3148, 1994.

Waters, C.Y., The influence of organic acids on leaching of heavy metals from contaminated mine tailings, M.S. Thesis, Department of Civil Engineering, College of Engineering, Kansas State University, Manhattan, Kansas, 1992.

Yeh, G.T., and V.S. Tripathi, A model for simulating transport of reactive multispecies components: Model development and demonstration, Water Resources Research, 27(12), 3075-3094, 1991.

Table 1. Uptake of metals (mg/g) by microbial biomass (from Volesky and Holan, 1995).

Metal	Fungi	Algae	Bacteria
Lead	373	436	189
Zinc	20	*	3.4
Chromium	31	*	118
Gold	176	400	70
Cadmium	56	215	101

*indicates non-availability of data

Table 2. Simulation parameters for column study.

Nodal Spacing (dx)	=	0.0125 m
Time Increment (dt)	=	0.005 hr
Water Flux at the Top (q)	=	0.005 m/hr*; 0.015 m/hr; 0.05 m/hr
Free Drainage Condition for Water at the Bottom Biomass Concentration in Aqueous Phase at the Top (C_b)	=	1 mg/L
Lead Concentration Entering at the Top (C)	=	4.8 μmol/L
Dispersion coeffecient for the Metal (D)	=	0.1 sq.m/hr
Soil Bulk Density (ρ)	=	1.3 kg/L
Saturated Hydraulic Conductivity (K_{sat})	=	0.05 m/hr
Air-Entry Pressure (Ψ_b)	=	-15 cm of water
Initial Water Content (θ)	=	0.1
Saturated Water Content (θ_s)	=	0.43

Rate Constants for Microbial Metal Uptake (Ting et al., 1991)		
R_1	=	0.10/hr
R_2	=	0.22
Metal Partitioning Coefficient to Biomass (K_p)	=	1 g/L; 3.5 g/L*; 10 g/L
Maximum Growth Rate (μ_m)	=	0.5/hr
Half-Saturation Constant (K_s)	=	100 mg/L
Substrate Concentration (C_s)	=	40.0 mg/L
Biomass Partitioning Coefficient (K_{db})	=	10 kg/L; 30.0 kg/L*; 50 kg/L; 100 kg/L
Metal Partitioning Coefficient (K_p)	=	*0 L/kg; 50 L/kg; 205 L/kg
Biomass Dispersion (D_b)	=	0.5 m²/hr
Endogenous Decay (K_d)	=	0.001/hr
Cell Yield (Y)	=	0.4 gm biomass/gm substrate

*Indicates a base case simulation value for the parameter. This vailue is used in all simulations, except where it is varied with the values shown.

Table 3. Physical characteristics for soil (He, 1997).

Sand	:	35%
Silt	:	42%
Clay	:	23%
Organic matter	:	2.5%
pH	:	4.8
CEC (meq/100gms)	:	19

Table 4. Adsorption eqiuilibrium data for lead for soil (He, 1997).

with 5 mmole/L of citric acid as substrate

Aqueous-Phase Lead (mg/L)	Adsorbed Lead (mg/L)
0.0	0.0
5	186
7.5	254
12.5	374
22.5	584

with 0 mmol/L of citric acid as substrate

Aqueous-Phase Lead (mg/L)	Adsorbed Lead (mg/L)
0.0	0.0
1	270
2.5	588
4	878
5	1063

Journal of Hazardous Substance Research **9-17**

Figure 1. Processes affecting the fate of metals (adapted from Pierzynski et al., 1994).

PLANT UPTAKE

BINDING BY
SOLUBLE ORGANIC
COMPLEXES
(ROOT EXUDATES)

PRECIPITATION BY
MINERALS

FREE METAL ION

IN

AQUEOUS PHASE

ADSORPTION
TO
SOIL CLAY
AND ORGANIC
MATTER

ADSORPTION TO
THE SOIL BIOMASS

LEACHING
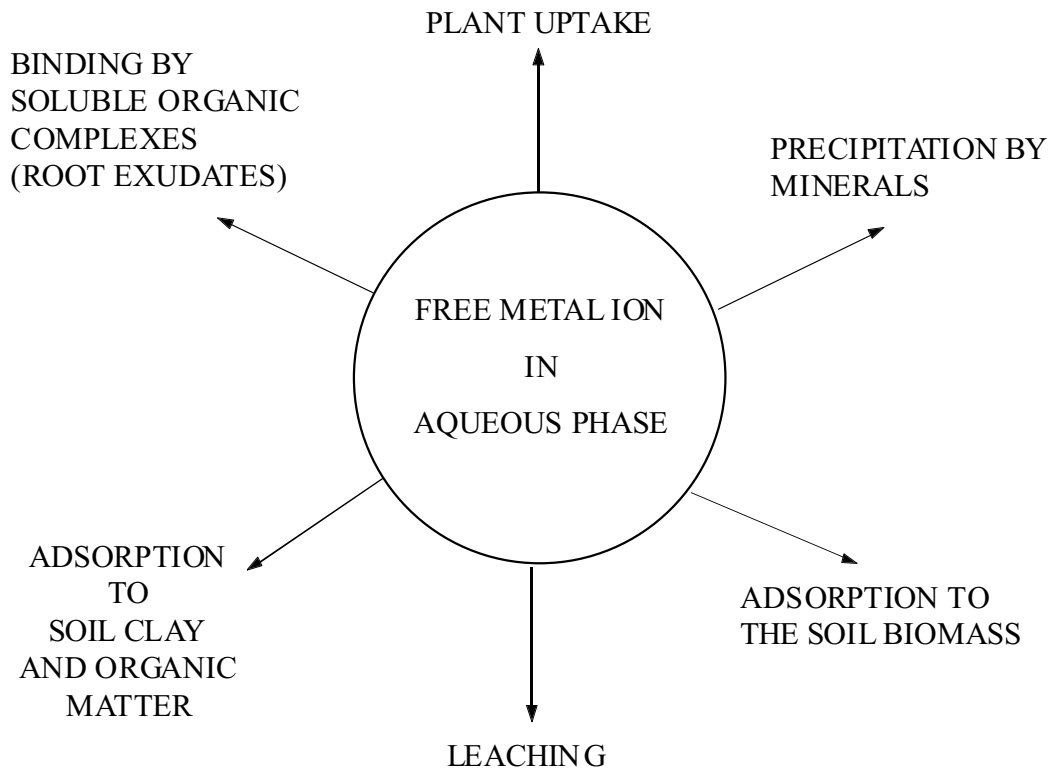
Figure 2. Passive and active adsorption processes for a metal and a typical microbial cell.

ACTIVE
ADSORPTION

C

PASSIVE
ADSORPTION

C

CELL SURFACE

$Cp + E = CE$

Cp

$CE = E + Ca$

Ca

E

E

MEMBRANE

CELL INTERIOR
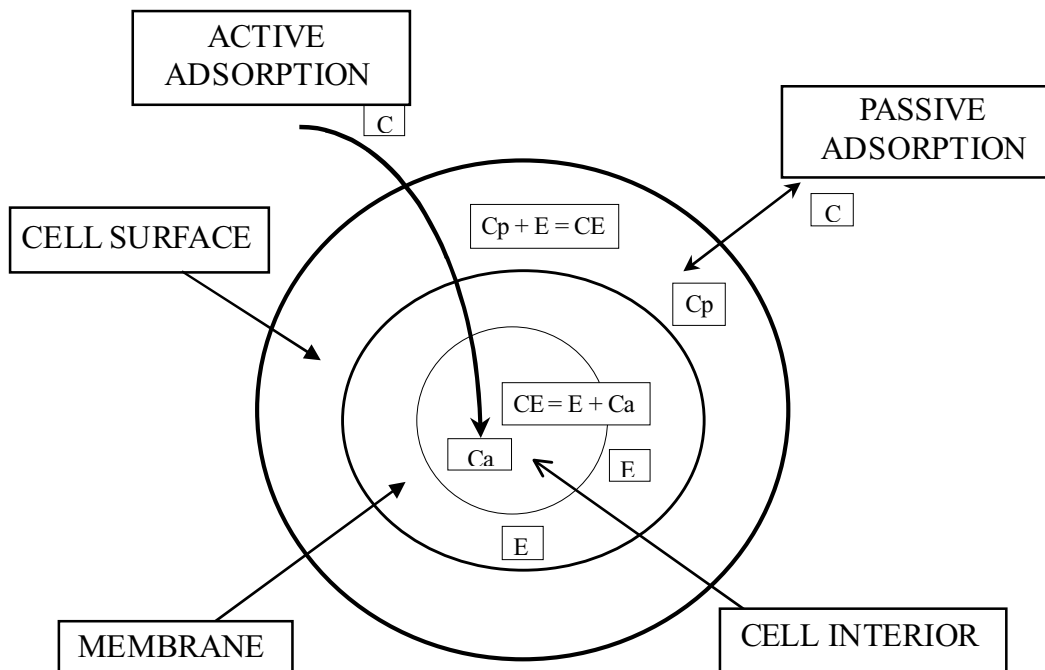
Figure 3. Overall simulation solution strategy.
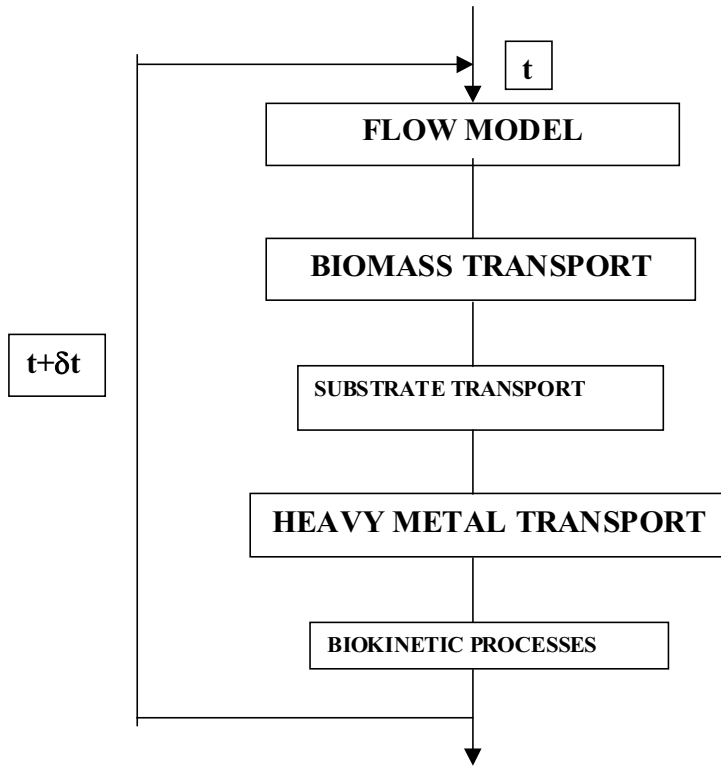
Figure 4. Illustration of lead uptake with the simultaneous growth of biomass.
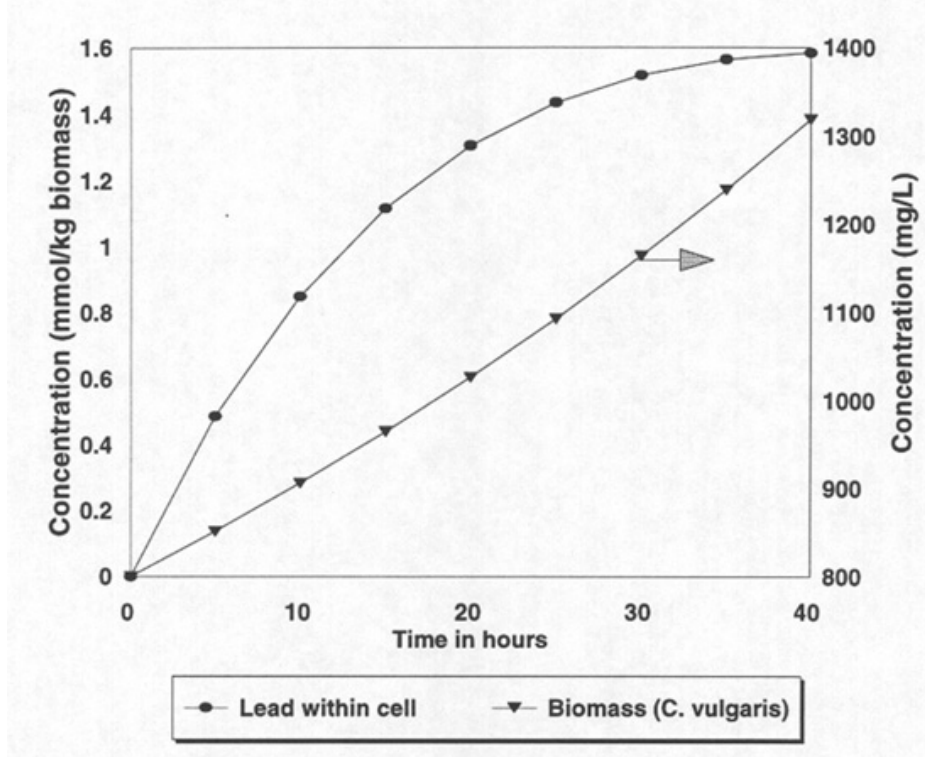
Figure 5. Concentration of lead in aqueous phase, as influenced by c.vulgaris.
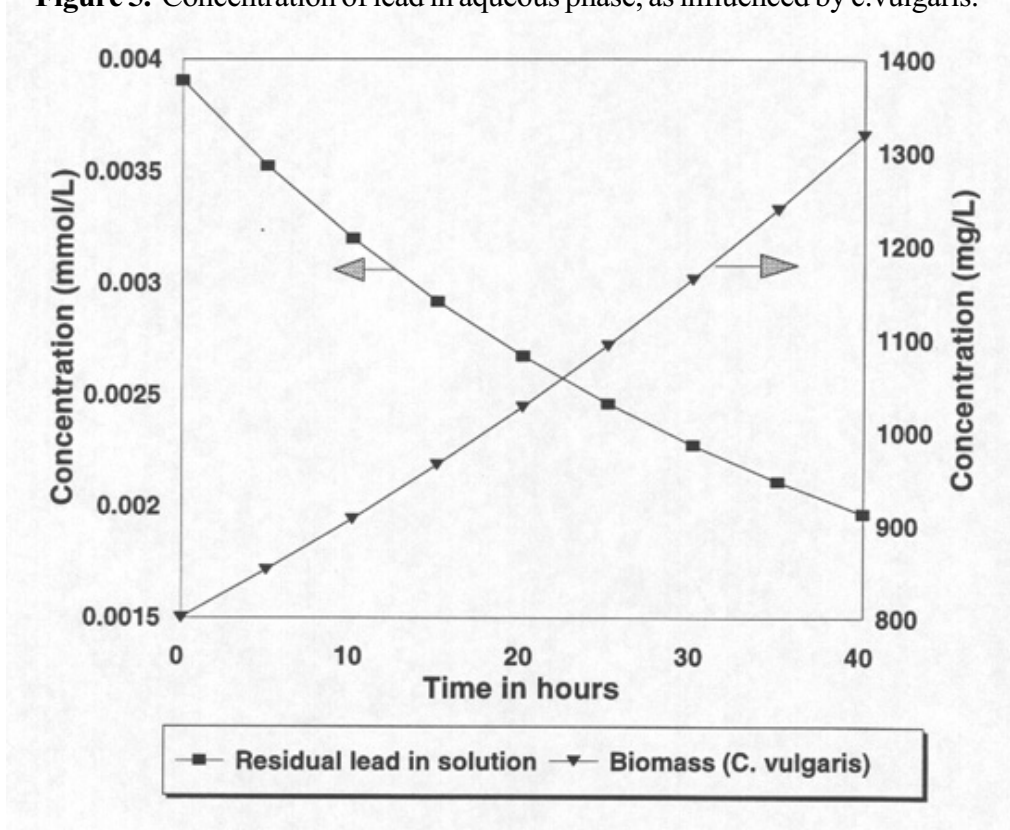
**Figure 6.** Illustration of cadmium uptake with the simultaneous growth of biomass.
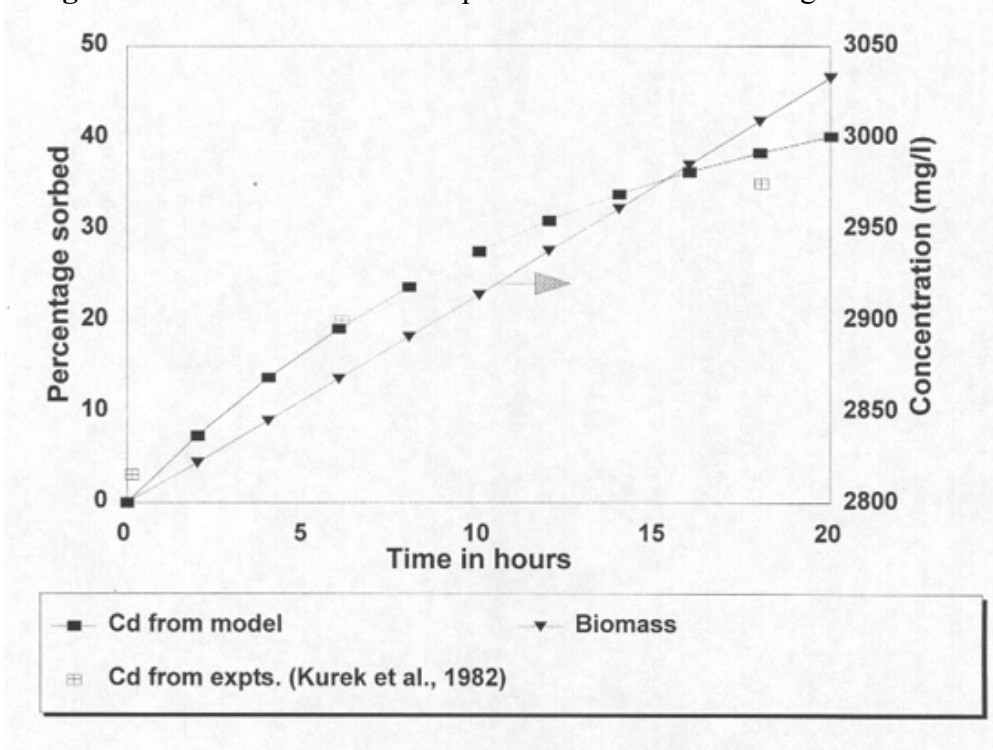
Figure 7. Comparison of influences of biosorption and physical adsorption to soil on lead transport after 48 hours of simulation. Biomass is growing at the rate of 0.1/hr and the partitioning coefficient of lead to soil is 50 L/kg.
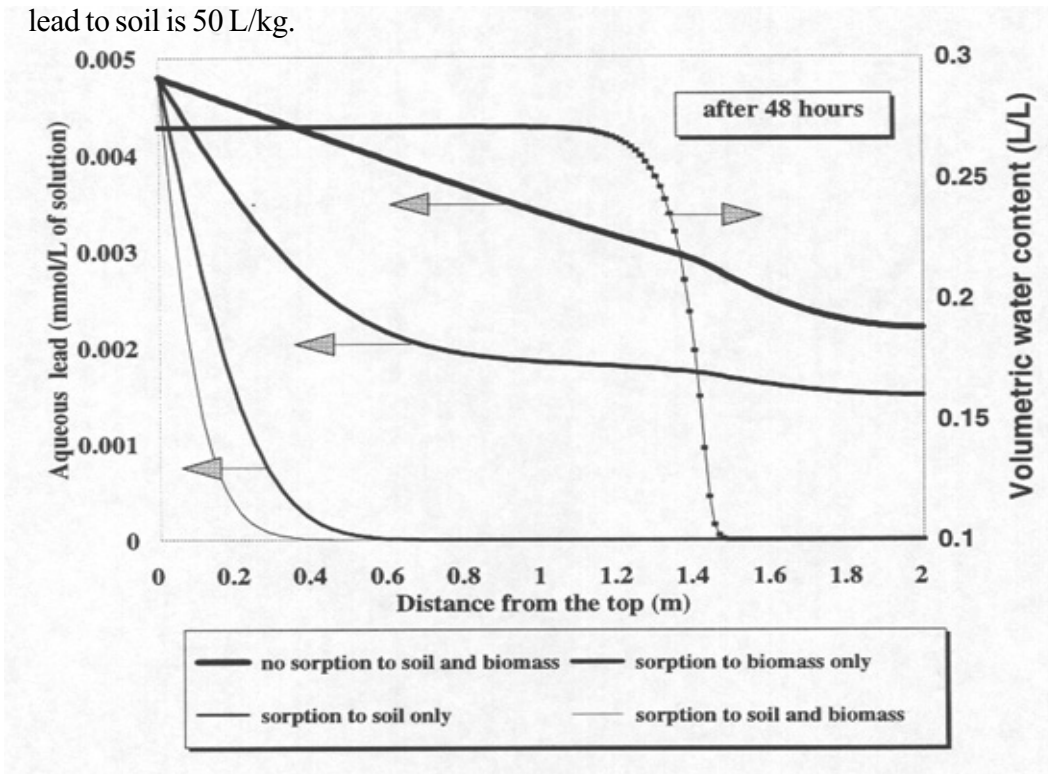
Figure 8. Effect of growing biomass on total and aqueous lead in a soil column. The substrate concentration is always maintained at 40 mg/L throughout the entire column.
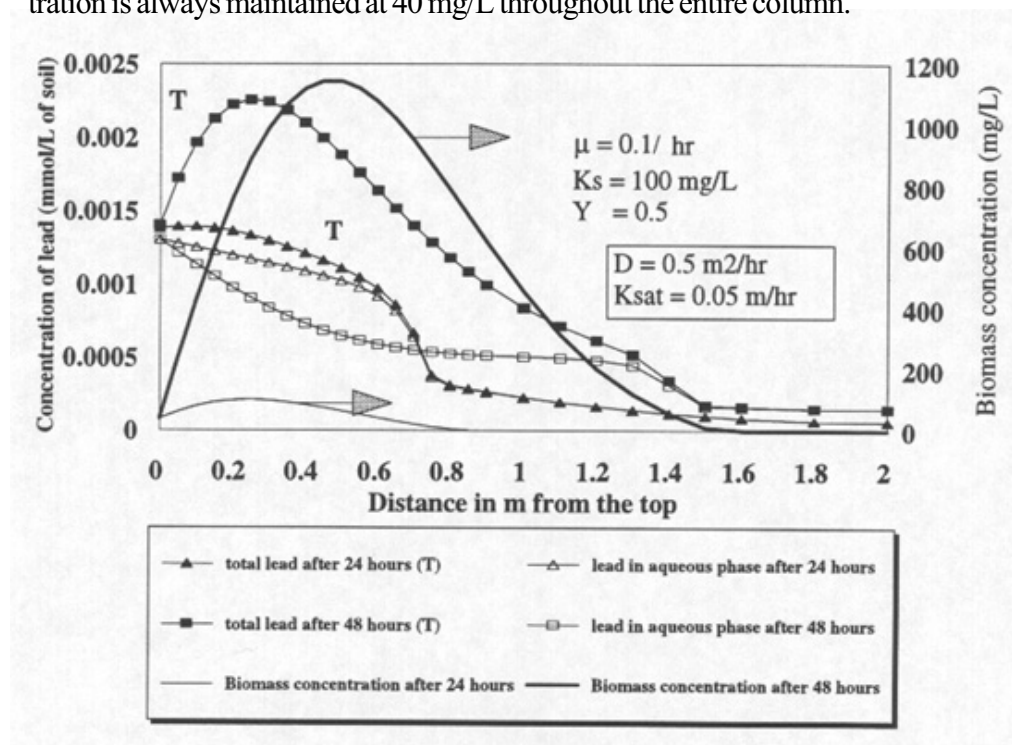
Figure 9. Influence of the unsaturated zone on water movement in a soil column with drainage due to gravity at the bottom. The unsaturated zone is maintained by controlling the water flux (also called superficial velocity) at the top of the soil column at different values. The superficial velocity is 0.005 m/hr for case (a), 0.015 m/hr for case (b), and 0.05 m/hr for case (c).
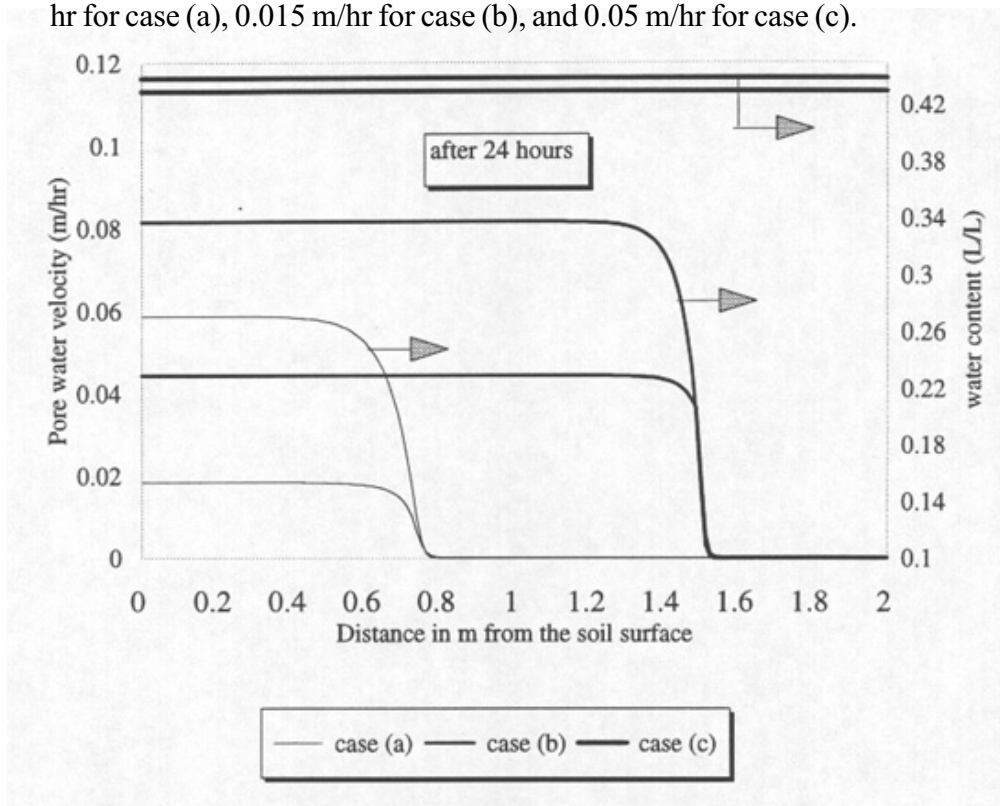
Figure 10. Influence of the unsaturated zone on total biomass concentration in a soil column for three inlet water fluxes at the top of the column. The substrate concentration is always maintained at 40 mg/L throughout the entire column.
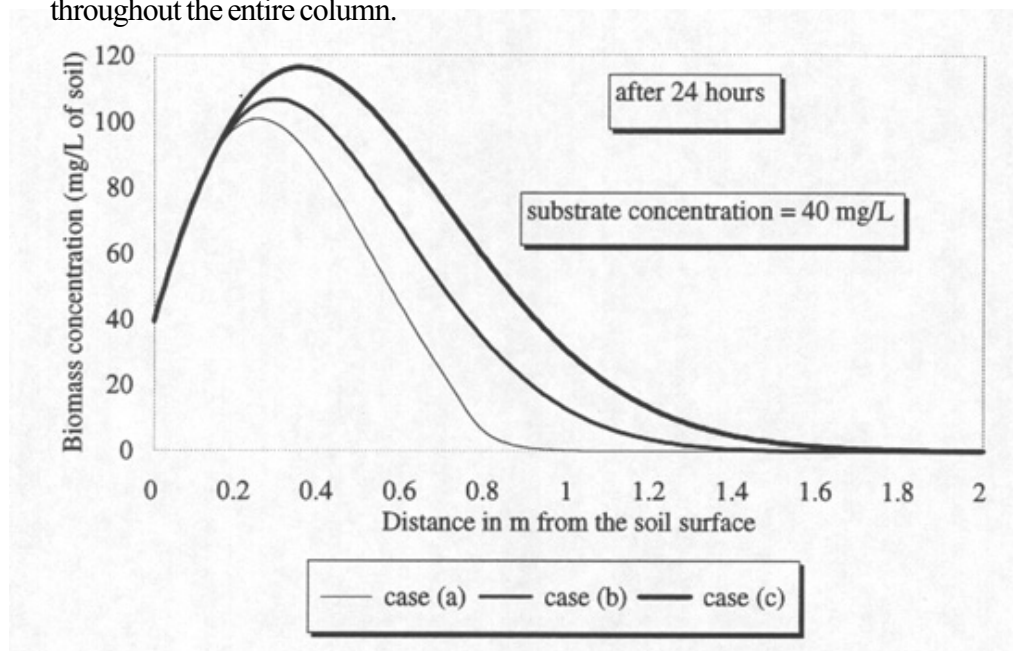
Figure 11. Influence of the water flux on lead concentration in a soil column for three inlet water fluxes at the top of the column.
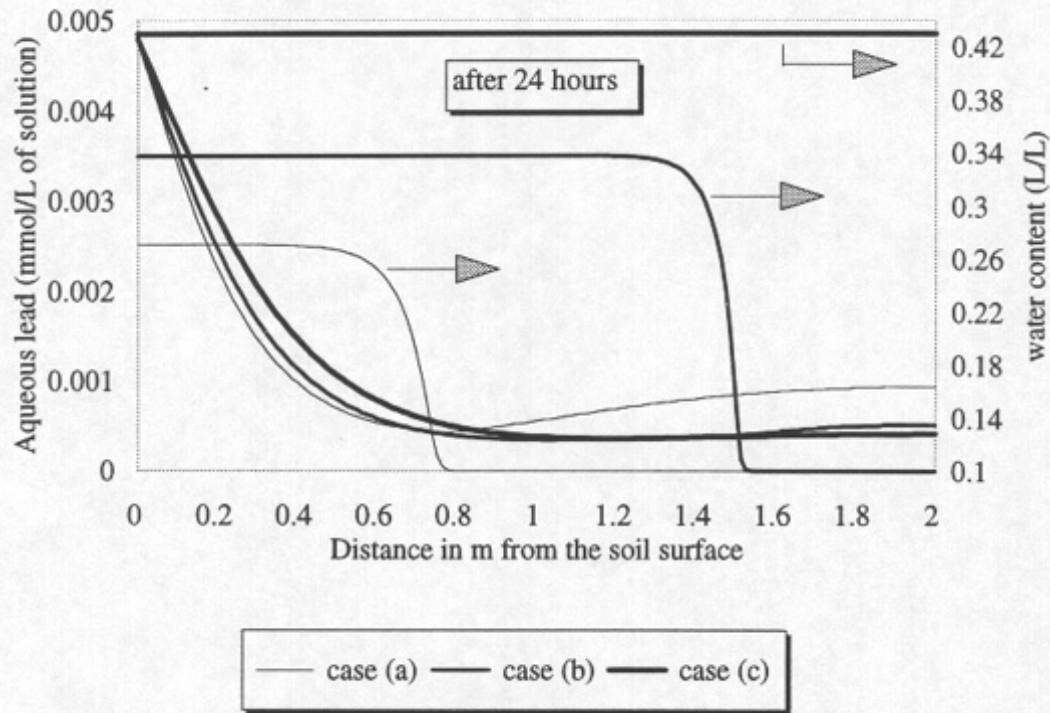

Figure 12. Effect of biomass partition coefficient on lead movement in the aqueous phase. The partition coefficient is 1 kg/L for case (a), 10 kg/L for case (b), and 100 kg/L for case (c). The substrate concentration is always maintained at 40 mg/L throughout the entire column. The superficial velocity at the top of the column is maintained at 0.005 m/hr with free drainage at the bottom.

Journal of Hazardous Substance Research   **9-23**
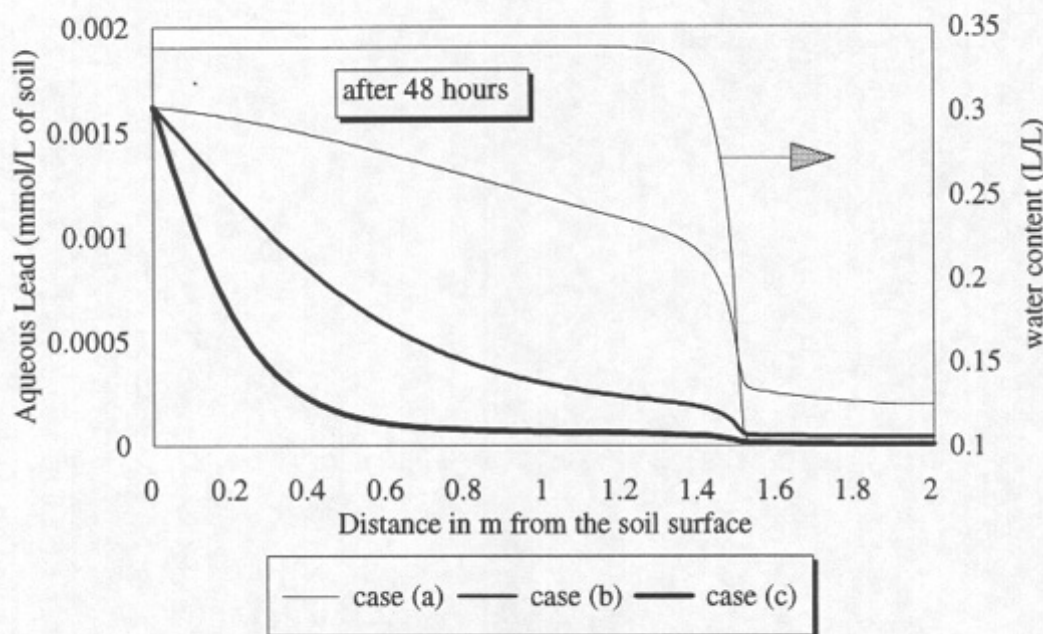
Figure 13. Effect of metal partitioning between the aqueous phase and biomass. The partition coefficient is 1,000 mg/L for case (a), 3,500 mg/L for case (b), and 10,000 mg/L for case (c). The substrate concentration is always maintained at 4 mg/L throughout the entire column. The superficial velocity at the top of the column is maintained at 0.005 m/hr with free drainage at the bottom.
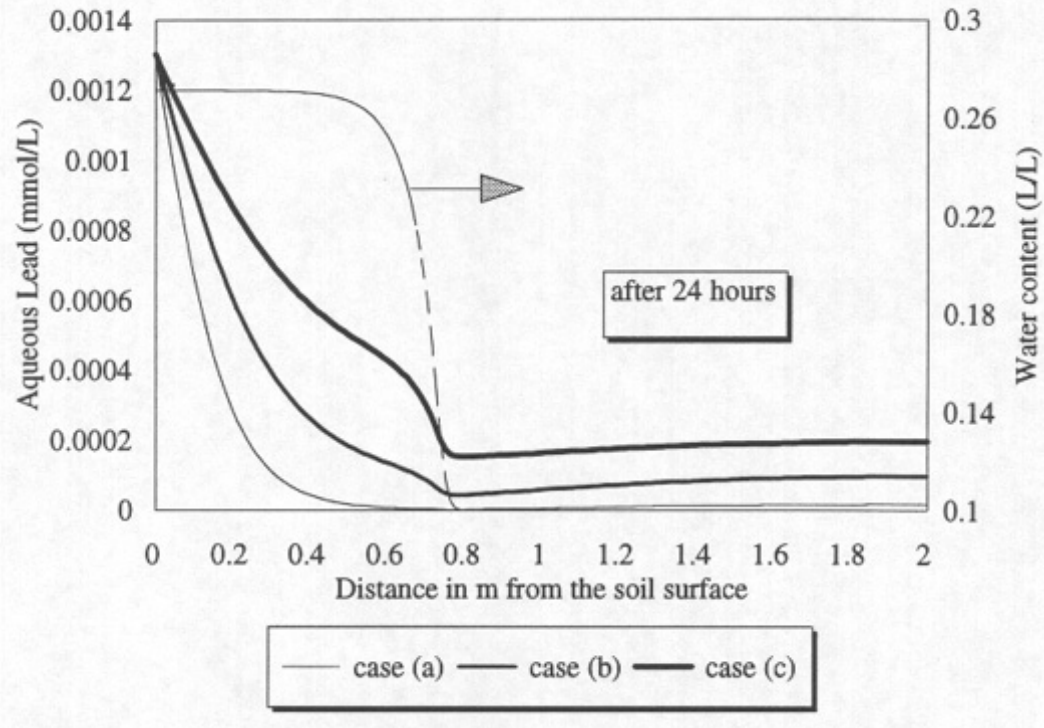
Figure 14. Effect of growing biomass on total and aqueous lead in the soil column after 24 hours. Substrate availabillity for the biomass is limited by the advection and dispersion of the substrate throughout the soil.
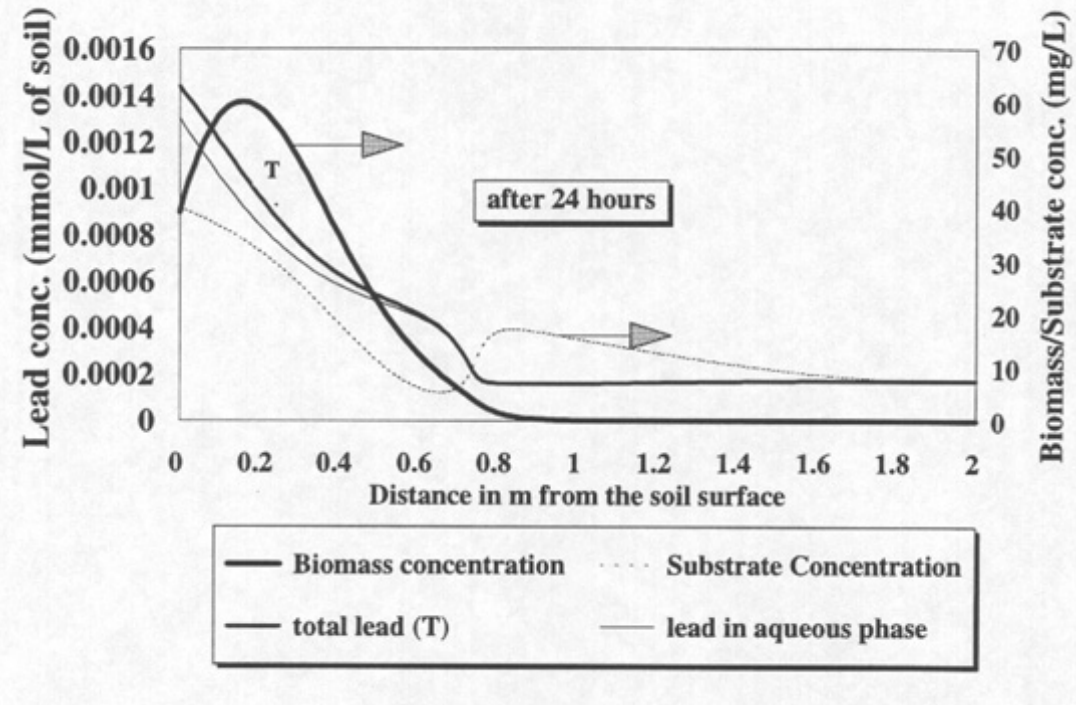
Figure 15. Effect of growing biomass on total and aqueous lead in the soil column after 48 hours. Substrate availability for the biomass is limited by the advection and dispersion of the substrate through the soil column. Enhanced uptake of lead by biomass is observed after 48 hours when compared to that in Figure 14 after 24 hours.
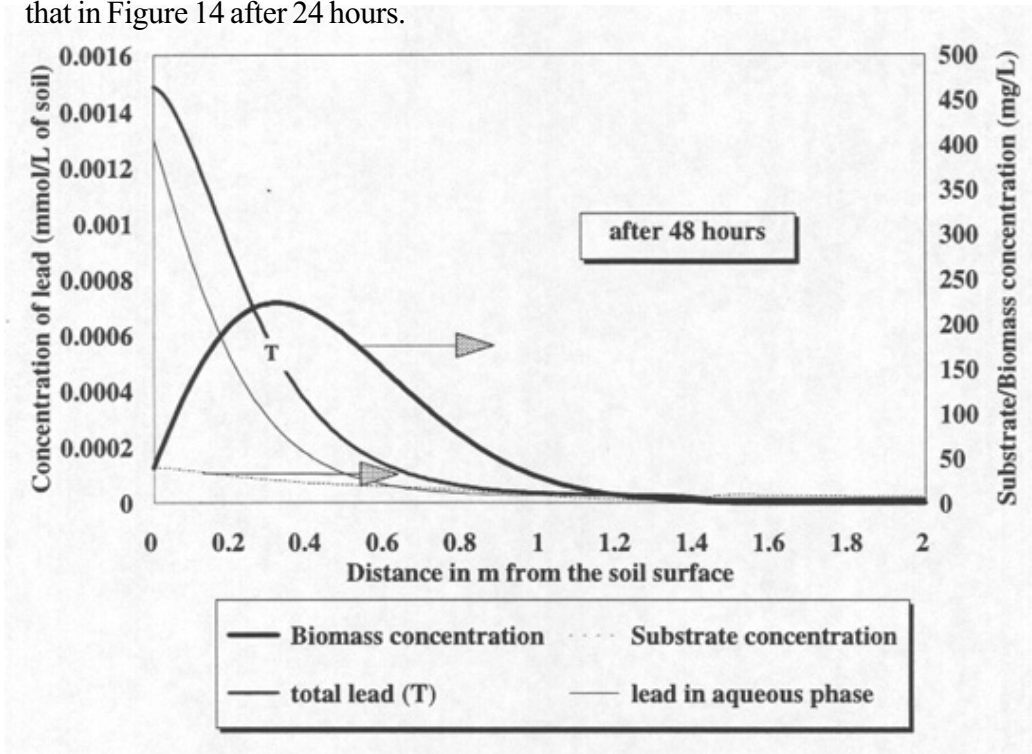
Figure 16. Effect of physical adsorption of lead to the soil on lead in the aqueous phase within the column.

Journal of Hazardous Substance Research   **9-25**