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TRANSPORT OF PATHOGENS THROUGH SOILS AND AQUIFERS

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1. INTRODUCTION

Between 1971 and 1980, the use of untreated groundwater was responsible for more than one-third of the waterborne disease outbreaks in the United States (1). This statistic points to the potential for subsurface contamination by pathogenic microorganisms. The major sources of pathogens are waste water effluents, residual sludges from waste treatment (2), and septic tank effluents (3, 4). The number of septic tanks in the United States has been estimated at 22 million units. These on-site treatment systems serve approximately one-third of the U.S. population (4). They are major contributors to the contamination of subsurface environments. The contaminants are household chemicals (nitrate, heavy metals, organic toxicants), pathogenic microorganisms, and parasitic cysts.

The fate of pathogenic microorganisms in soils and aquifer material is primarily governed by their transport and persistence in these environments. The survival and transport of pathogens in soils and aquifers are controlled by four major factors: climate (e.g., temperature, rainfall), type of soil or aquifer material (*e.g.*, texture, pH, water holding capacity, cation exchange capacity), properties of pore fluids (*e.g.*, chemistry, saturation), and type of pathogen (5). In the following sections, we review the major factors controlling the persistence and transport of pathogens and discuss modeling efforts to assess pathogen fate in soil and subsurface environments.

2. PERSISTENCE OF PATHOGENIC MICROORGANISMS IN SOILS AND GROUNDWATER

The topic of enteric bacterial survival in soils has been extensively reviewed (5-9). The major factors that control their persistence in this complex environment are temperature, moisture content, sunlight, pH, organic matter, bacterial type, and antagonistic microflora. Persistence is generally highest at low temperature, high soil moisture content, and abundant organic matter that may allow the growth of certain bacteria. Some pathogens, such as mycobacteria, are extremely hardy and survive for several months in soils. Pathogen survival is generally lower in acidic soils. Sunlight has a detrimental effect at the soil surface. Competition and antagonisism from other bacteria, as well as predation by protozoa, can contribute to the decline of pathogenic and indicator bacteria in soils.

Many of the abiotic factors that affect survival of bacteria in the subsurface also influence the persistence of viruses (10). Development in the late 1970s of better methods for virus recovery from soils increased our knowledge of the persistence of viruses in this complex environment. In general, all methods of virus recovery include an elution step followed by a concentration step. The most commonly used eluants include glycine-EDTA, beef extract, casein, and non-fat dry milk (11, 12). These procedures result in a recovery efficiency of approximately 50%. Viruses have also been recovered by eluting soil with deionized water and concentrating the eluant by sorption onto aluminum hydroxide (Al[OH]₃) flocs (13).

Soil temperature (14–16) and moisture (15, 17–21) are the primary factors affecting virus survival in soils treated with waste water effluents or sludge. Viruses survive best in moist soils under low temperature. Soil type may also influence survival, although the effects are probably related to the degree of viral adsorption (10). There is considerable variation among virus types in their ability to survive in subsurface environments. Among the enteric viruses, hepatitis A virus (HAV) appears to be among the most resistant to inactivation in soils (22).

Virus persistence in groundwater was studied under laboratory as well as field conditions, using traditional glass containers and McFeters-type diffusion chambers. These investigations indicate that bacteria and viruses persist longer in groundwater than in surface waters (21, 23-25). The decay rate (k) of poliovirus

in groundwater was found to be 0.0019 hr^{-1} (23). Similar decay rates were found for enteroviruses (poliovirus 1; echoviruses 6, 11, and 24; coxsackievirus B6) incubated in dialysis tubes under *in situ* conditions (25); decay rates of viruses in well water varied between 0.0004 to 0.0037 hr⁻¹. Virus survival in groundwater appears to be influenced by water temperature, dissolved oxygen, and, possibly, by indigenous microorganisms in groundwater. Yates and Gerba (26, 27) also showed that temperature was important in controlling phage persistence and that survival was also dependent on the groundwater source.

Geostatistical techniques (*e.g.*, kriging, cokriging, and combined kriging and regression) have been considered for estimating virus inactivation in groundwater in order to predict safe septic tank setback distances for drinking water well installation in the vicinity of septic tank drainfields (10, 28, 29). Results of recent laboratory investigations suggest that the relationship between virus inactivation rates and groundwater temperature may be described by a linear equation (10, 30). Combined use of kriging, which can be used to calculate rates of viral inactivation at unsampled locations from values obtained for samples collected nearby, and a linear regression equation relating viral inactivation and temperature allows estimation of setback distances between wells and contamination sources by knowing only the groundwater temperature and flow characteristics (10). Setback distances (D), in meters, can be calculated with the following equation:

 $D = (tKi)/\theta$

where t is the travel time required (days) for a 10 million-fold decline in number of active viruses, which is calculated from kriged estimates of virus inactivation; K is the hydraulic conductivity (m/day); i is the hydraulic gradient (m/m); and θ is the effective porosity (29).

3. PATHOGEN TRANSPORT THROUGH SOILS AND AQUIFER MATERIALS

3.1 Factors Controlling the Transport of Pathogenic Microorganisms

3.1.1. Bacterial Transport

Straining, which occurs within pores that are smaller than the limiting dimension of the cell, and adsorption onto particles are the major factors controlling transport of bacteria through soils (5). Bacterial cells are negatively charged biocolloids and, as such, they interact with soil particles (sand, silt, clay minerals, metal oxides). Bacterial adsorption to soils is favored in the presence of cations, clay minerals, low concentrations of soluble organics, and at low pH conditions (5). Bacterial removal by straining is inversely proportional to particle size of soils. Bacteria strained at the soil surface promote the retention of finer particles in soil, a phenomenon occurring during mat formation resulting from the application of septic tank effluents. The biological clogging mat or crust appears to be an effective barrier to bacterial breakthrough. The clogging mat may be due to biological factors such as the presence of microbial polysaccharides (31, 32).

Bacterial transport through absorption fields of septic tanks is controlled by soil porosity and the degree of saturation with water. Under saturated conditions bacteria may be transported over much greater distances than under unsaturated conditions. Studies have shown that pathogen transport through soil is promoted by heavy rainfall, and bacterial contamination of wells was found to coincide with periods of heavy rainfall (33, 34). Rainfall generally lowers the ionic strength of the pore fluid and thus promotes bacterial transport through soils. Laboratory experiments with sludge–soil mixtures challenged with bacterial indicators, total coliforms (TC) and fecal coliforms (FC), have shown that only heavy rainfall (12.3 cm/day) promotes significant downward transport of the bacteria to the bottom of the 8 inch-deep column. Lower amounts of rainfall did not cause significant migration of the bacterial cells. Bacterial breakthrough in the column leachates never exceeded 2.4% of TC and 1.4% of FC inputs to the columns (34).

Agricultural scientists have studied the introduction of beneficial microorganisms in the rhizosphere for the control of soilborne plant pathogens. Transport and survival of these microorganisms (*e.g.*, *Rhizobium*) in the rhizosphere is of great interest to agronomists. These studies have shown that the vertical flow of water is an important transport mechanism for bacteria in the rhizosphere (35). Bacterial transport by percolating water is enhanced by preferred flow path structure (channels) created by plant roots and by the activity of soil invertebrates such as earthworms (36). Growing wheat roots were also found to enhance the downward transport of a genetically engineered microorganism by enhancing water flow through the soil (37).

3.1.2. Virus Transport

Because of their small size, viruses are less subject to straining in sandy aquifer sediments and soils than are bacteria. Therefore, bacteria do not serve as good indicators for virus transport into groundwater. Under field conditions, indicator bacteria, total coliforms, fecal coliforms, and fecal streptococci (TC, FC, and FS) originating from a septic tank system were efficiently retained by a sandy/loamy sand soil and were detected at levels less than 1 count per 100 ml of groundwater. However, polioviruses were transported to groundwater. (38).

The major abiotic impediment to the migration of viruses through the subsurface is sorption onto solid surfaces. Virus adsorption to solids, including soils, can be explained largely in terms of surface interactions between the amino acids on the capsid and biological or nonbiological surfaces. These interactions include both electrostatic and hydrophobic interactions (39-43). Several factors control virus adsorption to soils. Binding to soils is affected by soil texture. The presence of clays and other minerals such as hematite and magnetite increases virus retention (39, 44-46). Conversely, soil organic matter may decrease adsorption (47-50); organic-rich sediments ("mucks") have been observed to have a low retention capacity for viruses (50, 51). Increases in hydraulic flow rate through soils facilitate virus transport (52-54) by lessening the degree of adsorption.

Both batch and column experiments have shown that viruses do not readily sorb to or are released from soil particle surfaces when suspended in low ionic strength solutions (44, 49, 55–57). Rainwater, having low ionic strength, can be instrumental in the redistribution and transport of viruses within the soil profile (14, 52, 53). Stools from infants who received the trivalent oral poliovirus vaccine were used to inoculate a septic tank so that poliovirus could be used as a marker and followed downgradient in nearby groundwater (38). It was observed that virus numbers in groundwater increased as the distance from the drainfield increased. It was concluded that the relatively high ionic strength of the septic tank effluent (1,000–3,100 μ S cm⁻¹) allowed virus adsorption to take place in the vicinity of the septic system. Virus desorption and movement away from the septic tank was promoted by dilution of the relatively high ionic strength of the septic tank effluent (38).

In contrast, soluble organic compounds in waste water effluents inhibit virus adsorption to surfaces including soils. Their presence would thus promote virus transport through soils (42, 58–60). Humic and fulvic acids also interfere with virus sorption to soils (50, 61).

Several investigations have shown that virus adsorption to solids (*e.g.*, soil, activated sludge flocs, sediments) depends on the type and strain of virus under consideration (55, 60, 62, 63). Virus adsorption to a sandy loam soil varied between 0% for echovirus 1, strain V239, to 99.9% for poliovirus type 1, strain LSc (63), presumably because of differences in surface characteristics of the viruses. The adsorption patterns of HAV and rotaviruses can differ from those of polioviruses and phages (22, 59). Adsorption capacity may also vary between isolates of the same virus type (49). A field study carried out at a groundwater recharge site in Western Australia showed considerable variation with regard to virus adsorption by a sandy soil (64). Viruses such as echoviruses 11, 14, 24, 29, and 30, coxsackievirus B4, and adenovirus type 3 penetrated 3.0 m of soil. Echovirus type 11 was detected in groundwater at 9 m below the recharge basin. Conversely, indigenous and laboratory strains of polioviruses both displayed

higher affinity for the soil under study; it was also observed that the laboratory strains adsorbed better to soil than did the indigenous viruses.

Properly designed soil-column experiments offer a relatively rapid means for assessing virus transport patterns in sludge-amended soils (65). Soil column experiments as well as field studies have shown that sludge-associated viruses become immobilized at the soil surface and do not migrate significantly through soils (15, 16, 66–69). Hurst and Brashear (70), using a vacuum filtration technique to study leaching of viruses from sludge, indeed confirmed that viruses will not desorb significantly from sludge particles following land application of sludge. A similar trend was observed for sludge-associated bacteria. Liu (71), investigating the transport of sludge-associated bacteria into groundwater, has concluded that bacterial contamination from on-land sludge disposal was unlikely. Most of the sludge-associated bacteria were retained at the soil surface.

3.2. Use of Tracers To Study Transport in Aquifer Materials

Abiotic tracers have been used in a number of field and laboratory studies involving transport of microorganisms through soils and groundwater. Welldefined particulate and dissolved tracers can be useful in accounting for abiotic processes that contribute to or control movement of bacteria and viruses through subsurface media. Often, the processes controlling transport in situ are complicated by a number of geohydrological, chemical, and biological factors (72). Nonpathogenic microorganisms were employed in many of the co-transport studies involving tracers and microorganisms. However, such studies have shed light on a number of processes affecting the migration of pathogens. Dissolved tracers that are assumed to be conservative (nonreactive) were employed in a number of microbial transport experiments performed in aquifers. The tracers provide information about the hydrologic characteristics along the paths of travel. This can be difficult to determine solely from the concentration histories (breakthrough curves) of the organisms themselves, since microorganisms tend to interact with solid surfaces and do not necessarily follow the same mean flow paths as the groundwater in which they are suspended.

Tracer experiments that are employed to examine microbial transport behavior in aquifers are typically of two types: forced gradient and natural gradient (Fig. 5.1). In forced-gradient experiments, an artificial flow field is created by continuous pumping of water into an injection well (divergent tracer tests, Fig. 5.1A) or continuous withdrawal from a sampling well (convergent tracer test, Fig. 5.1B) at a rate that will override ambient flow conditions in the aquifer. The injectate containing the nonindigenous or labeled organisms and the abiotic tracers is thereby forced through the aquifer to the sampling well. One disadvantage of forced-gradient experiments is that the distance over which the introduced microbes can be followed is limited because of a rapid decrease in their



Fig. 5.1. Schematic representations of the design of small-scale groundwater injection experiments involving microorganisms and abiotic tracers. A: Divergent, forced gradient. B: Convergent, forced gradient. C: Natural gradient.

concentrations caused by dilution with groundwater beyond that caused by normal dispersion. The forced dilution is a consequence of the radial flow (convergent or divergent) created by the continuous pumping or injection. The hydrological component of tracer and microbe transport is also more difficult to model in forced-gradient tests, because the flow velocity changes along the flow path. These problems may be ameliorated in natural-gradient experiments, which involve passively injecting the tracers into the aquifer and monitoring the injectate as it moves with the natural flow of groundwater past wells that have been installed along the path of travel (Fig. 5.1C). However, correct placement of downgradient sampling wells can be problematic when the hydrology of the site is poorly understood because of physical heterogeneity, geohydrologic complexity, or lack of instrumentation. Bromide is often used as the conservative tracer in *in situ* subsurface transport studies with microorganisms, because it is geochemically and biologically nonreactive, generally has a low background concentration, and can be readily assayed in the field. The differences in the concentration histories (breakthrough curves) at the sampling well for bromide and bacteria can provide useful information about the importance of physical factors that influence microbial transport: mean groundwater velocity, porosity, and longitudinal dispersion. The use of dissolved, conservative tracers also allows determination of a retardation factor for the microbial transport, which involves comparison of the time of arrival of the peak concentration of bromide to that of the microorganism that had been coinjected with bromide into the aquifer.

Retardation factors (mean flow velocity of the groundwater/mean transport velocity of the microorganisms) for the transport of microorganisms co-injected with bromide into freshwater aquifers are listed in Table 5.1. For small-scale (2-7 m) and forced- and natural-gradient tracer tests within a sandy aquifer in Massachusetts, U.S.A. (73, 74), arrival time of the maximum abundance of labeled, indigenous bacteria was nearly coincident with that of bromide, resulting in a calculated retardation factor of 1.0. In contrast, retardation factors for the other injection tests employing microorganisms were substantially less than 1.0, indicating that transport of microorganisms not immobilized by the medium was, on average, significantly faster than that of the conservative tracer and, presumably, mean groundwater flow. The apparent enhancement in transport velocity of the unattenuated microorganisms was greatest in the experiment that involved a fractured crystalline-rock aquifer (75). This phenomenon may be caused by preferential transport of organisms along preferred flow paths (large pores, fractures, and channels), because they may be excluded from the smaller pores on the basis of size. The absence of more rapid transport for the indigenous bacteria relative to bromide in the tracer tests on Cape Cod may be caused by the absence of a secondary pore structure; this is consistent with the well-sorted nature of the aquifer sediments at that site.

The results of the various *in situ* transport experiments suggest that the usefulness of halide tracers in studying the transport behavior of bacteria depends on the pore structure of the aquifer. At the Massachusetts site, the patterns of breakthrough for bacteria and bromide were similar (Fig. 5.2). Breakthrough of bromide and stained bacteria each exhibited single peaks and followed similar temporal patterns. In this system, bromide was useful in the construction of the hydrological portion of the overall bacterial transport model (74). In contrast, the breakthrough patterns observed for bacteria in forced-gradient experiments performed in other types of aquifers were quite different from that of bromide (Fig. 5.3). The earlier arrival of peak abundance and shorter duration of breakthrough for the bacteria relative to bromide (75) suggest that there are significant differences in paths of travel between bacteria and dissolved species in fractured or

Type of Distance Retardation							
Microorganism	aquifer	test	(m)	factor ^b	References		
E. coli	Fractured crystalline rock	Forced gradient	12.7	0.1	Champ and Schroeter (75)		
Indigenous isolate (Bacillus sp.)	Layered basalt	Forced gradient	27	0.6	Harvey, Voss and Souza, (unpublished data)		
Saccharo- myces cerevisiae	Sand and gravel (with clay and carbonate)	Forced gradient	1.5	0.7	Wood and Ehrlich (77)		
Indigenous	Well-sorted	Forced	1.7	1.0	Harvey et al. (73)		
bacterial population	sand and gravel	gradient	3.2	0.6 ^c			
Indigenous bacterial population	Well-sorted sand and gravel	Natural gradient	6.7	1.0 ^d	Harvey and Garabedian (7)		

TABLE 5.1. Differences in Apparent Transport Velocity Between Microorganisms and Bromide in Small-Scale Groundwater Tracer Experiments^a

^aReproduced from Harvey (104).

^bCalculated as the ratio of transport velocity at peak abundances of bromide to that of the microorganisms.

^cThe reason for the retardation factors substantially less than one for the sampler at 3.2 m is not clear. This was not observed in any of the other Massachusetts tracer tests involving bacteria.

^dValue is for both 8.5 and 9.1 m depths.

fractured-layered rock aquifers. Therefore a great deal of caution should be used in employing dissolved, nonreactive tracers in bacterial transport tests in such systems.

In addition to bromide, a number of other dissolved organic and inorganic constituents have been used as tracers in studies of microbial transport through subsurface materials. These include rhodamine WT dye used in studies involving *Escherichia coli* migrating through a sandy New Zealand aquifer (76), iodide in studies involving yeast (*Saccharomyces cerevisiae*) introduced in a sand and gravel aquifer in Texas (77), and thiocyanate and pentafluorobenzoic acid in studies involving the bacteriophage f2 moving through fractured rock (column experiment) (78). In the latter experiment, the dissolved, nonreacting tracers were retarded by a factor of 1.2–2.0 relative to fluid flow through the fractured-



Fig. 5.2. Dimensionless concentration histories for bromide (closed triangles) and fluorochromelabeled bacteria (closed circles) at a multilevel sampler 6.9 m downgradient from the point of injection. Calculated using data from Harvey and Garabedian (74). The experiment was a naturalgradient tracer test in layered, sandy aquifer sediments. A: 9.1 m below land surface; B: 8.5 m below land surface.

rock cores. Because the f2 phage remained in the fractures, estimates of dispersion and effective porosity applicable to the phage could not be calculated from data provided by the nonreactive tracer.

The advantage of using microbe-sized microspheres instead of dissolved tracers to delineate abiotic aspects of bacterial transport behavior is that the microspheres and bacteria should follow the same flow paths, even in aquifers with substantial preferred flow-path structure. The disadvantage of using microspheres is that the nature and intensity of interactions with solid surfaces can differ substantively from those experienced by bacteria. The differences in relative transport velocity and breakthrough between bromide or chloride and various types of bacteria-sized microspheres in small-scale, natural-gradient tests are listed in Table 5.2. Differences in transport behavior between microspheres and bacteria can involve the magnitude of retardation. In these experiments, bacteria at peak breakthrough were not retarded (relative to a conservative tracer), whereas most of the microspheres were subject to substantial retardation



Fig. 5.3. Dimensionless concentration histories for bromide and bacteria (*E. coli*) downgradient from point of injection in the Chalk River Aquifer (Ontario, Canada). The tracer test was a forced gradient in fractured, crystalline rock. (Redrawn from Champ and Schroeter [75].)

Colloid	Туре	Diameter (µm)	Retardation factor ^b	Relative break- through $(\times 10^{-2})^{c}$	References
Bacteria	Indigenous population	0.6	1.0	15–21	Harvey and Garabedian (74)
Plain latex	Uncharged	0.6	~1.0	0.05	Harvey et al. (73)
Polyacrolein	Carbonyl surface gps.	0.8	1.3	3.1	× ×
Carboxylated latex	Carboxyl surface gps.	0.5	1.4	0.04	

TABLE 5.2.	Differences in Apparent Transport Velocity and Magnitude of
Breakthrough	for Bacteria-Sized Colloids Relative to Bromide in Small-Scale,
Natural-O	Fradient Groundwater Tracer Experiments (Mass. U.S.A.) ^a

^aReproduced from Harvey (104).

^bCalculated as the ratio of transport velocity at peak abundances of bromide to that of the microorganisms or microspheres.

°Calculated as the integral of the dimensionless concentration history normalized to that of bromide.

(73). Only the neutral (uncharged) microspheres traveled at approximately the same rate as bromide or chloride. However, the degree of attenuation (immobilization at solid surfaces) for the neutral microspheres was several hundredfold higher than that observed for the bacteria in similar experiments. Although the polyacrolein microspheres exhibited rates of immobilization much closer to those exhibited by bacteria, they were significantly retarded, as were the carboxylated latex spheres (Table 5.2).

All of the particulate or dissolved tracers considered herein appear to have limitations for use in investigations of subsurface microbial transport behavior. In at least one aquifer having well-sorted sediments and little preferred flow-path structure, dissolved, nonreactive tracers, such as bromide, appear useful in delineating the hydrologic aspects of bacterial transport behavior (74). Even in fractured media, halides are useful as indicators of flow and may provide information regarding the potential movement of nutrients that sustain growth of moving bacterial populations. None of the bacteria-sized microspheres examined in the Massachusetts study exhibited the same combination of attenuation, retardation, or apparent dispersion as bacteria (71). However, microspheres appeared to be useful in the delineation of certain abiotic aspects of bacterial transport behavior.

Polioviruses as well as bacterial viruses have been routinely used as model indicator viruses in studies on transport through soils. An attempt was also made to use positively and negatively charged dyes (methylene blue and amaranth) and proteins with various isoelectric points (ferritin, myoglobin, and cytochrome c) to predict virus transport through soils (79). Dye adsorption to soils did not correlate with viral adsorption. However, ferritin (isoelectric point = 4.5) and cytochrome c (isoelectric point = 9.3) adsorption appeared to correlate well with the adsorption of specific types of viruses proposed by Gerba et al. (48).

4. MODELS DESCRIBING PATHOGEN TRANSPORT

4.1. Approach

Increasing contamination of soils and shallow aquifer sediments by microbial pathogens has led to a growing interest in the development of more accurate models that describe their transport. Because of the complexity involved in modeling microbial transport through subsurface environments, the approach can be as important as the mathematical descriptions. As discussed above, the extent of pathogen transport in soils may be affected by factors influencing their survival as well as their movement (5). Consequently, a number of modeling efforts involving pathogenic bacteria and viruses in subsurface environments has emphasized both persistence and transport (34, 72, 80–82). Often, a deterministic

approach is employed. However, accurate simulations of microbial transport over distances that are on larger spatial scales than those of the physical heterogeneity in soils or aquifer sediments may require a probabilistic (stochastic) approach.

Both conceptual and data-based approaches have been used in the development of models for simulating subsurface transport of microorganisms. Recently, theoretical models for describing microbial transport through soils and other porous media have been developed (83, 84). However, verification of these conceptual models using field observations is problematic in part because of the complexity of the models and the number of parameters that would need to be determined a priori. Without adequate testing, it is difficult to ensure that proper emphasis is given to the various factors controlling transport and that the mathematics are adequate. There is at least some evidence to suggest that the predictive value of the conceptual models may be limited. For example, it has been reported that when flow parameters in the theoretical model described by Corapcioglu and Haridas (84) are stressed to permissible limits, the predicted extent of bacterial transport through unsaturated soil within a 2 week period is only 0.2 m (85). In contrast, Smith et al. (86) demonstrated that E. coli could penetrate through a column of undisturbed soil to a depth of 0.3 m within 20 minutes, and transport over several meters through well-sorted aquifer sediments has been observed for labeled bacteria and bacteria-sized microspheres (73). Nevertheless, comprehensive theoretical models provide a valuable conceptual framework for the development of more realistic models that are grounded on field observations

A number of recent advances have been made in understanding the processes that govern microbial transport through soils and groundwater as a direct result of laboratory microcosm and *in situ* transport studies. Column experiments, which offer a greater degree of control than *in situ* studies, have facilitated improvements in the understanding of several factors governing microbial migration through the subsurface, including bacterial and viral adsorption to grain surfaces (65, 78, 87, 88) and bacterial growth and taxis (89–91). However, it has been shown that the transport behavior of microorganisms in aquifer sediments repacked in columns can be quite different from that observed *in situ*, even when flow velocity, porosity, and physicochemical conditions are similar (92). Consequently, the overall modeling of microbial transport through soils and aquifer sediments may require information derived from *in situ* experiments, since repacking soils and sediments into columns destroys the secondary (preferred flow-path) pore structure (86).

4.2. Bacteria

A number of biotic and abiotic factors can promote or inhibit the subsurface movement of bacterial pathogens. Several important factors that affect their



Fig. 5.4. Schematic representation of the role of biotic and abiotic processes in modeling transport of bacteria through the subsurface. "Antagonistic" and "synergistic" refer to the promotion and inhibition, respectively, of microbial transport in the direction of groundwater movement.

transport in the direction of flow are schematically depicted in Figure 5.4. An evaluation of the various factors as they relate to modeling transport of bacteria in aquifers is given by Harvey (93). Bacteria can move through soils and aquifer sediments by several mechanisms, including continuous, discontinuous (intermittent), and chemotactic migration. The relative contribution of each mechanism is not always well understood (93). Much of the modeling effort has focused on continuous transport, which assumes passive, unretarded transport of bacteria. However, discontinuous transport because of reversible sorption onto solid surfaces can be a primary mechanism of bacterial movement through the subsurface, especially where substantial distances are involved.

This discontinuous transport creates an apparent retardation of the bacteria relative to conservative, nonreactive transport. Retardation factors as high as 10 have been reported for bacterial populations traveling through porous aquifers in central Europe (72). The contribution of bacterial motility to overall transport can also be significant, because bacterial movement by taxis is much faster than that caused by random thermal (Brownian) motion. Jenneman et al. (90) reported that the rate of bacterial penetration through Berea sandstone cores in the presence of a nutrient gradient was up to eightfold higher for a motile bacterium (*Enterobacter aerogenes*) than for a nonmotile species (*Klebsiella pneumoniae*). However, for most pathogens in nutrient-limiting soils and aquifer sediments, the extent of transport may be limited largely by the geohydrology, interactions with the solid phase, and survival.

Recent efforts to model transport of enteric bacteria through unsaturated soils have employed data from laboratory column studies. Results of these modeling efforts suggest that pathogens may penetrate to greater depths in soils than was

generally believed. Germann et al. (85) used a kinematic wave approximation (employed previously in describing fluid flow through a variety of porous media) to model published data involving migration of *E. coli* through columns containing four different types of soils (94). Their model suggested that few microorganisms will be transported any deeper than 3 m when durations and intensities of naturally occurring precipitation events are considered. However, transport to depths greater than 100 m was predicted for prolonged and/or intensified precipitation events. To predict the likelihood of substantive bacterial migration in coarse soils, Peterson and Ward (95) applied Monte Carlo (probability) simulations to a one-dimensional, transport model that had been verified using earlier observations of FC penetration into a column of fine sand (96). Their simulations suggested that, under reasonable conditions, enteric bacteria can penetrate well beyond the 1.2 m depth in unsaturated soil, which is the minimum standard depth used in the installation of on-site drain fields.

Few attempts have been made to model in situ observations of bacterial transport through soils and saturated subsurface sediments. Harvey and Garabedian (74) describe a model that simulates breakthrough curves for nongrowing, fluorochrome-labeled indigenous bacteria populations injected into sandy aquifer sediments 6.7 m upgradient from the sampling well. Their model includes terms for storage, advection, dispersion, reversible and irreversible adsorption and superpositions, and separate solutions for each size class of bacteria and, where appropriate, for each uniformly conductive zone or layer along the path of travel. Because the aquifer sediments are well sorted with a fairly uniform distribution of pores, coinjection of a conservative, nonreactive tracer with the labeled bacteria facilitates construction of the hydrologic portion of the model. This deterministic approach works well for their small-scale test, because the length of the layers having similar conductive properties (as delineated by hydraulic conductivity profiles) appears to be on the order of several meters (97). However, a stochastic approach probably would be required for modeling transport over longer distances or thicknesses of the aquifer because it would be difficult to define aquifer structure deterministically at a larger scale. Both stochastic and deterministic approaches incorporating physical heterogeneity into transport models are facilitated by information concerning the variability of hydraulic and sorptive properties within the aquifer.

A major determinant in the transport of bacteria through subsurface sediments and a major component of most transport models involves their immobilization at grain surfaces. In fine grain sediments, both sorption and straining can result in significant removal of unattached bacteria as they are transported downgradient. It is generally believed that straining becomes an important mechanism for removal of microbes in groundwater when the average cell diameter is greater than 5% of that of the average grain size (98), although the range of pore sizes in the sediments is also important. In some models of subsurface microbial transport, straining and sorption are not differentiated. However, these two mechanisms of microbial immobilization can lead to different results. For example, straining preferentially removes smaller bacteria, whereas the reverse is true for sorption.

The sorptive-filtration theory (99), commonly employed to describe colloid removal during packed-bed filtration in water treatment applications, appears to offer a number of advantages in model descriptions of bacterial immobilization during transport through porous media; it is relatively simple, and it accounts for the abiotic mechanisms by which microbes contact grain surfaces and for the effect of cell size. Results of previously described experiments with differentsized microspheres suggest that sorption and not straining was primarily responsible for immobilization of bacteria traveling in the Cape Cod aquifer (73). Although there are a number of uncertainties in the application of colloid-filtration to microbial immobilization in the subsurface, it appears to be useful in multicomponent descriptions of microbial sorption in transport models (74).

In addition to immobilization, reversible sorption can be an important determinant of bacterial transport. However, it is not always clear how best to represent reversible sorption in models. In the Massachusetts study, it was found that the breakthrough curves could be accurately modeled by using either of two fundamentally different approaches to adsorption (74). The first assumes that adsorption is instantaneous and linear, but that the propensity of the bacteria for solid surfaces differs among segments of the population. The second employs a kinetic approach that assumes that all the bacteria are equally capable of interacting with solid surfaces, but that the rates of adsorption and desorption are different. The column studies described by Hendricks et al. (87) suggest that bacterial sorption onto soils may be modeled in terms of classic chemical thermodynamics.

Since bacteria that are weakly attached to solid surfaces can later desorb, particularly in response to chemical or physical changes, the ultimate limiting control of pathogen migration through the subsurface is the duration of viability. For nonindigenous pathogens in groundwater and soils, mortality can be very significant. Following an initial period of relatively constant abundance, temporal changes in abundance of displaced, nonindigenous bacteria may be modeled by exponential decline. The initial period before exponential decline may vary from days in highly contaminated water to months under oligotrophic conditions (72). In general, decay times for bacterial pathogens in soils and groundwater appear to be slow, and a one order of magnitude reduction in abundance can involve several weeks or longer (23, 100).

4.3. Viruses

Many aspects of modeling subsurface transport of bacteria are also applicable to viruses. One approach taken in modeling virus transport has involved modi-

fication of the one-dimensional solution of the classic advection-dispersion equation (101) to include sorption and decay of the virus (*e.g.*, (82). Use of the one-dimensional model is amenable to transport experiments performed in columns and in sandy aquifers where the transverse and vertical dispersivity are negligible compared with that along the direction of flow (longitudinal dispersivity). However, a three-dimensional approach may be required in modeling transport in a number of subsurface environments, particularly where there are sharp gradients in physical and/or chemical properties within the aquifer. In some instances, viruses may be restricted on the basis of size from some of the finer pore structure. For example, it was observed that MS-2 bacteriophages were excluded from 35%-40% of the void volume in a recent study involving a 'saturated soil column (78).

Sorption of viruses in transport models is generally assumed to be reversible and is often described mathematically by Langmuir or Freundlich isotherms. A retardation of the viruses relative to fluid flow may, therefore, be expected in aquifer sediments and can be calculated from known values of porosity, bulk density of the aquifer material, and the empirical distribution coefficient (81). However, there is a good deal of experimental evidence to suggest that many viruses may move faster through soils and aquifer sediments than conservative tracers of mean fluid flow, because of size-dependent exclusion of the viruses from some of the finer porosity (78). Adsorptive behavior of viruses in the presence of soils varies greatly with virus (48) and soil (47, 88) type.

Model descriptions of viral survival in the subsurface generally involve simple functions of time and temperature (82). Although temperature was found to be the most important (30), a number of other factors can affect the inactivation of viral pathogens in soils and aquifer sediments. These factors include moisture content, temperature, pH, organic matter, antagonism from soil microflora, aggregation, association with particulates, soil properties, virus type, and hydrostatic pressure (5, 102, 103).

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