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# Effect of Soil Permeability on Virus Removal Through Soil Columns

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Laboratory experiments were performed on four different soils, using 100 cm long columns, to determine the extent of virus movement when wastewater percolated through the soils at various hydraulic flow rates. Unchlorinated secondary sewage effluent seeded with either poliovirus type 1 (strain LSc) or echovirus type 1 (isolate V239) was continuously applied to soil columns for 3 to 4 days at constant flow rates. Water samples were extracted daily from ceramic samplers at various depths of the column for the virus assay. The effectiveness of virus removal from wastewater varied greatly among the different soil types but appeared to be largely related to hydraulic flow rates. At a flow rate of 33 cm/day, Anthony sandy loam removed 99% of seeded poliovirus within the first 7 cm of the column. At flow rates of 300 cm/day and above, Rubicon sand gave the poorest removal of viruses; less than 90% of the seeded viruses were removed by passage of effluent through the entire length of the soil column. By linear regression analyses, the rate of virus removal in soil columns was found to be negatively correlated with the flow rate of the percolating sewage effluent. There was no significant difference in rate of removal between poliovirus and echovirus in soil columns 87 cm long. The rate of virus removal in the upper 17 cm of the soil column was found to be significantly greater than in the lower depths of the soil column. This study suggests that the flow rate of water through the soil may be the most important factor in predicting the potential of virus movement into the groundwater. Furthermore, the length of the soil column is critical in obtaining useful data to predict virus movement into groundwater.

Land treatment of wastewater has been considered as a reliable alternative for management of wastewater. Human pathogenic viruses are known to exist in sewage effluents and sludge (23); thus, the removal of viruses after land treatment is essential to prevent the contamination of groundwater sources. The primary mechanism for virus removal from wastewater by soils is considered to be due to the adsorption of virus to the soil surface (2, 5, 13). Some physical and chemical properties of the soil that are considered to influence the adsorption of virus include texture, infiltration, cation-exchange capacity, ionic strength, organic matter content, and pH (1, 5). The ability to predict the movement of pathogenic human viruses through soil systems is a main concern in land disposal operations. However, because of the complexity of natural soils, it is difficult to establish definitive predictive correlations between virus adsorption and soil properties.

The rate of wastewater flow through the soil is controlled by soil physical properties such as texture, structure, and bulk density (4). Few studies have been conducted to evaluate the effect of soil permeability on the movement of viruses through soils. Lance and Gerba (11) found that increasing the flow rate of percolating sewage from 0.6 to 1.2 m/day through a coarse sand soil column 240 cm long resulted in more virus movement through the soil. However, increasing the flow rate from 1.2 to 12 m/day did not greatly affect virus movement. Robeck et al. (17) reported that poliovirus adsorption was not affected by flow rate until the rate exceeded 1.2 m/day. Above that flow rate, adsorption decreased with increasing velocity. Vaughn and Landry (19) studied the effects of an increased infiltration rate on virus removal in a coarse sand-gravel basin (7.6-m depth to the water table). They detected some virus movement through the basin at a flow rate of 144 cm/day and little virus retention at flow rates of 18 to 24 m/day. The purpose of this study was to further define the influence of flow rate on virus movement through soils of various compositions and permeabilities. The rates of virus removal by four soil columns were compared and correlated with the flow rates of the percolating wastewater.

## MATERIALS AND METHODS

**Virus and virus assay.** The viruses used in this study were a plaque-purified strain of poliovirus type 1 (strain LSc) and echovirus type 1 (V239; originally isolated from groundwater by the Baylor Laboratory). They were grown and assayed on the continuous buffalo green monkey (BGM) kidney cell line by the plaque-forming unit method (16). Previous experiments have shown that poliovirus 1 adsorbs well to different soils, whereas echovirus 1 adsorbs poorly (8).

**Soils.** Four different soils were used to construct soil columns. The chemical and physical characteristics of these soils are presented in Table 1. These soils have been studied previously in batch experiments to determine their abilities to adsorb viruses (8). All soil samples were air-dried and screened through an 0.8-mm sieve before use.

**Wastewater.** Secondary sewage effluent (pH 8.2 to 8.36; conductivity, 770 to 875 μmhos/cm) was applied to the soil columns. It was collected from a local activated sludge treatment plant (West University, Houston, Texas) before chlorination.

**Soil column experiments.** To investigate the movement of viruses in soil columns, air-dried soil samples were uniformly packed into polyvinyl chloride tubes measuring 100 cm long by 4 cm, inner diameter. The porosity and bulk density of each soil column are shown in Table 1. The lower 6 cm of each soil column was filled with pea gravel before packing. After leveling and smoothing the soil surface, the columns were wetted from the bottom with tap water to minimize air entrapment. The columns were then leached with 2 pore volumes of secondary effluent. In each experiment, secondary sewage effluent containing virus concentrations of $10^4$ to $5 \times 10^4$ plaque-forming units per ml was percolated through each soil column for 3 to 4 days under saturated steady-state flow conditions, using a Mariotte siphon device. Different flow rates of wastewater through each column were obtained by adjusting the height of the outlet to various levels.

The reservoir of sewage-virus suspension was replaced daily during each experiment to minimize the effect of virus die-off. The experiments were conducted at room temperature. Water samples (2 ml) were extracted daily from the ceramic samplers located at depths of 2, 7, 17, 27, 37, 47, and 67 cm and from the outlet line at 87 cm for the virus assay. The ceramic samplers are able to recover more than 90% of the seeded viruses during sampling (20). Soil columns were allowed to drain for at least 1 week before subsequent experiments were performed.

All data are plotted as $\log_{10}$ Nd/No versus soil column depth (centimeters). Nd/No is the ratio of virus concentration at different depths (Nd) to influent virus concentration (No). Linear regression analysis was used to determine the slopes of the curves which indicate the rates of virus removal from the soil columns and to evaluate the relationship of flow rate to rate of virus removal.

## RESULTS

The removal of poliovirus and echovirus by four soils as wastewater percolated through the soil columns under saturated steady-state conditions is shown in Fig. 1, 2, 3, and 4. The values presented in each plot are averages of samples taken on 3 to 4 flooding days. Of the four soils studied, the Anthony sandy loam removed 2 $\log_{10}$ of seeded poliovirus by passage of wastewater through the entire length of the column (Fig. 1). However, Rubicon sand with the highest flow rates (282–1,352 cm/day) gave the poorest removal of viruses (Fig. 4). Less than 1 $\log_{10}$ of the seeded viruses was removed when the wastewater percolated throughout the 87-cm-long soil column. In fact, virus concentration in the sewage effluent did not decrease significantly below the 17-cm depth of the soil column, indicating

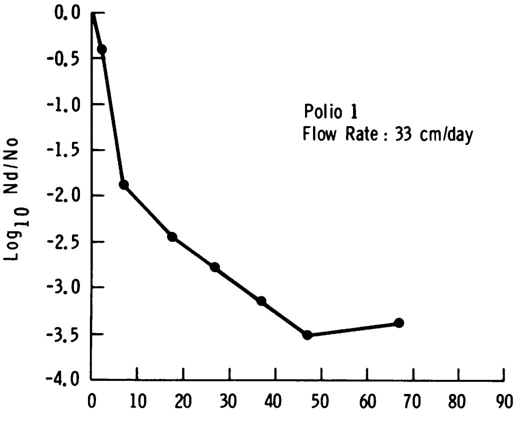

FIG. 1. *Removal of poliovirus in Anthony sandy loam.*

TABLE 1. *Characteristics of soils used*

| Soil type | Particle size distribution (%) | | | Location | pH | Cation-exchange capacity (meq/100 g) | Organic matter (%) | Column bulk density (g/cm³) | Column pore space (%) |
|---|---|---|---|---|---|---|---|---|---|
| | Sand | Silt | Clay | | | | | | |
| Flushing Meadows sand | 89 | 8 | 3 | Ariz. | 7.8 | | 0.9 | 1.55 | 42 |
| Rubicon sand | 92 | 4 | 4 | Mich. | 5.5 | 5.6 | 0.4 | 1.67 | 39 |
| Pomello sand | 89 | 8 | 3 | Fla. | 7.1 | 6.5 | 3.64 | 1.46 | 45 |
| Anthony sandy loam | 77 | 10 | 13 | Ariz. | 8.2 | 4.2 | 0.27 | 1.56 | 41 |

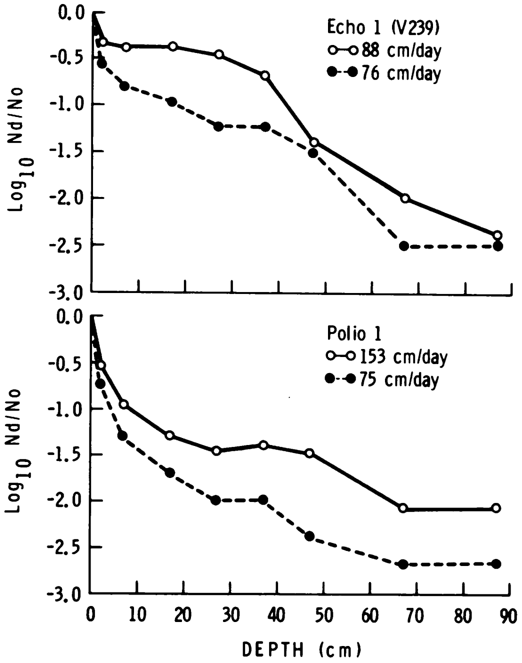

FIG. 2. *Removal of viruses in Flushing Meadows sand.*

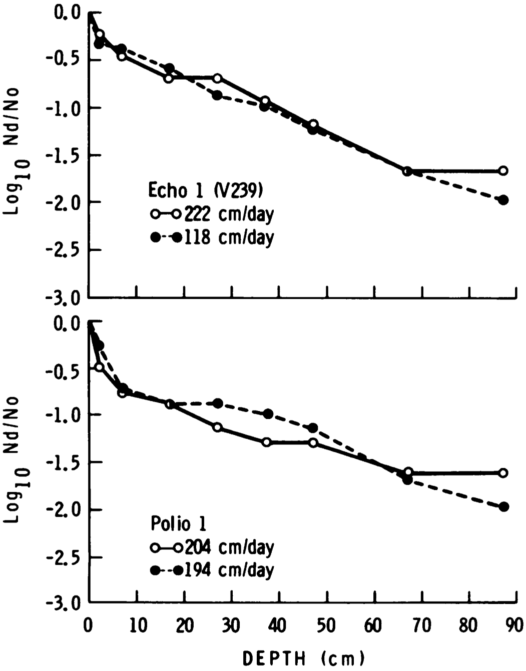

FIG. 3. *Removal of viruses in Pomello sand.*

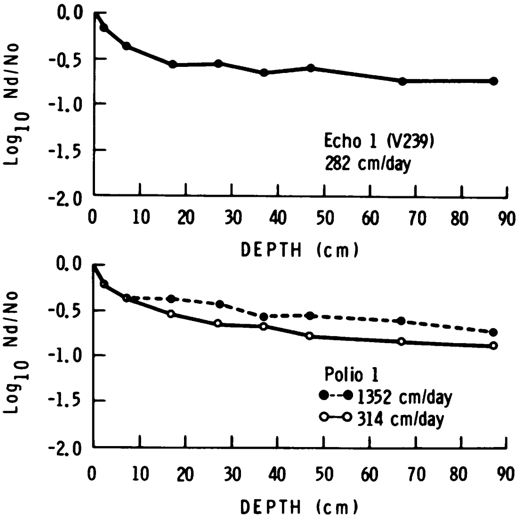

FIG. 4. *Removal of viruses in Rubicon sand.*

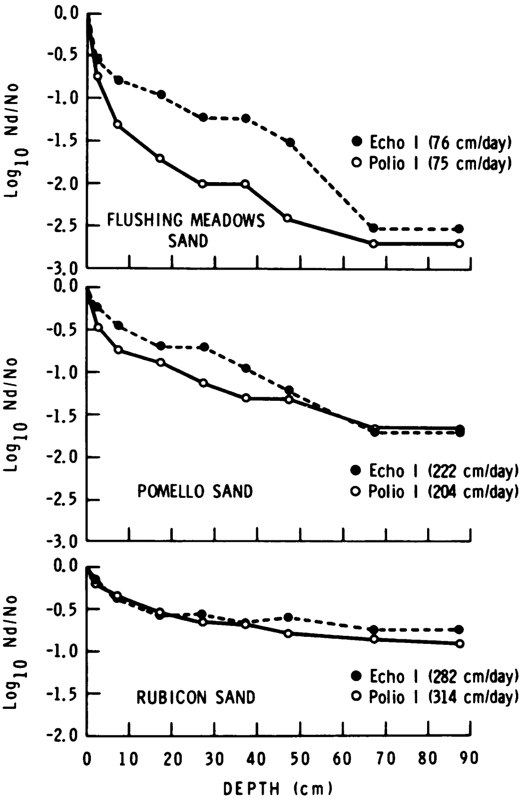

FIG. 5. *Comparison between poliovirus and echovirus removal in soil columns.*

that there was very little virus adsorption below this level in Rubicon sand.

When the data plotted in Fig. 5 for echovirus and poliovirus removals at similar flow rates were compared, the adsorption patterns of these two viruses appeared to be very similar in Pomello sand and Rubicon sand. In Flushing Meadows sand, poliovirus was removed more efficiently than echovirus above the 47-cm depth. However, the concentrations of these two viruses at the lower depths were similar. Similar results have been obtained by Lance and Gerba (11), concerning the removal of enteroviruses from wastewater percolated through soil columns of Flushing Meadows sand. The adsorption patterns of echovirus 1 and poliovirus 1 were dissimilar only above the 40-cm depths, with less echovirus being removed by the soil than poliovirus.

To determine the rates of virus removal in soil under various flow rates, linear regression analyses were performed on the data shown in Fig. 1, 2, 3, and 4. The numerical values for the rates of virus removal are shown in Table 2. Correlation and regression analyses were again used to determine the relationship between the rate of virus removal by the soil column and soil hydraulic flow rate. A significant and negative correlation was found to exist between the rate of virus removal and the flow rate of wastewater through the soil column. The $r^2$ value for rate of virus removal against flow rate was 0.83, indicating that 83% of the rate of virus removal in soil could be predicted from the soil hydraulic flow rate. The negative relationship indicates that the rate of virus removal decreased as the flow rate increased (Fig. 6). However, this linear relationship between flow rate and rate of virus removal was not significant when the data for the flow rate of 1,352 cm/day were used for analysis. Thus, virus removal by soils may at least be affected by the flow rates between 33 and 314 cm/day. Increasing the flow rate to 1,352 cm/day did not greatly affect virus removal.

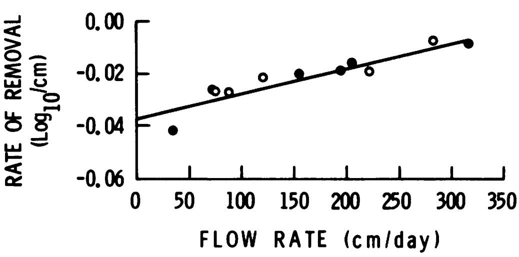

FIG. 6. *Relationship of flow rate to virus removal. The data are for both poliovirus (●) and echovirus (○) and were obtained from Table 2.*

It appears from Fig. 1, 2, 3, and 4 that the rate of virus removal in the upper part of the soil column is generally greater than in the lower depths. Therefore, the rates of virus removal above 17-cm depths and below 17-cm depths were determined (Table 3) from the data in Fig. 1, 2, 3, and 4. Statistical analysis indicated that the rate of virus removal in the upper 17-cm depth of the soil column was significantly greater than in the lower depths of the soil column at the 5% level. This indicates that a short soil column may not be suitable for predicting virus movement through the lower levels of the soil.

TABLE 2. *Rates of virus removal in soil columns*

| Soil type | Virus type | Flow rate (cm/day) | Rate of removal ($\log_{10}$/cm)[a] |
|---|---|---|---|
| Flushing Meadows sand | Echo 1 (V239) | 76 | −0.027 |
| Flushing Meadows sand | Echo 1 (V239) | 88 | −0.026 |
| Pomello sand | Echo 1 (V239) | 118 | −0.022 |
| Pomello sand | Echo 1 (V239) | 222 | −0.019 |
| Rubicon sand | Echo 1 (V239) | 282 | −0.007 |
| Anthony sandy loam | Polio 1 | 33 | −0.040 |
| Flushing Meadows sand | Polio 1 | 75 | −0.027 |
| Flushing Meadows sand | Polio 1 | 153 | −0.020 |
| Pomello sand | Polio 1 | 194 | −0.019 |
| Pomello sand | Polio 1 | 204 | −0.016 |
| Rubicon sand | Polio 1 | 314 | −0.009 |
| Rubicon sand | Polio 1 | 1352 | −0.007 |

[a] In an 87-cm-long column.

## DISCUSSION

Since human enteric viruses are not completely removed by conventional wastewater treatment processes, it is important to understand the fate of these viruses in land treatment systems. Viruses in wastewater disposed on land are assumed to react with the soil before the wastewater reaches the groundwater. However, the potential of groundwater contamination with viruses should not be neglected (18, 22). Before predictive designs for disposal of virus-containing wastewater can be established, factors influencing virus migration rates in soils must be identified and evaluated.

Soil column experiments concerning virus movement through the soil matrix into groundwater have been conducted previously in our laboratories, using sandy soils from the Flushing Meadows groundwater recharge basins near Phoenix, Arizona (12, 13). It has been suggested that these soil columns are good models for studying virus removal under field conditions (10, 11). Virus removal rates for four soils were evaluated in this study through the employment of similar laboratory soil columns containing various sampling sites at different depths of the columns. Soil columns were saturated with seeded sewage effluent in each experiment to permit a relatively fast percolation in comparison with unsaturated flow. Since the macrospore spaces account for most of the saturated water movement through soil, this study can predict maximum movement of viruses in soils.

TABLE 3. *Comparison of rate of virus removal between upper and lower depths of soil column*

| Soil type | Virus type | Flow rate (cm/day) | Rate of removal ($\log_{10}$/cm) | |
|---|---|---|---|---|
| | | | 0 to 17 cm[a] | 17 to 87 cm[a] |
| Flushing Meadows sand | Echo 1 (V239) | 76 | −0.046 | −0.025 |
| Flushing Meadows sand | Echo 1 (V239) | 88 | −0.016 | −0.032 |
| Pomello sand | Echo 1 (V239) | 118 | −0.029 | −0.020 |
| Pomello sand | Echo 1 (V239) | 222 | −0.038 | −0.020 |
| Rubicon sand | Echo 1 (V239) | 282 | −0.032 | −0.003 |
| Anthony sandy loam | Polio 1 | 33 | −0.144 | −0.020 |
| Flushing Meadows sand | Polio 1 | 75 | −0.088 | −0.015 |
| Flushing Meadows sand | Polio 1 | 153 | −0.068 | −0.013 |
| Pomello sand | Polio 1 | 194 | −0.046 | −0.018 |
| Pomello sand | Polio 1 | 204 | −0.043 | −0.010 |
| Rubicon sand | Polio 1 | 314 | −0.028 | −0.005 |
| Rubicon sand | Polio 1 | 1352 | −0.018 | −0.005 |

[a] Soil column depth.

Our studies on the removal of enteroviruses indicate that echovirus 1 and poliovirus 1 are generally removed at a similar rate during infiltration of treated wastewater. In batch studies, echovirus 1 was found to adsorb to these soils to a lesser degree than poliovirus 1 (8). This suggests that the usefulness of batch studies of adsorptive behavior of viruses in soil systems may be limited in predicting virus behavior during land treatment of wastewater under field conditions.

The migration of poliovirus and echovirus in percolating soil varied among the different soil types. It appeared that a depth of 7 cm was sufficient for effective removal of these two viruses from Anthony sandy loam. Depths of 47 and 67 cm, respectively, were required to remove more than 98% of these seeded viruses from the Flushing Meadows and Pomello sands. However, the possibility of groundwater contamination with viruses may occur in Rubicon sand if the groundwater table lies within at least 87 cm of the soil surface. The flow rates of the percolating sewage through soils appeared to have a significant influence on the rate of virus removal, at least for flow rates ranging from 33 to 314 cm/day. At the Flushing Meadows groundwater recharge basins, the infiltration rate for wastewater through soil averaged 30 cm/day (7). No viruses were detected in water samples after percolating through the soil (6). Based on the results from this study, the absence of any detectable viruses in water samples could be attributed to the high rate of virus removal from wastewater at a flow rate of 30 cm/day. Therefore, soil permeability may be one of the most important factors in determining the potential for virus movement through a soil to the groundwater. Further studies on the relationship of flow rate to rate of virus removal with many more soils and viruses are necessary.

In the past, laboratory soil column studies on virus removal have been performed by using soil columns of 20 cm in length or less (3, 9, 14, 15, 21). The fact that the rate of virus removal in the upper levels of the soil column is greater than in the lower depths suggests that using the data obtained from a short column could lead to overestimates of virus movement in the subsurface of the soil profile. On the basis of this study, a column with a minimum depth of about 60 cm should be used to more accurately predict the potential of virus migration into groundwater.

In conclusion, these results suggest that detailed information on the soil physical properties is important if accurate predictions of virus removal in soil profiles are to be obtained.

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