# Influence of Depth and Texture on the Attenuation of Migration of Bacteria in Saturated Porous Media

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#### Abstract

The influence of depth and media texture in the attenuation of pathogenic organisms in porous sand media cannot be left out in preventing groundwater from contamination; this is also important in the proper design of a good natural filter as used in water processing units. The purpose of this research is to examine the effects of media depth and texture on the attenuation of bacteria transportation in porous media. Water flow through column packed with different sizes of sand matrices were used (1 meter long, 2.79 x 10<sup>-2</sup>m in diameter). Five different depths were considered (10, 20, 30, 40 and 50 cm). 2ml of bacteria solution (Echerichia coliform-rod gram negative organism) were dropped in to the media and the effluents were collected to determine the bacteria load of the effluents. Effluents were normalized  $(C/C_o)$  to determine the coefficient of attenuation by fitting the curve of C/Co with depth. The results showed that the coefficient of attenuation increases with depth which ranged from 0.250 to2.282 for 10cm to 50cm depth. The mean value of C/Co increases with increase in particle size i.e, 0.1975 – 0.3637 for sizes between 0.150mm and 0.600mm. Statistical analysis showed that the variation of depths with bacteria attenuation is exponential which is similar to theoretical findings. The R- square value fitted linearly was obtained as 0.836 and exponentially, as 0.894. This result has direct application in determining the efficiency of sand media as a contaminant filter and as a protecting layer rather than mere quantification through chemotactic assay as applicable to aquifer bioremediation.

Keywords: Bacteria, Media depth, Media texture, Attenuation, Migration, Aquifer

### 1.0 Introduction

The dominant of groundwater resources is clear and their use, its protection, therefore, is of fundamental importance to human life and economic activity. In U. S. A., where groundwater is important in all climatic regions, it accounts for about 50 percent of livestock and irrigation water use, and just under 40 percent of public water supplies [1]. In developing countries including Nigeria, where the majority of people live in rural and sub – urban area, rivers, streams, wells and more recently boreholes which all have their origin linked with groundwater serve as the main sources of water for drinking and domestic use. The underground water supplies are usually considered safe provided they are properly located, constructed and operated according to the World Health Organization Guidelines for drinking water [2]. However, pollutants released from industrial, domestic and agricultural activities to the ground can work their way down into groundwater. Movement of water and dispersion within the aquifer spreads the pollutant over a wide area, which can then intersect with groundwater, wells or find their way back into surface water, making the water supplies unsafe.

Industrial pollution may involve seepage of used water containing chemicals such as metals and radioactive compounds, or contaminated water from damaged pipelines infiltration into the boreholes. Domestic pollution involves seepage from broken septic tanks, pit latrines, cesspools and privies. Agricultural pollution is from irrigation water and runoff water after rains, carrying fertilizers, pesticides, herbicides and fecal matter [3]. Among the mentioned pollutants sources, increasing contamination of shallow aquifer by chemical wastes and microbial pathogens has led to considerable interest in transportation of bacteria through groundwater. Transport of non – indigenous bacteria through groundwater is an

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environmental concern, in as much as the appearance of microbial pathogen in water wells has contributed significantly to the number of water borne disease outbreak [4]. In view of this, there is need to prevent the public water supply, wells and sensitive aquifers from fecal contamination through the use of hydrogeological barrier. A hydrogeological barrier is defined as the physical, biological and chemical factors singly or in combination that protect a well from pathogenic organisms. Sources of fecalcontamination in groundwater include leakage from on – site sanitation systems or sewers, animal manures, wastewater or sewage sludge applied in agriculture [5]. Physical factor considered in the application of natural hydrogeological barrier is soil properties such as depth, texture and permeability which all helped to determine the rate of groundwater recharge as well as protection from contamination. Soil acts as a natural filter. The ability of soil to lessen the amount of, or reduce the severity of groundwater contamination is referred to as soil attenuation. During attenuation, soil holds essential plant nutrients for uptake by agronomic crops, immobilizes metals that might be contained in municipal sewage sludge as remove bacteria contained in human animal wastes.

However, soil attenuation depends among other physical factors mostly on the depth and texture of the materials employed. In this research work, the effect of media depth and texture was examined on the attenuation of bacteria under saturated condition. This will not only help in providing a better solution to environmental pollution resulting from contaminant fluid from soakaway and dumping site into the source of water supply for consumption [6], but also assist in providing a better understanding of some physical parameters that affect the transport of bacteria (motility) for more accurate predictions of performances of the remediation design, as well as improving the quality of water used in fishpond if employed in water recycling processes as in " slow and rapid sand filtration systems". Factors known to affect the migration and filtration of bacteria in a porous media include, but are limited to carrier solution, ionic strength [7], electrophoretic mobility and cell hydrophobicity [8], shape and size of the porous matrix material [9]. Barton showed through experimental studies that there is relationship between obstacles (soil particle)diameter and bacteria cell motility, and independent of the porosity of the system. Experiment on measurements of bacterial penetration in sand columns with uniform particle size have shown that the effective random motility decreases with decreasing particle diameter [10]. The effects of surface characteristics upon retardation and attenuation of bacteria during transport for 0.5 - 0.8 cm size classes of microspheres had been verified [4] and this showed also that the surface characteristics of microspheres had a marked effect upon retardation during transport in the natural gradient experiment. However, the effects of groundwater geophysical properties on microbial transport have not been fully elucidated under natural condition.

The objective of this work was to examine the effects of soil texture and depth on the attenuation of bacteria through porous media under natural condition using downward flow of water through column. The bacteria use was Echerichia coliform (E - coli) –a rod and gram negative organism.

### 2.0 Methods

### 2.1 Samples Collection

The fecal coliform was collected in sterile conical sample bottles from septic tank in the Department of Microbiology and Tedder Hall, University of Ibadan. The samples were subjected to microbiological analysis for isolation and characterization. Isolation was done using the modified pour plate method. The pure culture was sterilized at 121°C for 15 minutes and cooled. This was then incubated for 48 hours at 37°C. The inoculated broth was centrifuged by dispensing them into sterilize vials and spinned at 4000 rpm. The cells were washed two to three times, after centrifugation and suspended in normal saline to avoid osmotic burst. This was then kept aseptically in the refrigeratorat 4°C. 1ml of the suspension was serial diluted and plated to determine the colony forming unit per milliliter (CFU/ml).

### **3.0** Sand Preparation

Sand samples were collected from the riverbed in OsunRiver along Iwo – Gbongan road, Osun State. Sizeable sand quantities were brought to the laboratory in polythene bags, washed with deionised water in order to remove fine organic materials that were in the process of decaying as a result of the work of soil micro-organism. The samples were then boiled in 1M hydrochloric acid for 2 hours and latter treated with 1M of NaoH to remove metallic oxide coating on the sand and equilibrate the pH respectively. The resulting sand sample was washed again in deionised water two to three times, sundried and stony pebbles removed.

The treated samples were sieved with different sieve mess to get different soil texture. The mess sizes used are 0.150mm, 0.212mm, 0.300mm, 0.425mm and 0.600mm. Each particle sizes of different texture were packed into clear jars and sterilized with dry heat in oven at  $120^{\circ}$ C for48 hours. This was done prior to use to avoid microbial contamination.

### 4.0 Column Preparation and Operation

Glass columns (1m long, 2.79cm diameter) pyrex glass were washed and sterilized with 97% ethanol. The column cylinder which was covered with muslin cloth at outlet was blocked so as to prevent the water passage. The saturated sand was then poured into the water column up to height 'h' referred to the depth equal 10cm. the column was repeatedly tapped during

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packaging to prevent any entrapment of air bubble with the pore space [11]. The cover was removed and water was allowed to pool down, until the dripping water from the column has a frequency of one drop per 10 seconds. 2ml of bacterial suspension of known concentration was dropped onto the sand bed in the column with aids of 5ml sterile syringe and this was followed by intermittent supply of 250ml of sterile water. The effluents of the column was collected and analyzed for the concentration of the bacteria load. This was done five times to have five rainfall simulations as it may occur under natural condition. Five effluents were collected differently in sterile beakers and covered immediately and subjected to microbial analysis using plate count method. The effluents concentrations of bacteria were normalized to the respective influent concentration [12]. This process was repeated for four different depths (20, 30, 40 and 50cm).

#### 5.0 **Results and Discussion**

In this study, attention was focused on the attenuation of the bacteria migration with depth and media texture using saturated sand. Flow through columns packed with various porous matrices were employed as may be a true reflection of natural groundwater movement and infiltration processes that do occur at contaminated site with such vectors [12]. Table 1 and Table 2 shows the normalized concentration of bacteria load with the depths and sizes of the porous sand media; and the coefficient of attenuation here in referred to as filtration coefficient of the media. Figure 1a and Figure 1b) and Figure 2 shows the graphs depicting the relationship between the depth and normalized concentration; and coefficient of attenuation.

Table 1: Normalized values of bacteria efficients at different depuis and partic					
Particle	C/Co	C/Co	C/Co	C/Co	C/Co
Diameter(mm)	D = 10cm	D = 20cm	D= 30cm	D =40cm	D= 50cm
0.150	0.3030	0.2061	0.1878	0.1697	0.1212
0.212	0.4167	0.3167	0.3000	0.2583	0.1313
0.300	0.5087	0.3699	0.3179	0.3250	0.1438
0.425	0.4507	0.3944	0.3521	0.1977	0.1549
0.600	0.2938	0.3277	0.4450	0.3853	0.3670

 Table 1: Normalized values of bacteria effluents at different depths and particle sizes.

**Table 2:** Coefficient of attenuation (cm<sup>-1</sup>) with depth (cm)

Depth (cm)	Attenuation coefficient ( $\mu$ ) cm <sup>-1</sup>	
10.00	0.250	
20.00	0.575	
30.00	1.576	
40.00	1.777	
50.00	2.282	

### 6.0 Media Depth and the Attenuation

The coefficient of attenuation was determined using the empirical equation which recognized that particle removal from solution was of first order in particle concentration.

$$\frac{dc}{dx} = \lambda_0 C \tag{1}$$

Where C is the particle mass concentration,  $\lambda_o$  is the coefficient of filtration and x is the distance along the flow direction. The filter coefficient was typically evaluated by measuring the bacteria concentration after flow through a filter of length 'L' and calculating  $\lambda$  from the integral form of equation (1).

$$C(L) = C_0 e^{-\lambda o}$$

Co is the influent concentration while C is the effluent concentration through media of thickness 'L'

Values of coefficient of attenuation were determined by fitting an exponential plot of normalized concentration (C/Co) to depth (X).

(2)

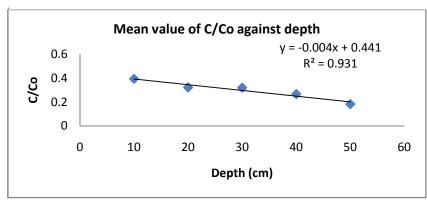


Figure 1 (a): The average value of Normalized concentration of different sizes against the depths in centimeter

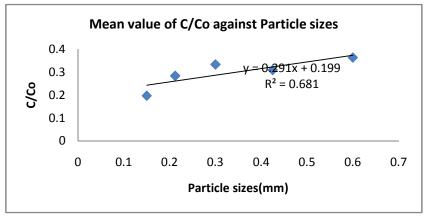


Figure 1 (b): The average value of Normalized concentration at different depth against particle sizes in millimeter

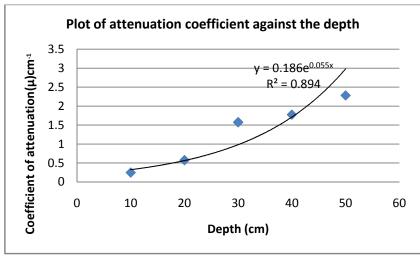


Figure 2. The plot of coefficient of bacteria attenuation with depth.

Figure 2 shows that the coefficient of attenuation increases with increasing depth, thus high efficiency of bacteria removal. The statistical model SAS was used to determine the R – square value (0.894) when the fitting was exponential and (0.836) when linearly fitted. The value 0.894 indicates that the variation of bacteria with depth is exponentially because of the high consistency in data than for the linear. This is in consistent with work in [13]; that the efficiency of a rapid filter is directly related to the media depth. There is no significant effect of soil depth on the fecal coliform removal from the fact that straining occurs over a relatively short time and medium depth had no significant effect on the pH of percolate water, these

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are in contrary to our studies [14,15]. It is a known fact theoretically, that the density of sand media increases with depth, so there would be a decrease in the permeability which will reduce the flow rate, it was reported [16], that the measured peak C/Co values for G4PRL cell decreases as velocity of flow decreased. From the statistical view, the test indicates that there is a significant relation between the depth and the bacteria transportation under natural condition. Most of the researches on bacteria migration always point towards bioremediation of contaminated aquifers using chemotactic mechanism; mechanism that involves the use of stimulant to attract the motile organisms towards the preferred direction. Theresult of this research thus provide information at protecting the aquifer from contamination from the likelihood sources, therefore provide an alternative to remediation by the use of sand porous media which necessitate the need to examine the influence of depth and texture of such media on the migration of bacteria under an intermittent mechanism.

The texture of the media has a significant effect on the bacteria attenuation. The mean value of C/Co ranges from 0.1956 to 0.3637 for particle size 0.150 mm to 0.600 mm. The normalized values of bacteria increase as particle sizes increases which shows that the larger the particle size, the higher the effluents collected, the lower the coefficient of attenuation and in [17]limited phosphorus mobility was reported. They observed that phosphorus movement was greater in coarser sands while effective Pseudomonas putida removal occurred in soil composed primarily of medium and fine sand. It was also reported in [18] that total organic phosphorus contents in soil samples increases as soil particle sizes decreased. These are all in agreement with our findings as shown in Figure 1.

### 7.0 Conclusion

The findings from this work show that there is a significant relationship between the coefficient of attenuation and the depth of the filter media. Depth considered range from 10cm to 50 cm. R – square value range between 0.0065 and 0.9661 while Pr> F range between 0.8974 and 0.0171 with standard error between 60.548 and 69.952. The mean value of normalized concentration (C/Co) is between 0.19756 and 0.36376 for particle size ranging from 0.150mm to 0.600mm. It was observed that influence of depth and texture of filter media should not be neglected when designing effective bioremediation of contaminated aquifer and they are of paramount importance when using a porous sand media to shield or reduce the severity of contaminants in groundwater.

It is recommended that further study should focus on the effect of hydraulic conductivity and porosity on the attenuation of bacteria in porous media under natural condition.

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