RESEARCHREPORT

PUBLIC HEALTH/WORKER SAFETY

Title: Quantifying Overland and Vertical Transport of Pathogens as Affected By Vegetated Buffer Strips - **NPB #01-148**

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II. Abstract:

Animal waste in the form of organic fertilizer is applied to agricultural land for crop production purposes. Pathogens in animal waste contain both beneficial as well as harmful bacteria such as Escherichia Coli O157:H7 and Salmonella. Such harmful bacteria pose human health risk if they reach to water and/or food supply. This study was designed to evaluate an effective vegetated buffer strip that can help prevent such harmful bacteria from reaching to water sources. Two sets of lysimeters one with 20% and the other with 5% were used to conduct the experiment. Both lysimeters were instrumented to monitor the surface and vertical transport of Pathogens. Soil on one side of the lysimeter with 20% slope is sandy loam, while on the other side it is clay loam. Each side of the two-sided lysimeter was divided into two sub-plots (6 m x 6.4 m), one with grass and the other with bare soil. Lysimeter with 5% slope contained clay loam soil and consisted of four sub-plots (4 m x 6.5 m), two with grass and the other two with bare soil. Plots were instrumented to collect runoff samples along the slope length (at three equidistant transects in 20% sub-plots and only at one location, 4.1 m from the edge of waste application area in sub-plots with 5% slope). Samples of runoff were also collected in a gutter at the edge of each plot for all sub-plots in both lysimeters. All plots with 20% slope were equipped with multi-sensor moisture probes to monitor real-time water content through the soil profile. No moisture sensors were installed in sub-plots with 5% slope. Bovine manure and Swine slurry, each at separate times was applied at the top of the slope of each plot in one-foot strips. Rainfall was simulated at 61 mm/hr and 81 mm/h, on 20% sub-plots and 5% sub-plots, respectively, using a portable rainfall simulator. Surface flow was measured and sampled at 2-5 minute intervals at three different transects in 20% sub-plots and one location in 5% sub-plots, and in the gutter for all sub-plots. Twenty four hours after simulations, soil samples were taken at incremental depths (0-50 cm).

Runoff and soil samples were analyzed for fecal coliforms (FC), E. Coli, and Salmonella. Results indicated that vegetated buffer strips retarded the flow, thus reducing the runoff of water and bacteria in both lysimeters and in all soil types examined in this study. Runoff from the bare

clay loam sub-plots contained virtually all the pathogens in both fresh manure and the swine slurry under both slopes, but runoff from the vegetated clay loam sub-plots had only 0.6 percent of the initial pathogen population in lysimeter with drastic slope of 20%. In comparison, runoff from bare sandy loam sub-plot with 20% slope contained 24 percent of the initial pathogen, but runoff from the vegetated sandy loam sub-plot had none. Similarly, vegetation seemed to filter both E.Coli and Salmonella in sub-plots with 5% slope, thus none or minor amounts of these pathogens were detected leaving the edge of the plot. This study concluded that even for drastic slopes such as the slope of 20% used in this study, grass buffer strip virtually stopped the movement of the pathogens. Results of this study has the real world implications in that such buffer strips may be implemented along the edges of the agricultural fields, especially in the regions of the country receiving animal waste, thus preventing the transport of pathogens to the surface bodies of water. Results also indicated that pathogens do not move much beyond 10 cm of the soil profile even in the vegetated sandy loam soil with the highest infiltration rate, thus indicating no threat to the groundwater. Results so far lead us to conclude that having a buffer strip of about 3-4 meters at the edge of the fields with sandy loam texture and about 4-5 meters at the edge of the fields with clay loam soil will prevent the delivery of pathogen from the fields receiving animal waste to the adjacent stream systems.

III. Introduction:

Recent evidence indicates that there are multiple environmental sources of pathogenic organisms including humans, companion animals, wildlife, and farm animals. Nonetheless, land application of animal waste is frequently cited as a major source of water-borne pathogen contamination. Land application of animal waste is recommended to recycle nutrients and organic matter to enhance crop productivity and soil quality. However, contaminated waste may pose a public health threat if parasites and/or pathogens are transported to potable or recreational waters. Potential modes of water contamination include leaching to groundwater via preferential flow and/or runoff to rivers, streams, reservoirs, etc. There is insufficient information, however, to assess the relative magnitude or significance of these transport pathways or to recommend effective mitigation strategies.

The risk associated with surface water contamination by animal waste is, in part, a function of waste volume, site hydrology, and proximity to surface waters. Large, confined animal feeding operations (CAFOs) generate substantial volumes of animal waste held or applied within a relatively small area. High rates of land-applied manure, particularly where rates exceed soil assimilative capacity, increase the risks for surface or groundwater contamination. Risk may be offset by low rainfall, dryness, minimal land slope, relative isolation of the animal population, and methods of composting, storing, and spreading manure.

Numerous variables affect rates and extent of pathogen transport including slope, soil characteristics, vegetation and rainfall intensity/duration. In addition, because pathogens are particulate in nature, factors affecting adhesion to soil particles or entrapment also impact transport. Pathogens applied or deposited onto soil surfaces may infiltrate into the soil profile or, alternatively, may runoff to surface waters. Since both processes can occur simultaneously, a thorough understanding of the controlling factors is critical in predicting which process will predominate. Agricultural management practices which decrease runoff and sedimentation (e.g., vegetative buffer strips) have been proposed as a means of reducing pathogen transport to surface waters. These practices reduce runoff by decreasing overland water flow and providing additional soil surface for infiltration (Magette et al., 1989). A more comprehensive understanding of how environmental parameters control pathogen transport is required in order to evaluate design criteria for remediation methods such as vegetative buffer strips.

A substantial body of literature exists describing both leaching/infiltration of microorganisms through soils and runoff to surface waters. Early studies documenting subsurface bacterial transport from septic effluents have been reviewed by Hagedorn *et al.* (1981) and Bitton and Harvey (1992). These studies document bacterial transport from a few meters to 830 meters depending on soil texture (sand vs. gravel), water content (saturated vs. unsaturated) and time. Recently laboratory-scale studies have focused on elucidating soil parameters affecting leaching. These studies indicate that the predominant factors affecting leaching in tilled soils are soil structure/texture and porosity/bulk density in conjunction with bacterial size (Gannon *et al.*, 1991a,1991b; Huysman and Veerstracte, 1993; Tan *et al.*, 1991), while in no-tilled soils the predominant factors are distribution and continuity of macropores (preferential flow pathways) in conjunction with initial water content (McMurry *et al.*, 1998; Paterson *et al.*, 1993). In general, higher leaching rates are observed in no-till than tilled soils (Smith *et al.*, 1985; van Elsas *et al.*, 1991). Limited research suggests that parasites (*Cryptosporidium parvum*) leach more slowly through soils than bacteria (Mawdsley et al.,1996a, 1996b; Rouhi et al. unpublished data). Still, quantitative relationships have yet to be established describing rates or extent of pathogen leaching at the field or watershed scale.

Studies addressing runoff of bacteria to surface waters have been reviewed by Baxter-Potter and Gilliland (1988), Khaleel *et al.* (1980), and Patni *et al.* (1985). The major factors controlling bacterial densities in runoff are manure management practices, wildlife activity, channel/bank storage, microbial mortality, and watershed hydrology. However, due to the complexity of interactions and nonlinear effects, few quantitative relationships have been established between the various factors. For example, several researchers have reported the absence of a significant correlation between fecal coliform densities in surface waters and the presence of beef or dairy operations; similar results have been reported for karst aquifers (Pasquarell and Boyer, 1995). This could be due to effective livestock/manure management practices and hydrologic characteristics, which do not favor transport, high microbial mortality rates, high background concentrations from wildlife, or a combination of the above. In particular, very few studies have addressed the role of fecal coliform/pathogen contributions from wildlife (Atwill *et al.*, 1997). Similarly, studies suggest that stream/river banks and channels may serve as reservoirs for fecal coliforms and that increased concentrations observed during storm events are due to microbial re-suspension during periods of turbulent flow (McDonald *et al.*, 1982).

We are unaware of any studies examining both vertical and surface transport simultaneously. Since both processes occur simultaneously under natural conditions, a thorough understanding of the controlling factors is critical to predicting which process will predominate. This lack of understanding is, in part, responsible for the limited success in developing design criteria for mitigation strategies, such as vegetative buffer strips (VBS). Although the basic principles of VBS are well established, results from previous VBS studies are contradictory. For example, using a VBS, to purify feedlot runoff, Young *et al.* (1980) observed that effluent fecal coliform and fecal streptococci concentrations were reduced by 61% and 87%, respectively. Similarly, Schelinger and Clausen (1992) observed a 30% decrease in bacterial concentrations from a dairy manure detention pond after transport through a VBS. Conversely, Chaubey *et al.* (1994) did not find any significant decrease in effluent fecal coliform concentrations from swine manure applied to VBS of differing lengths; comparable results were reported by Srivastava *et al.* (1996) on the same plots with different manure application rates. Note that all of the above studies were conducted on VBS designed primarily to minimize runoff of nutrients and sediments; no attempt was made to optimize VBS for the interception/detention of microorganisms.

Bovine manures have been documented to contain various pathogenic microorganisms. Field application of manures can potentially results in surface and groundwater contamination.

Consequently manure management practices are required to prevent or minimize the deterioration of water quality from manure-born pathogens. One method to reduce transport of pathogens to water sources is using vegetated filter strips. The majority of published findings support, to various degrees, the role of filter strips in removing pathogens from overland flow (Coyne et al., 1995; Crane et al., 1983). Moore et al. (1988) recommended that vegetative filter strips should be at least 3.0 m wide and have a slope of 0 to15%. Conflicting results on the effectiveness of vegetative filters on removing bacteria (Srivastava et al., 1996; Hunt et al., 1979; Jenkins et al., 1978; Dickey and Vanderholm, 1981) indicate that surface conditions, (e.g. soil type, types of vegetation), manure application technology, as well as the rainfall parameters may affect the efficiency of filter strips. The goal of this project was to elucidate critical soil, hydrological and meteorological parameters controlling rates and extent of simultaneous pathogen overland flow and vertical transport. Results will be used to establish design criteria for vegetative buffer strips (VBS) to minimize pathogen transport to surface waters.

IV. **Objectives**:

The goal of this project is to elucidate critical soil, hydrological and meteorological parameters controlling rates and extent of simultaneous pathogen overland flow and vertical transport. Results will be used to establish design criteria for vegetative buffer strips (VBS) to minimize pathogen transport to surface waters.

V. **Procedures**:

Materials and Methods

The experimental site was located in Beltsville, MD. A two-sided lysimeter (12.7 m wide by 21.5 m long) with 20% slope on both sides was instrumented to monitor the surface and vertical transport of pathogens (Fig.1). Soil on one side was sandy loam down to 60 cm, while on the other side it was clay loam to the 30 cm depth, and then sandy loam down to 60 cm. Both sides had gravel layer below 60 cm. The organic matter content of the top 10 cm from five random samples was determined to be 2.7±0.9% and 1.7±0.9% at the clay loam and sandy loam soils, respectively. Soil pH in the same samples was 5.9±0.6 and 6.2±0.3% in the clay loam and sandy loam soils, respectively. Both sides were divided into two sub-plots (6m x 6.4m), one bare soil and the other planted with orchard grass. A V-notch weir was installed at the end of the gutter to measure total surface runoff (Fig. 2). Nine mini-flumes were installed along three transects in each plot to measure spatial distribution of runoff. Mini-flumes were connected to sampling funnels outside the lysimeter via food-grade tygon tubing buried about 20 cm depth below the soil surface. Four capacitance-based soil moisture sensors (SENTEK) were installed at each plot. The sensors measured volumetric water content within 5-15, 15-25, 25-35, and 35-45 cm depth intervals with 2 min measurement frequency and were connected to the data logger outside the plots. A four-nozzle rainfall simulator was calibrated at the site to maintain the uniformity coefficient of approximately 90% for rainfall intensity of 61 mm h^{-1} (a 10-year, 24-hour storm for Maryland).

Another lysimeter (12.5m x 18.7m) was also constructed at 5% slope and filled with clay loam soil. Four sub-plots (2 bare and 2 grassed with 4 m in width and 6.5m in length) were delineated an instrumented to collect overland flow samples both at 4.1m from the edge of the slurry (swine slurry) application area and at the outlet of the plot. Sub-plot length in each case was in the direction of slope. Funnel and v-notch weir setup used in this lysimeter was similar to that used in the lysimeter with 20% slope. No soil moisture probes were used in this lysimeter.

Experimental Procedures

The evening prior to rainfall simulations, a 30 cm wide strip of bovine manure was uniformly applied to the top of plots in the lysimeter with 20% slope. FC concentrations were determined in manure at the time of application and just prior to rainfall simulations. During simulations, all runoff from mini-flumes as well as gutter were collected for every five minutes interval. The initial populations of FC in manure and swine slurry were measured in all plots right before the initiation of rainfall simulation. The duration of experiments for subplots with 20% slope was 55 min for bare soil plots, 90 min for vegetated clay loam, and 120 min for vegetated sandy loam, during which 61 mm/h of rainfall was simulated. Rainfall intensity of about 80 mm/h for a duration of 60 minutes was simulated on all subplots with 5% slope.

After simulations in both lysimeters, water samples from mini-flumes were stirred and 50-ml sub-samples were taken for laboratory analysis. The same afternoon or the following day, soil samples were taken adjacent to mini-flumes and in the manure application strip at incremental depths using a 2.54 cm ID core sampler. The remaining manure/swine slurry residue was collected from the application strip in bare plots for determining the remaining FC population.

Runoff samples were centrifuged at 100 x g in 12-ml conic tubes for 10 minutes. Two replicated 50 µL sub-samples of the supernatant were placed onto Mac Conkey Agar using a Spiral Bio-Tech auto-plater. Plates were incubated at 44.5º C for 24 hours. A Synoptics Limited Protocol Colony Counter was used to count FC in Colony Forming Units (CFU) in each plate. Soil samples were diluted 1:10 with distilled water, dispersed for 2 minutes at high-speed blender, and then processed as described above for runoff samples.

The standard error of the fraction of FC in runoff s_f, was computed as:

$$s_f = f\sqrt{\frac{(s_Q)^2}{Q} + \frac{(s_{p-r})^2}{p-r}} = f\sqrt{Q\left(\frac{s_a}{a}\right)^2 + \frac{(s_p)^2 + (s_r)^2}{p-r}} \qquad (1)$$

where, $f = Q/(p\text{-}r)$ is the fraction of FC in runoff, p is the average count of FC before the simulation, r is the average count of FC remained at the surface of the application area, q is the average count of FC in runoff, Q is the average adjusted count of bacteria, $Q=q/a$, a is the average extraction efficiency, s denotes the standard error of the value in subscript.

Modification of the Project from Original Proposal:
The only modification that we have done is to use a 20% slope lysimeter instead of 10% slope. We felt that 20% would provide the critical hydrologic conditions for examining the pathogen transport, especially under grassed covered plots.

VI. **Results:**
Results of hydrology and pathogen transport are presented for both lysimeters (e.g., 20% slope and 5% slope). Since soils and slopes were different from one lysimeter to the other, results are reported separately to avoid the confusion.

Hydrology:

A. For Lysimeter with 20% slope: The hydrographs of Fig. 2 indicate that both in bare clay loam and in bare sandy loam, runoff started much earlier as compared to the vegetated plots, and they both had very sharp rising limb in the early minutes of simulation. Results also indicate that on the bare clay loam after a sharp rise within the first 10 minutes, the runoff gradually was increasing until the rainfall simulator was turned off. On the bare sandy loam, runoff maintained almost a constant rate after a sharp rise in first 15 minutes of the test. Surface sealing of the soil seemed to develop at the bare clay loam site, thus causing decreased infiltration and increased runoff rate. Soil moisture probe (SENTEK) measurements support this conclusion (data not shown). The very flat hydrograph on vegetated sandy loam soil also indicates the significance of soil texture on runoff and infiltration under vegetated surfaces. Results from both vegetated plots show that vegetation drastically attenuates the surface flow of water (less runoff). Quantitatively, data show that percent of simulated precipitation lost to surface runoff was 70% and 7 % for bare and grassed plots, respectively. This indicates that grass enhances infiltration and reduces runoff significantly.

B. For Lysimeter with 5% slope: Figure 3 shows the runoff hydrographs for all the plots (both bare and vegetated) in the lysimeter with 5% slope. As the data indicate plots 1 and 3 with vegetated surface had the lower runoff than the plots 1 and 4 (bare plots). Considering that the soil was the same in all plots, the only culprit to cause difference in recorded runoff hydrographs is the surface cover. Vegetation may have helped in several ways to enhance infiltration and reduce runoff. First, vegetation provides a cover for the surface, thus preventing the compaction and sealing of the surface of the soil and preventing the reduction in soil's infiltration capacity. Second, biological loosening effect of the grass roots causes several fold increase in soil's hydraulic conductivity (Shirmohammadi et al., 1984) and enhance infiltration rate. Additionally, vegetation retards the flow velocity and provides more chance for water to infiltrate. Results of Figure 3 also show that runoff initiated almost immediately after the initiation of rain simulation in bare plots (plots 1 and 4), but it did not begin until about 10 minutes and 13 minutes after the initiation of rain simulation in plots 3 and 1, respectively.

Bromide:

A. For Lysimeter with 20% slope: Data was also collected on the fate of bromide as a tracer at the sampling location. Data indicate definite spatial variability in Bromide concentrations on the bare plot (plot1) collected at each row along the slope (Figure 4). Figure 4 also shows that funnels 1, 2, and 3 (top row) had similar bromide concentration to those collected in funnels 4, 5, and 6 (middle row). Figure 5 shows the average Bromide concentrations at each row as a function of time for the bare plot (plot 1). Again, data indicate spatial variability in the longitudinal direction, where middle row seemed to have a higher peak concentration than the top row. However, the bottom row always had a lower bromide concentration than both top and middle rows. Similar results regarding the spatial variability were obtained for the grassed plot as well. Bromide is a soluble non-reactive tracer, thus spatial variability observed for this tracer may indicate that flow on the surface moves preferentially as a concentrated flow rather than uniform sheet flow. Results of bromide study show that water and pollutants (both non-reactive and reactive) move preferentially in the landscape, which may be a phenomenon to be considered in modeling transport processes through such systems.

B. For Lysimeter with 5% slope: Similar to sub-plots at 20% slope, data on transport pattern of bromide was collected in for both vegetated (Figure 6) and bare sub-plots at 5% slope. Breakthrough curves in Figure 6 show that bromide concentrations peaked within about 25 to 20 minutes of the simulated rain event both at 413 cm (funnels) and 620 cm (gutter) from the edge of the application area. It is apparent that while maintaining similar breakthrough pattern, grass

filter reduced the peak bromide concentrations leaving the edge of the plots by over two fold. It is apparent that peak concentrations observed in sub-plot 3 are twice as much as those observed in plot 1 despite apparent similarity in soil, slope, and surface cover. This difference may be attributed to preferential movement of water and dissolved compound such as bromide on the land surface due to natural difference in depressions and possibly density of grass between two sub-plots.

Figure 7 shows the breakthrough curves for bromide runoff for sub-plots 2 and 4 with bare surfaces, but with the same soil and slope conditions of sub-plots 1 and 3(vegetated surface). Data indicate that peak concentrations were observed within 5-7 minutes of the initiation of the simulated rain event in bare surfaces. This is a retardation time of peak arrival of about 10-15 minutes compared to vegetated surfaces. It is also apparent that peak concentrations observed under bare sub-plots were at least an order of magnitude higher than those observed under vegetated sub-plots, thus indicating attenuation capability of the grassed filters. Figure 7 also shows that most of the bromide was flushed out within10-12 minutes of the initiation of rain event on bare sub-plots, indicating negative impact of bare surfaces on runoff of dissolved chemicals in landscape. Comparing data presented in figures 6 and 7 makes it apparent that vegetated surfaces retard the overland flow of water and dissolved chemicals such as bromide and provide more opportunity for the water to infiltrate into the soil profile. Soil profile may then act as a filter for some of the dissolved chemical and remain within the soil pores and be used by vegetation.

Fecal Coliforms:

A. For Lysimeter with 20% slope: Concentrations of FC in runoff decreased with time at various distances from the source on both bare clay loam (Fig. 8a) and bare sandy loam (Fig.8b). Maximum FC concentrations in runoff decreased with the distance from the source. Comparison of Figs. 8a and 8b show that the pattern of decrease is affected by soil texture. On the bare clay loam plot, concentrations of FC at 95, 285, and 485 cm from the ridge were similar and higher than the FC concentration at 600 cm from the ridge. On the contrary, on the bare sandy loam plot, FC concentrations were similar at 285, 485, and 600 cm from the ridge, and all were less than the concentrations at 95 cm from the ridge. Unlike the bare soil, vegetated soil surface creates a much less uniform transport pattern with respect to time for FC (Fig. 8c). Only at the 95 cm distance from the ridge, the pattern of FC transport seems to have a steady decrease with time. Vegetation changes transport patterns and levels of FC concentrations much more significantly than the soil texture does.

FC levels in the soil profile decreased with depth (Fig. 9). The exponential decrease at a distance of 95 cm from the ridge was similar to what was observed by Fleming et al. (1990). At longer distances, FC concentrations decreased fast in the top layer of the soil profile and exhibited much slower decrease in the bottom layers of the soil profile. FC moved to a deeper depth in the soil profile on vegetated sandy loam soil (Fig.9c) than in vegetated clay loam soil (Fig. 9a). Soil texture seemed to play an important role for FC vertical transport. Vegetation promoted vertical FC transport close to the source, but did not seem to have much effect at distances away from the source. This may be due to the fact that very little FC was transported far away from the source via surface runoff (Fig. 8c). The same FC contents were found at 30-cm depth in vegetated sandy loam soil (Fig. 9c) and at 10-cm depth in vegetated clay loam soil (Fig. 9a) at a 95 cm distance from the ridge. No significant FC was found in the surface runoff at larger distances from the source in vegetated sandy loam, indicating most of the FC was transported vertically at the top of the plot near the source.

The mass balance for water and FC are shown in Table 1. Data in Table 1 show that runoff was decreased by 81.2% (92.6-11.4) in vegetated clay loam, and by 57.3% (59.8-2.5) in vegetated sandy loam soil as compared to bare clay loam and bare sandy loam, respectively. Both vegetation and soil texture affected water balance. Much higher reductions of surface runoff are observed where vegetation is coupled with soil textures that favor infiltration. Table 1 also shows that vegetated filter strips significantly attenuated FC in the surface runoff, thus reducing its population to non-detectable levels in the vegetated sandy loam plot.

B. For Lysimeter with 5% slope: For sub-plots in this lysimeter, E-Coli and Salmonella were measure and the results are presented in the following section.

E-Coli and Salmonella:

A. For Lysimeter with 20% slope: Genetically engineered salmonella failed to show itself under microscope in the samples collected from the vegetated sub-plots. This may be attributed to the existence of other microorganisms in the grass matrix. Obviously, organisms in the grass have identical morphology to that of salmonella, thus hindering its count under the microscope. Results indicate that on bare soils both E-Coli and Salmonella behaved very similar (Figures 10a and 10b). Data in Figures 10a and 10b show that E-Coli and salmonella have similar transport characteristics, which helped us to estimate the salmonella count in the vegetated plots based on the E- Coli count using their correlated relationship.

The concentration ratios verses time on both bare clay loam (Fig. 11a) and bare sandy loam (Fig. 11b) indicate the double exponential release rate for both *E-coli* and *Salmonella*. The same results also indicate the fast release trend in the first 10 minutes of the simulation event and slow release trend for the rest of the simulation period. Such results are indicative of the population dynamics of both pathogens with respect to their availability for transport at different time steps during the simulation process. Contrary to the bare soils, there is no unique flow pattern for *E-coli* and *Salmonella* on vegetated soils (Figure 12). These results clearly reflect the effect of vegetated surface strip in distortion of the overland flow pattern due to canalization and concentrated flow regime.

Results from vegetated plots showed colonies resembling *Salmonella* all over the plots. It was determined that those organisms were caused by the residual of mowed grass that remained on the plots prior to the experiment. Further experiments were necessary to be conducted to clarify these colonies. Results from entro-tubes in the laboratory indicated that *Salmonellas* were not present in runoff samples collected from vegetated plots, thus reflecting the consistency of *Salmonella* detection with that of *E-coli*. Samples of culture of *E.coli* and *Salmonella* with random samples of each from runoff were also examined at 400X with a Zeiss Axioskop epifluorescence microscope equipped with a fluorescein isothiocyanate-Texas red dual wavelength filter. The results of this experiment also indicated that those colonies were not *Salmonellas*. Other physiological characteristics tests were also conducted to characterize these organisms. Results of this later test also showed that the growth on specific media for *Salmonella* was not *Salmonella*. Based on the results of bare soils and the laboratory experiments, it was then concluded that, both *Salmonella* and *E-coli* have very similar release rate and transport patterns in all plots. By knowing the fact that these two organisms structurally and morphologically are very similar, the final conclusion was that the transport dynamics of *E-coli* and *Salmonella* is expected to be similar as it was predicted.

B. For Lysimeter with 5% slope: Results of the relative concentration of both Salmonella and E-Coli bacteria versus time are depicted in figures 12a, 12b, 13a and 13b for plots 1 (vegetated), 2 (bare), 3 (vegetated) and 4(bare) for the lysimeter with Clay Loam soil and 5% slope. Data in

figures 12a and 13a indicate that vegetation retarded the movement of both bacteria, thus resulting in reduced concentrations both at 413 cm and 620 cm from the edge of the swine slurry application area. It should also be noted that despite variability in relative concentrations of both pathogens between plots 1 and 3 (both vegetated), vegetation retarded the movement of pathogens and produced less steep breakthrough curves. However, breakthrough curves observed for bare surfaces (plots 2 and 40 seem to be relatively similar and have steep gradients (12b and 13b). Data also indicates that bulk of pathogen discharge the plots with bare surfaces within first 10 minutes of the simulated rain event (Figures 12b and 13b) as compared to about 30 minutes in vegetated plots (Figures 12a and 13a). Results of this study concludes that grass filters provide an excellent buffer between the waste (i.e., swine slurry) application area and the stream system. Results also show that vegetated filter strips of about 4-5 m (13 -16feet) should provide a sufficient buffer in Clay Loam soils with 5% slope.

VII. Discussion: Results of this study showed that Vegetated filter strips reduced both surface runoff and pathogen transport significantly. Results also indicated that combination of porous sandy loam soil and vegetated filter strips played a major role in diminishing the overland transport of water and pathogens. On the contrary, bare surfaces offered no resistance to flow and pathogen transport. One may conclude that having a vegetated filter strip of about 3-4 meters for sandy loam soils at the bottom of even steep slopes such as 20% will prevent pathogen loss from the edge of the filter to the surface bodies of water. However, width of the vegetated buffer strip will depend on both slope and soil texture. Results also lead us to conclude that for fine textured soils such as clay loam soils (representative of Midwest soils) with even small slopes of about 5%, vegetated buffer strips of about 4-5 m at the bottom of the slope are required to prevent pathogen transport to water bodies. Finer the soil texture, lower is the infiltration rate, thus providing more opportunity for surface flow and surface transport of contaminants (e.g., bacteria). In such cases, width of vegetated filter strips need to be wider than those designed for coarse textured soils such as Sandy soils tested in this project. We feel that results of this study may be used as a recommendation for implementing vegetated buffer strips at the edge of agricultural fields that are adjacent to the stream systems and are receiving either bovine manure or swine slurry as a nutrient source for crop production.
Results also indicated that Salmonella and E- Coli have similar transport characteristics. Results obtained in this project show that by determining the transport characteristics of E-Coli, one may be able to estimate that of the Salmonella and save both time and money regarding sampling and analysis.

VIII. Lay Interpretation: Farmers use animal waste (e.g., bovine manure and swine slurry) to fertilize their fields and pastures. However, existence of pathogens such as E-Coli and Salmonella in the animal waste poses environmental and health problems. A cooperative study between USDA-ARS, Animal waste Pathogen Laboratory (AWPL), USDA-ARS, Environmental Quality Laboratory, and the University of Maryland was conducted to examine the effectiveness of vegetated filter strips in retarding the runoff of pathogens to the edge of field. To perform this study, two lysimeters with two different slopes of 20% and 5% were constructed. The lysimeter with 20% slope is a two –sided lysimeter, one side with sandy loam soil and the other with clay loam soil. The lysimeter with 5% slope was only constructed with clay loam soil to represent the soils and topography of the Midwest USA. Then, two grassed plots and two bare plots were constructed and formed in each lysimeter. All plots were exposed to the natural climatic conditions for grass to grow. An in-house designed and built overhead rainfall simulator was used to simulate desired storm events and sample for bacteria transport during the simulated rain event at different time intervals. Both bovine manure as well as swine slurry were inoculated with genetically engineered E-Coli and Salmonella strands and applied along the top of each plot right before rain simulation. Then, water samples were collected at different distances from the

edge of the waste application area along the plot slope. Samples were also collected in a gutter at the edge of each plot to measure both runoff and pathogen population leaving each plot under the specific simulated rain event.

Grass filters were very effective in removing pathogens in both lysimeters. Combination of sandy loam soil with high infiltration capacity and vegetation reduced the pathogen discharge to zero at the edge of the plots even under drastic 20% slope and once in 20 year storm event for Maryland area. Vegetated filters were also very effective in clay loam soil (low infiltration capacity) with 5% slope, but not as effective as in the porous sandy loam soil. Results of this study concluded that using vegetated filter strips of about 3-4 meters (10-13 feet) in porous sandy soils and 4-5 meters (13-16 feet) in clay loam soils may be a safe buffer width at the edge of agricultural fields that are adjacent to the stream systems and are receiving animal waste as a source of nutrient for crop or pasture growth.

Table 1. Components of mass balance of water and fecal coliforms for the lysimeter with 20% slope.

Component of the balance	Clay loam		Sandy loam	
	Bare	Vegetated	Bare	Vegetated
Water, cm				
Rainfall	5.6	9.2	5.6	12.7
Runoff,%	92.6	11.4	59.8	2.5
Bacteria, 10^9CFU				
Applied	7.5	11	56	32.3
Remained after rain	3.3	0	4.4	0
Runoff	5.1	0.1	12.8	ND
Soil	0.4	9.9	18.6	3.6
In runoff, %	121.4(24.2)$^{\perp}$	0.9(0.6)	24.4(19.2)	ND

$^{\perp}$Standard error in parentheses; ND - not detected

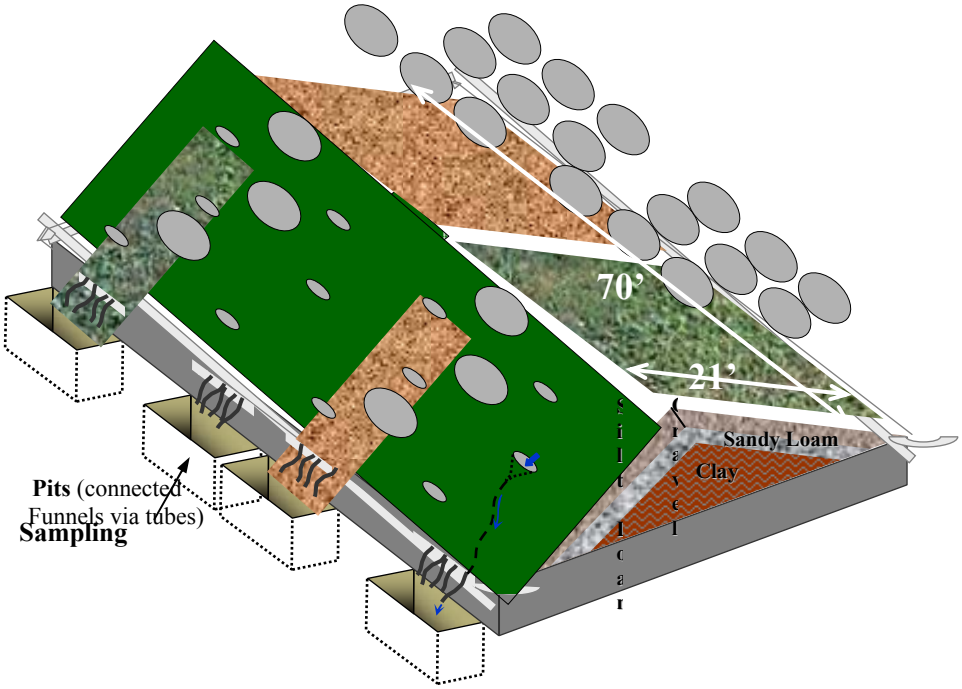


Figure 1. Schematics of the Lysimeter.

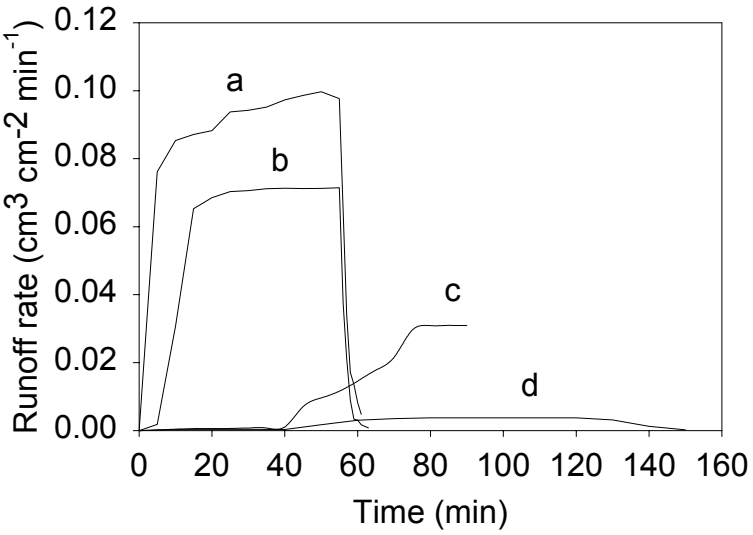


Figure 2. Runoff hydrographs for sub-plots with 20% slope; a,c - clay loam, b,d, - sandy loam, a,b, bare plots, c,d - vegetated plots.

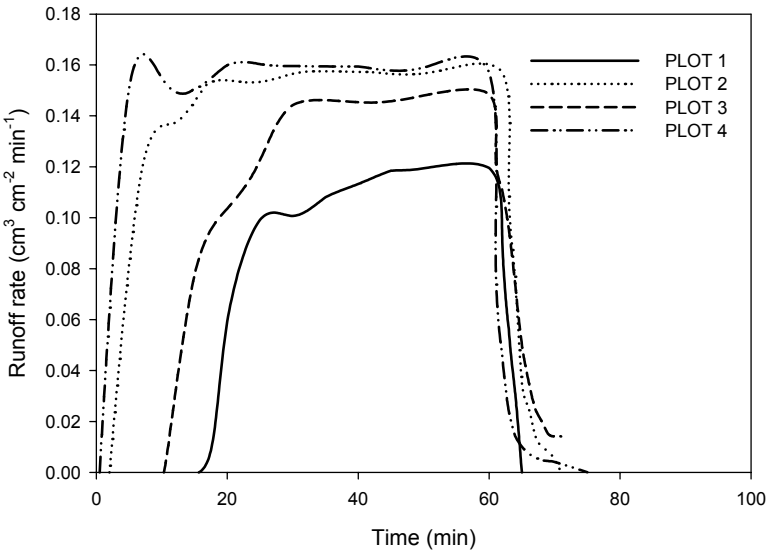
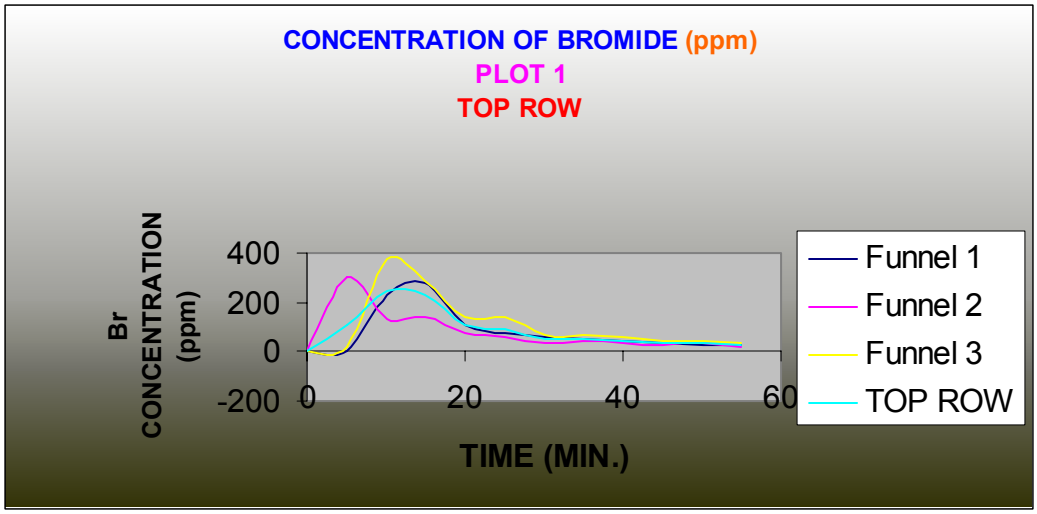
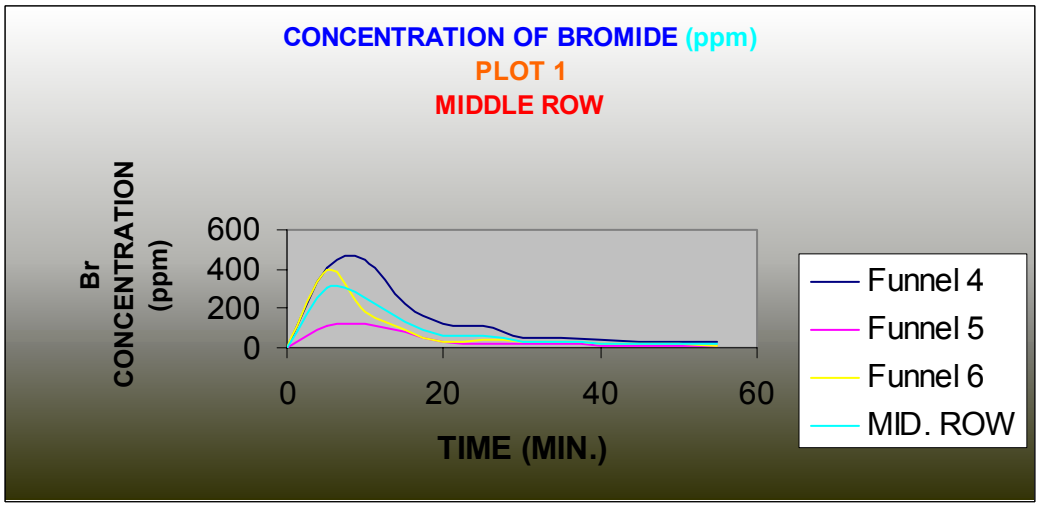


Figure 3. Runoff hydrographs for sub-plots with 5% slope in clay loam soil; plots 1&3 (vegetated) and plots 2&4 (bare).





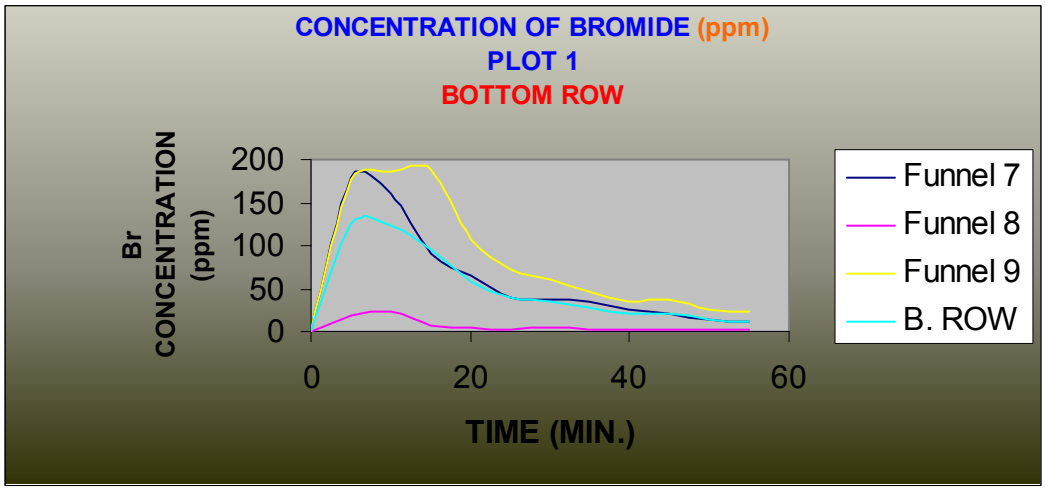


Figure 4. Bromide breakthrough curves at different distances for bare clay loam sub-plot at 20% slope (Funnels 1, 2, & 3 at 95 cm; Funnels 4,5, & 6 at 285 cm; and 7, 8, & 9 at 490 cm from the top of the ridge) and the average breakthrough curve at each distance.

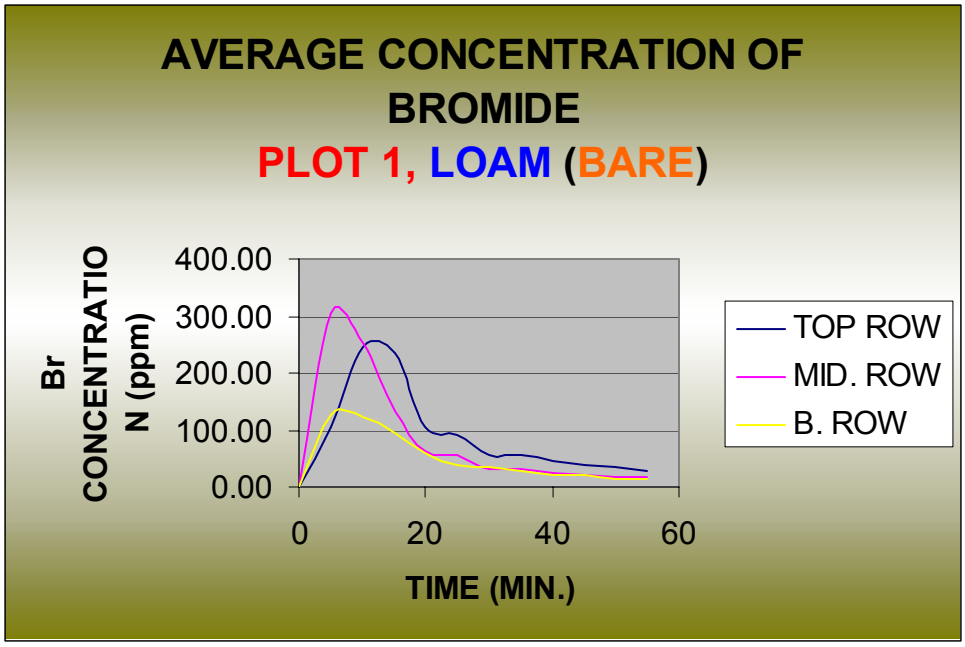
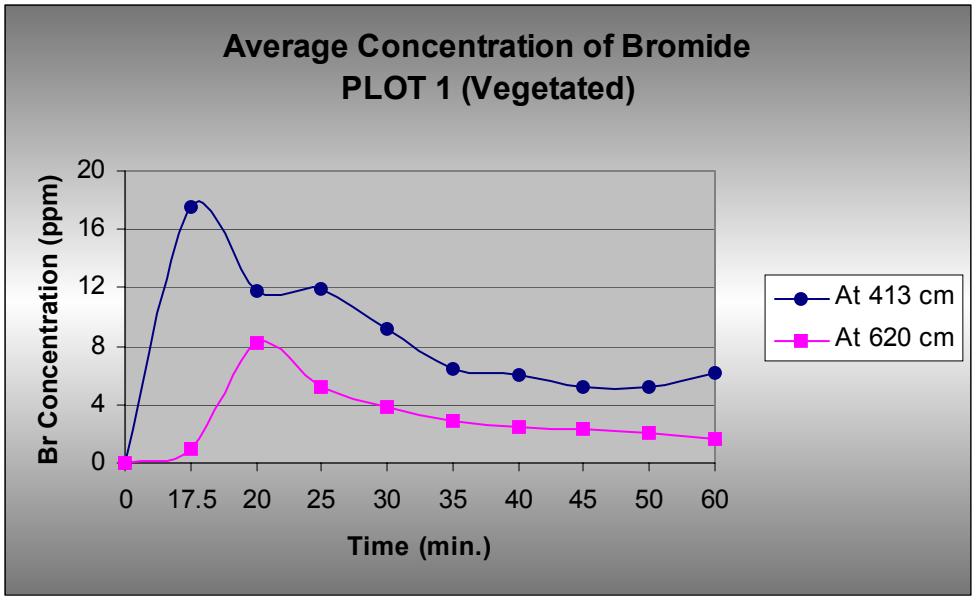


Figure 5. Average Bromide breakthrough curves at 95 cm, 285 cm, and 490 cm from the top of the ridge in bare clay loam soil sub-plot at 20% slope.



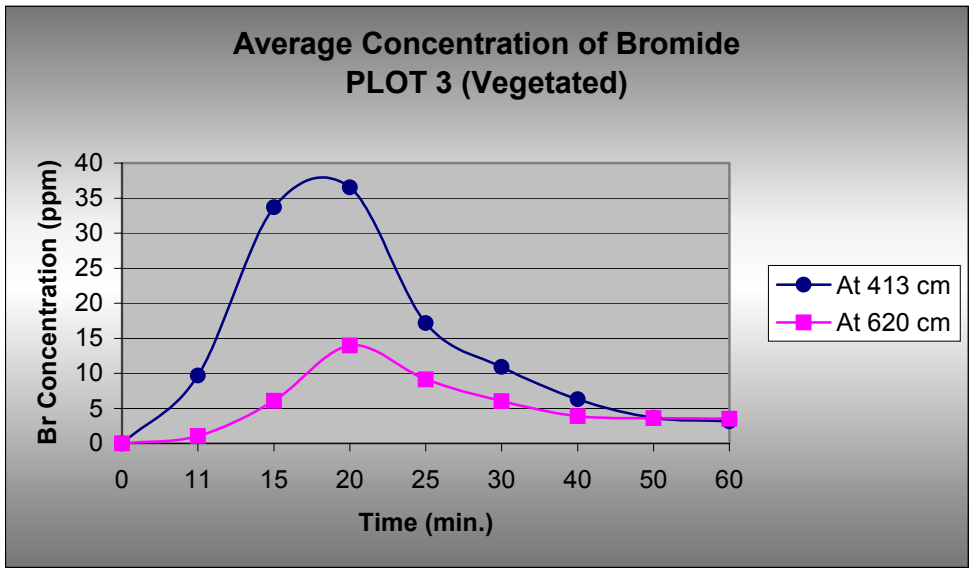
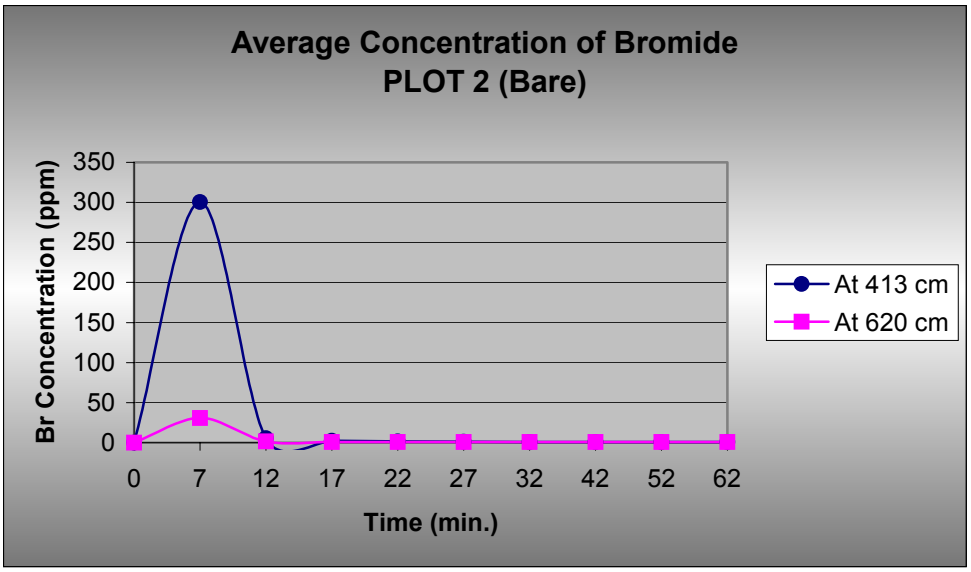


Figure 6. Average Bromide breakthrough curves at 413 and 620 cm from the edge of application area in vegetated clay loam soil for sub-plots 1 and 3 with 5% slope.



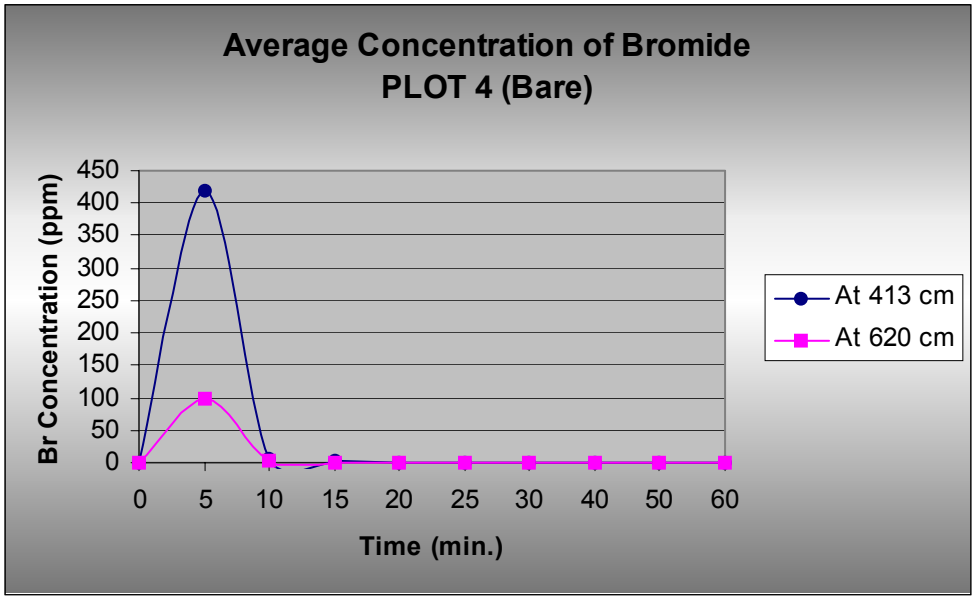


Figure 7. Average Bromide breakthrough curves at 413 and 620 cm from the edge of application area in bare clay loam soil for sub-plots 2 and 4 with 5% slope.

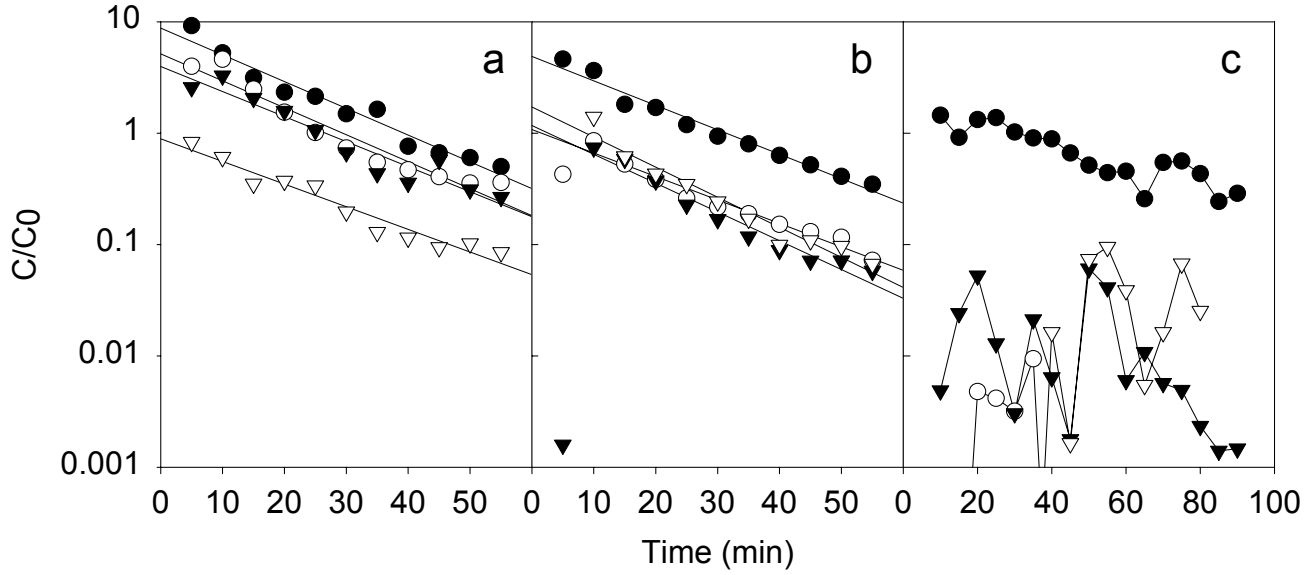


Figure 8. Relative concentrations of fecal coliforms in runoff; a - bare clay loam, b - bare sandy loam, c - vegetated clay loam; □ - 95 cm, ○ - 285 cm, ▼- 485 cm,.▽ - 600 cm from the ridge in the lysimeter with 20% slope.

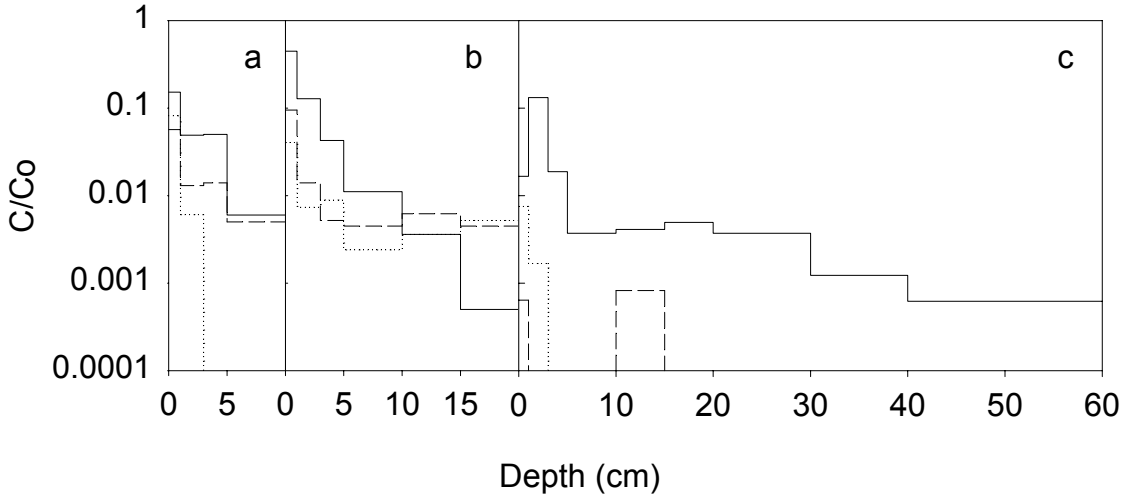


Figure 9. Relative concentrations of fecal coliforms in soil; a - vegetated clay loam, b - bare sandy loam, c - vegetated sandy loam; _____ - 95 cm, - - - - 285 cm, ……. - 485 cm from the ridge in the lysimeter with 20% slope.

PLOT 1
Bare Clay Loam
Regression of E-coli Concentration Ratios(C/C_0)
to Salmonella Concentration ratios(C/C_0)

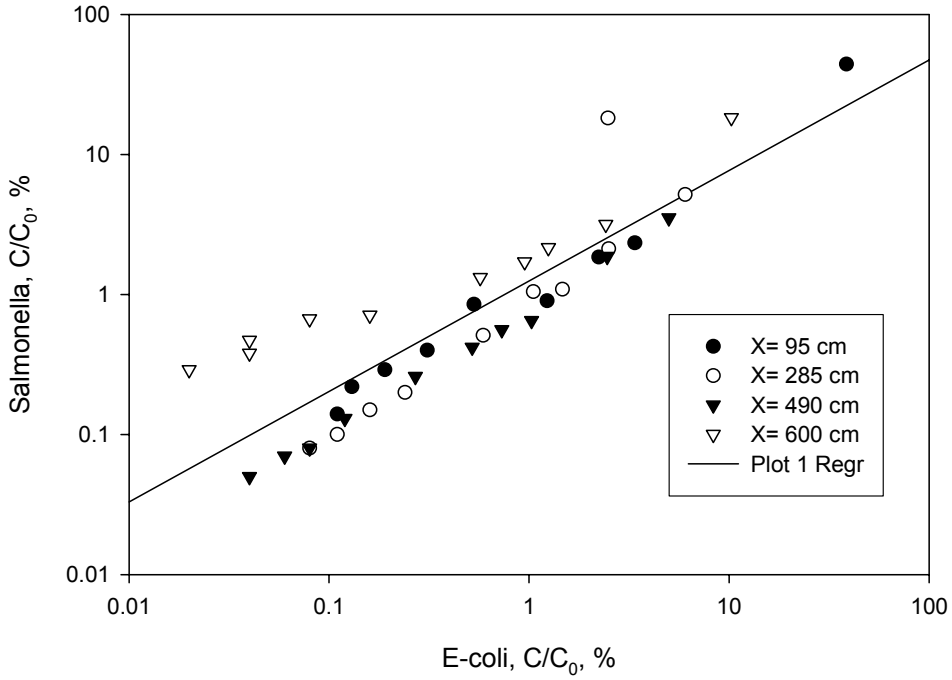


Figure 9a. Relationship between Salmonella and E.Coli for Bare Clay Loam in the lysimeter with 20% slope.

$S_{95\ cm}$=-0.2437+1.1493$E_{95\ cm}$ R^2=0.9985

$S_{285\ cm}$=0.0025=0.8548$E_{285\ cm}$ R^2=9977

$S_{490\ cm}$=0.031+0.705 $E_{490\ cm}$ R^2=0.9977

$S_{600\ cm}$=0.155+1.7402 $E_{600\ cm}$ R^2=0.9931

PLOT 3
Bare Sandy Loam
Regression Of E-coli Concentration Ratio (C/C_o)
to Salmonella Concentration Ratio (C/C_o)

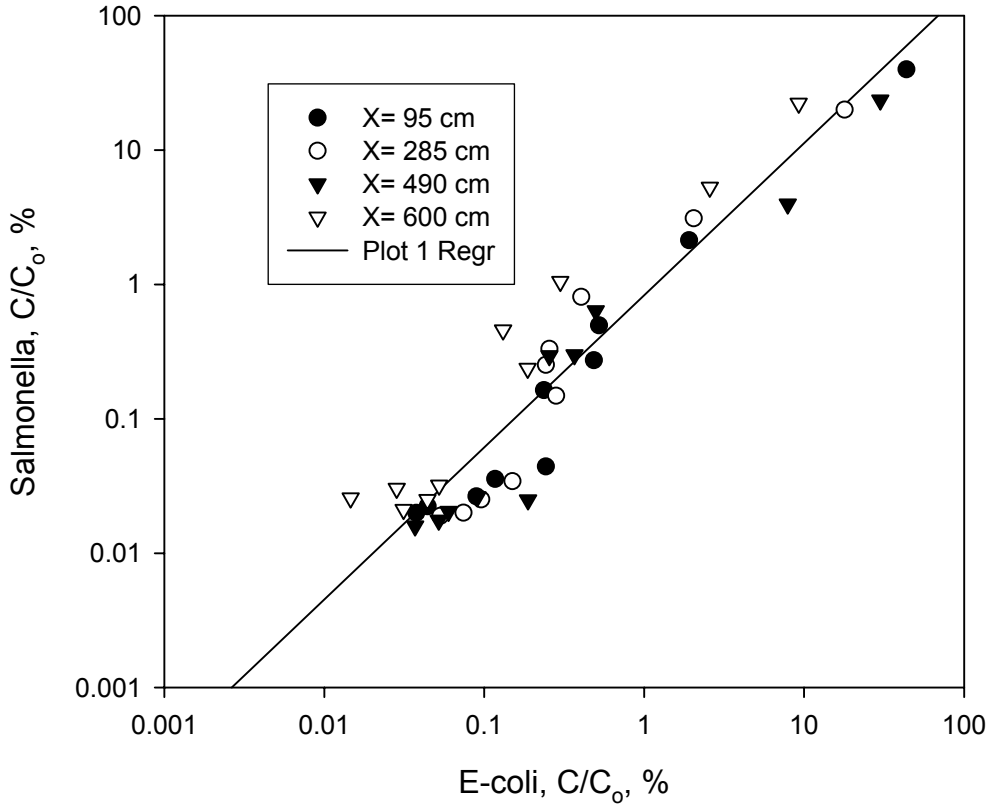


Figure 9b. Relationship between Salmonella and E.Coli in Bare Sandy Loam in the lysimeter with 20% slope.

$S_{95\ cm} = -0.01921+0.913633E_{95\ cm}$ $R^2=0.99984$

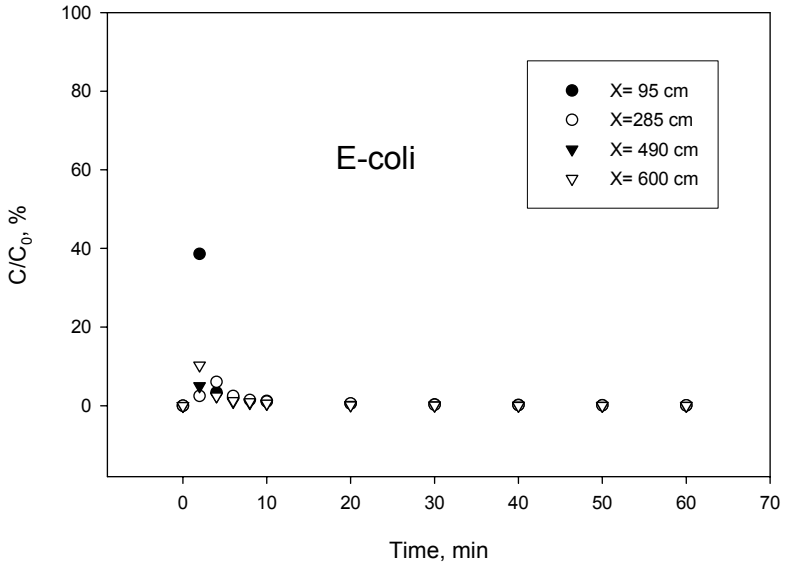
$S_{285\ cm} = 0.07245+1.112383\ E_{285\ cm}$ $R^2=0.99761$

$S_{490\ cm} = -0.16809+0.774424E490\ cm$ R2=0.99054

$S_{600\ cm} = -0.07175+2.383434E_{600\ cm}$ $R^2=0.99798$

$S_{overal} = -0.08015+1.1323827E_{overal}$ $R^2=0.8989$

PLOT 1
Bare Clay Loam
Concentration Ratios (C/C_0)
of E.coli and Salmonella



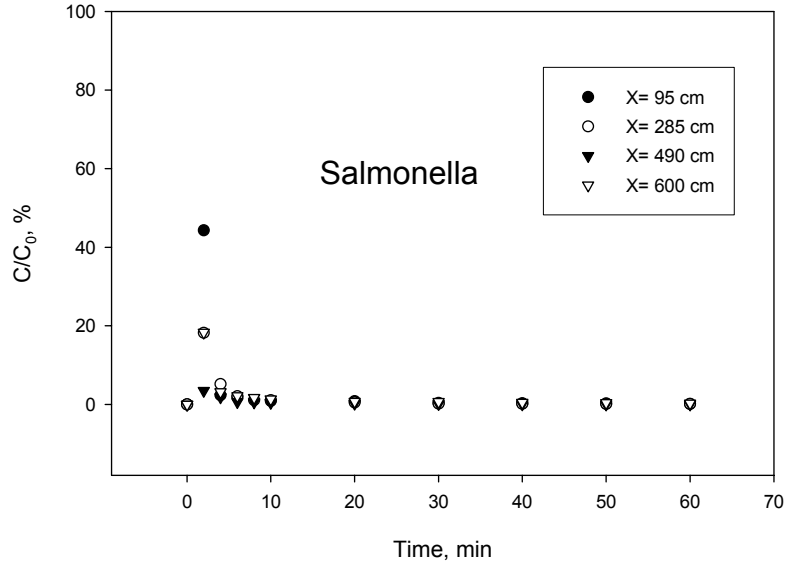
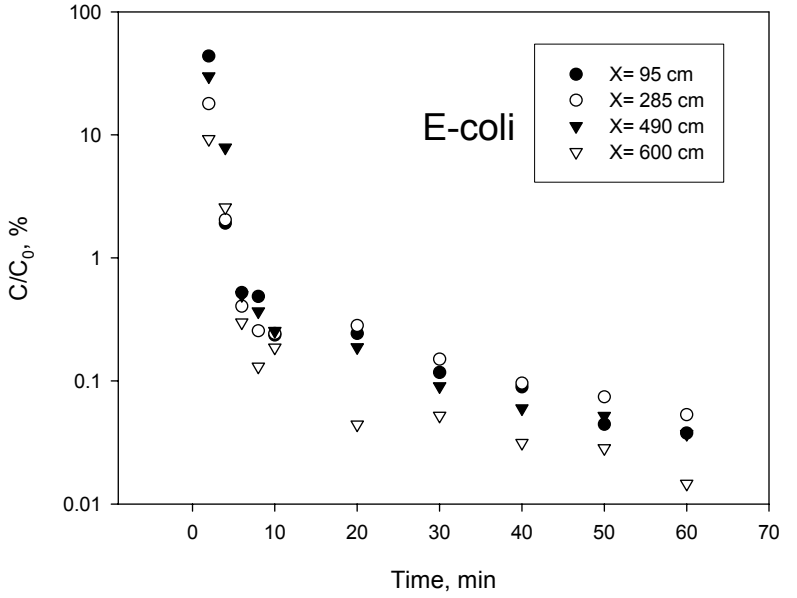


Figure 10a. Concentration ratios (C/C_0) with time for E.Coli and Salmonella in bare Clay Loam in the lysimeter with 20% slope.

PLOT 3
Bare Sandy Loam
Concentration Ratios (C/C_0)
of E.coli and Salmonella



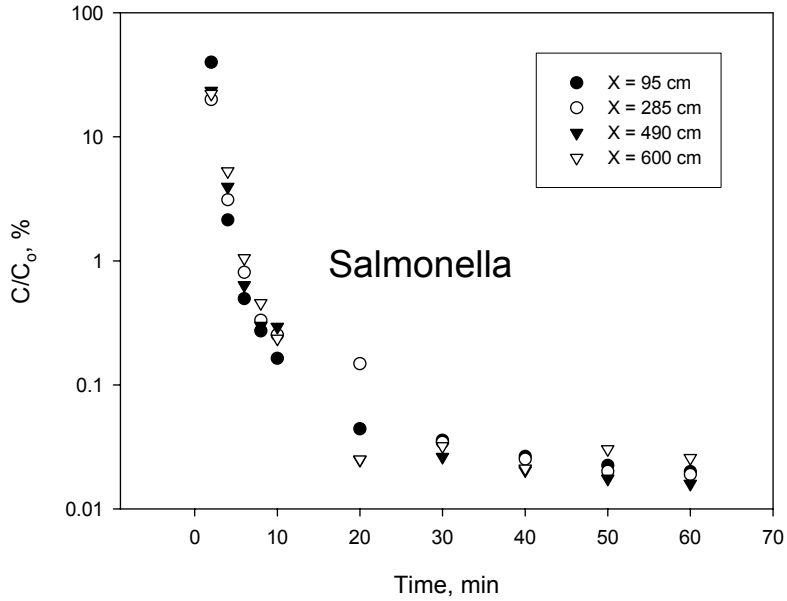
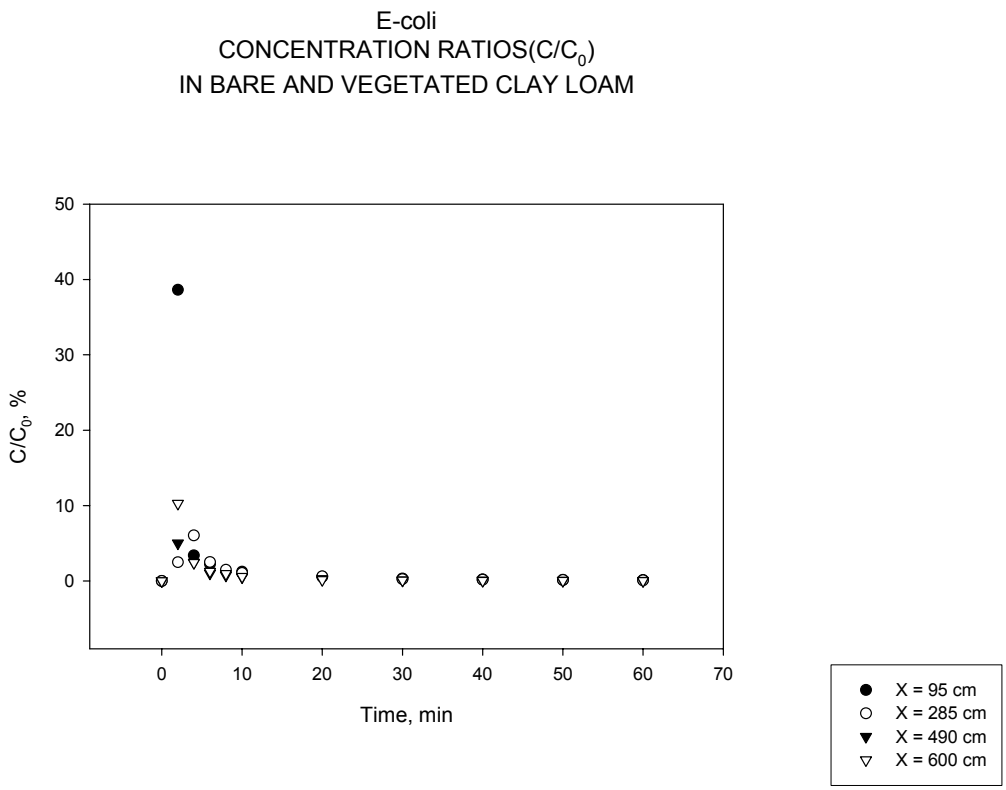
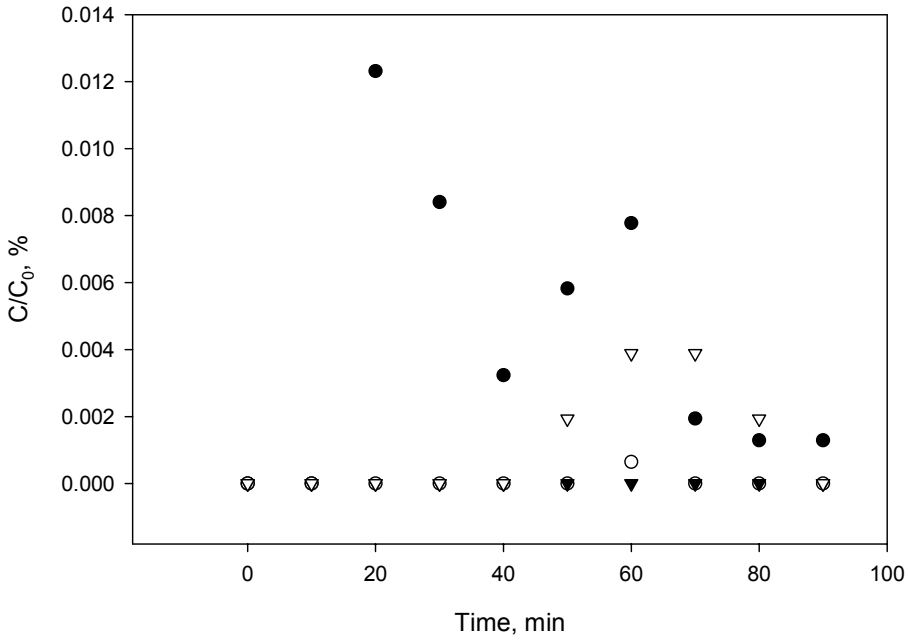


Figure 10b. Concentrations ratios (C/C_0) with time for E.Coli and Salmonella for bare Sandy Loam soil in the lysimeter with 20% slope.



(a) Bare Clay Loam



(b) Vegetated Clay Loam

Figure 11. Concentration Ratios (C/C_0) of E.Coli with time for both bare (a) and vegetated (b) Clay Loam soil in the lysimeter with 20% slope.

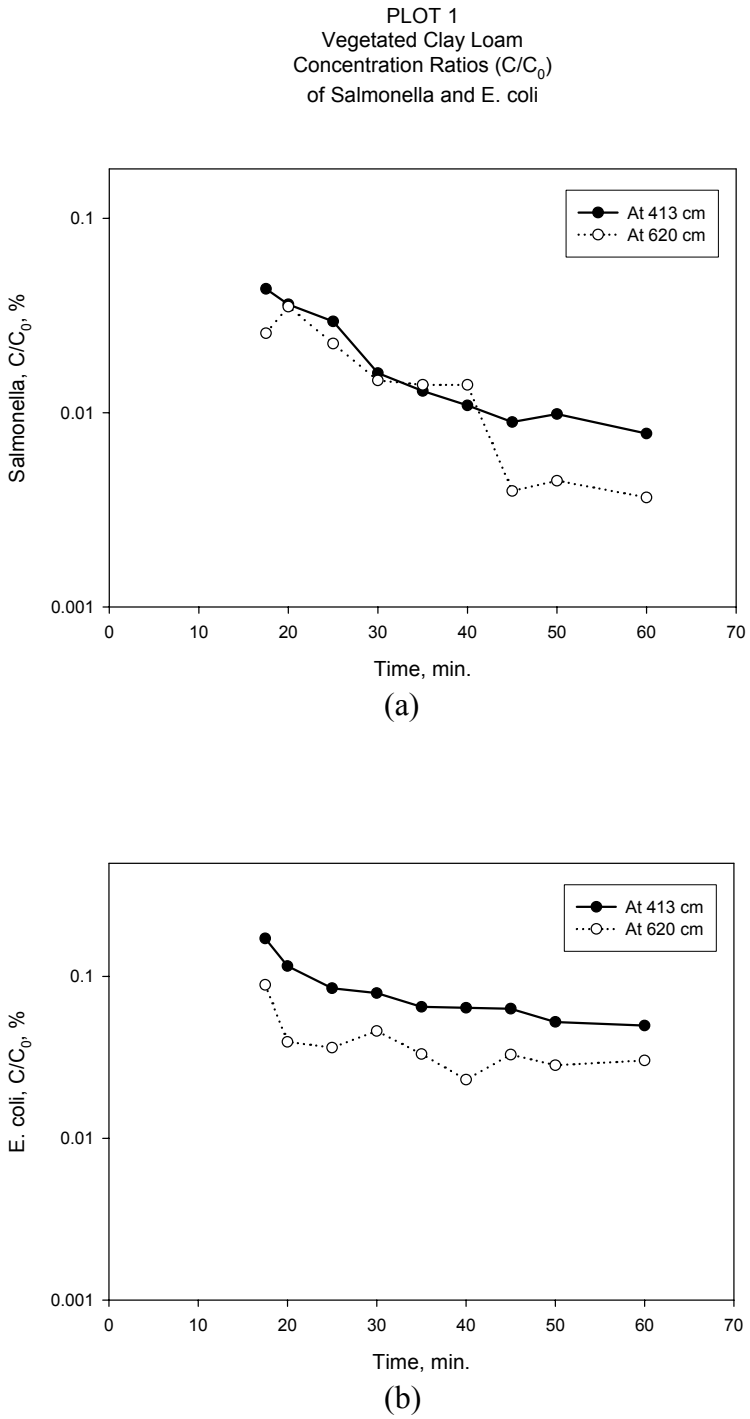


Figure 12a. Concentration Ratios (C/C_0) of Salmonella (a) and E-Coli (b) for the vegetated Clay Loam sub-plot (plot 1) with 5% slope.

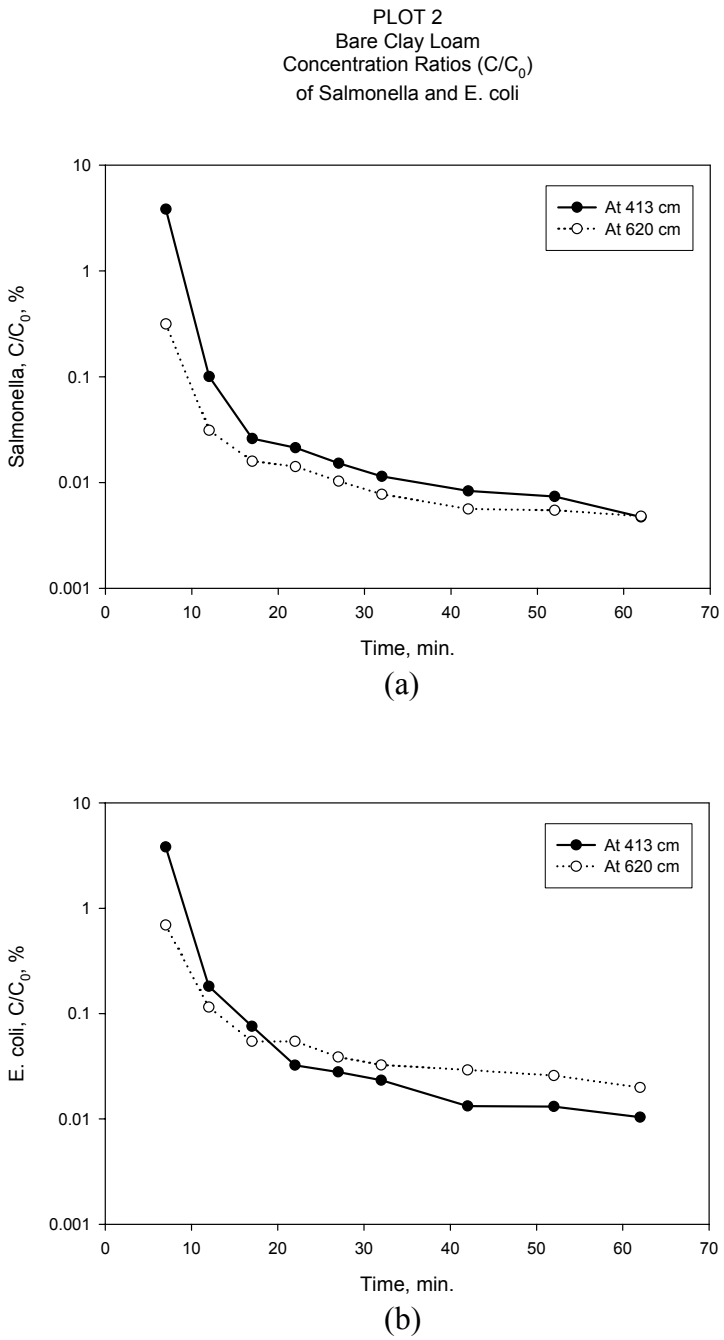


Figure 12b. Concentration Ratios (C/C_0) of Salmonella (a) and E-Coli (b) for the bare Clay Loam sub-plot (plot 2) with 5% slope.

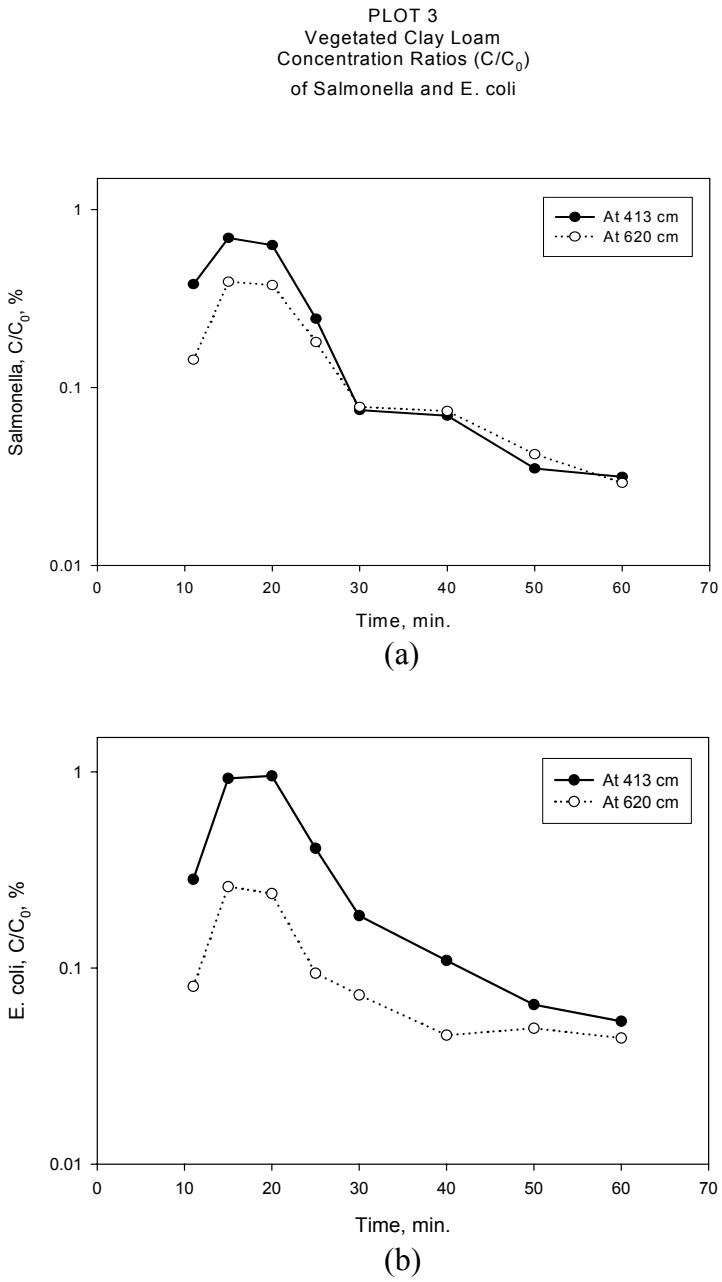


Figure 13a. Concentration Ratios (C/C_0) of Salmonella (a) and E-Coli (b) for the vegetated Clay Loam sub-plot (plot3) with 5% slope.

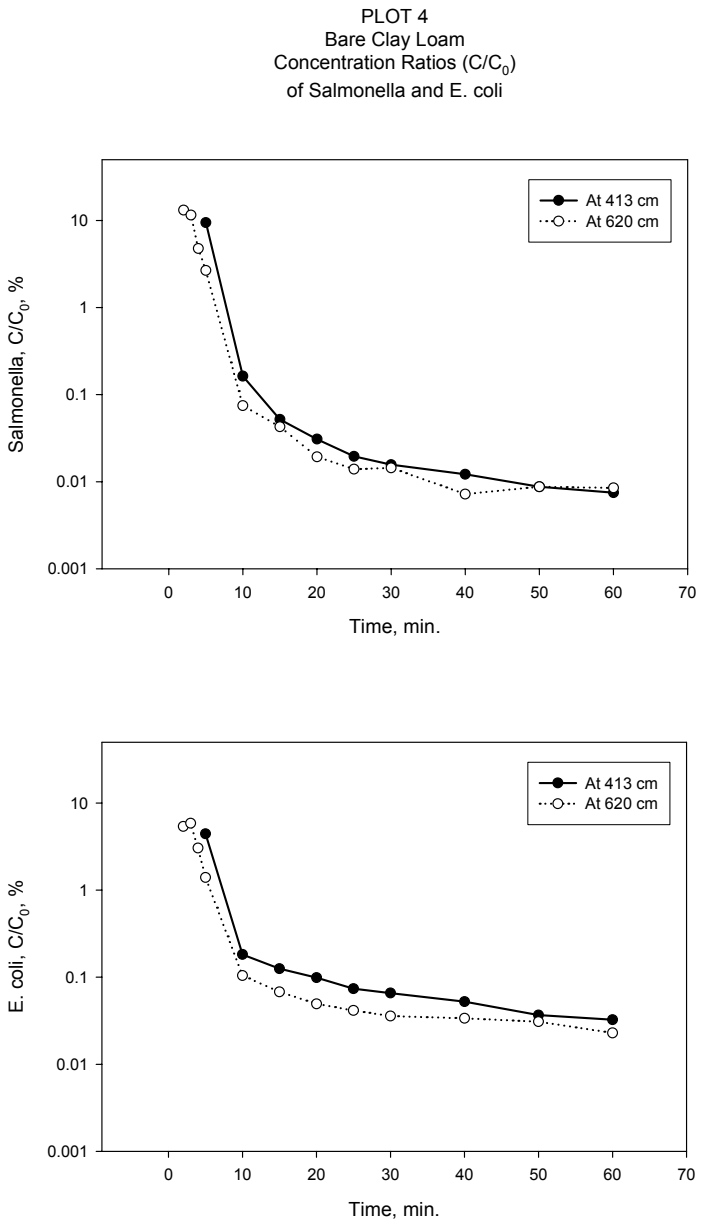


Figure 13b. Concentration Ratios (C/C_0) of Salmonella (a) and E-Coli (b) for the bare Clay Loam sub-plot (plot4) with 5% slope.