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Runoff transport of faecal coliforms and phosphorus released from manure in grass buffer conditions

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ABSTRACT

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Aims: To test the hypothesis that faecal coliform (FC) and phosphorus (P) are transported similarly in surface runoff through the vegetative filter strip after being released from land-applied manure.

Methods and Results: The Hagerstown soil was packed into boxes that were 10 cm deep, 30 cm wide and 100, 200 or 300 cm long. Grass was grown in boxes prior experiments. Same-length boxes were placed under rainfall simulator and tilted to have with either 2% or 4% slopes. Dairy manure was broadcast on the upper 30-cm section. Rainfall was simulated and runoff samples were collected and analysed for Cl, FC and total phosphorus (TP). Mass recovery, the concentration decrease rate k, and the ratio FC : TP showed that there was a consistent relationship between FC and TP in runoff.

Conclusion: The FC and TP transport through simulated vegetated buffer strips were highly correlated.

Significance and Impact of the Study: As a knowledge base on the effect of the environmental parameters on P transport in vegetated buffer strips is substantially larger than for manure-borne bacteria, the observed similarity may enhance ability to assess the efficiency of the vegetated buffer strips in retention of FC currently used as indicator organisms for manure-borne pathogens.

Keywords: faecal coliform, manure, phosphorus, runoff transport, vegetated filter strip.

INTRODUCTION

Large amounts of manure are applied to agricultural land in localized areas. In many cases, this can elevate levels of micro-organisms and phosphorus (P) in runoff (Sharpley *et al.* 1998; Johnson *et al.* 2003). Vegetative filter strips (VFS) have been widely adopted as a conservation measure to decrease edge-of-field losses of both micro-organisms and P to surface water (Sanderson *et al.* 2001; Abu-Zreig *et al.* 2003). The mechanisms of the VFS functioning related to decreasing edge-of-field losses include: (i) decrease surface water flow, resulting in loss of transport capacity which leads to deposition of sediment and adsorbed potential pollutants; (ii) enhancing infiltration of water and potential pollutants into soil; and (iii) adsorbing pollutants onto litter, vegetation and surface layer of soil, all of which can lower outflow concentrations (Crane *et al.* 1983).

There is little information on whether filter strips can effectively decrease both microbial and P transport in surface runoff. Edwards *et al.* (1996) found that manure-borne pollutants are transported mostly in aqueous phase. This suggests similarities in microbial and chemical transport. Other studies, however, have suggested that VFS removal of bacterial and chemical constituents in surface runoff from manured fields are unrelated, indicating different transport mechanisms for bacterial and chemical manure constituents (Crane *et al.* 1983; Barfield *et al.* 1998). In such cases, VFS should be designed to enhance infiltration as a well as retard particulate transport, regardless of the type of

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pollutant. Srivastava *et al.* (1996) and Edwards *et al.* (2000) showed that substantial FC and P removal may occur within the first several metres from the edge of cattle manure application area.

The objective of this work was to test the hypothesis that FC and P have similar runoff transport characteristics at short distances from the edge of the application zone.

MATERIALS AND METHODS

Soil and manure collection

A surface sample (0–10 cm depth) of the Hagerstown silt loam, widespread in the northern US soil was collected for this study from an agricultural field to which no manure or fertilizer P had been applied in the last 10 years (Table 1). The soil was air-dried and sieved (4 mm). The manure was scraped from a free-stall barn housing lactating Friesian-style dairy cows (*Bos taurus*), thoroughly mixed and stored at 4°C for 2 days prior the runoff experiments.

Runoff boxes and rainfall simulation

Soil was packed into boxes that were 10 cm deep and 30 cm wide, had back walls 2·5 cm higher than the soil surface, 5-mm diameter drainage holes in the base, and a canopied gutter to channel runoff water to collection containers (Kleinman *et al.* 2002). Boxes were 1-, 2- and 3-m long; six boxes of each length were prepared. Box bottoms were covered with the cheesecloth, soil was packed into boxes to achieve an approximate bulk density of 1·3–1·5 g cm^{-3}, and seeded with Kentucky Bluegrass (*Poa pratensis* L.) at a rate of 50 g m^{-2}. Volumetric soil moisture content was determined using a capacitance sensor (ThetaProbe; Delta-T Devices, Ltd, Cambridge, UK) at five locations within each box. Soil moisture was kept at field capacity (approximately 0·30 m^3 m^{-3}) by frequent spray irrigation for 14 weeks during grass growth. The grass was fertilized 4 and 7 weeks after planting with 40 kg N ha^{-1} as urea. Once established, the grass was clipped at 2-week intervals and prior to rainfall application to a 7·5-cm height.

Six packed boxes of the same length were placed under the rain simulator. Three of them were randomly selected to have a 2% slope and the remaining three had a 4% slope. Rainfall was applied using a simulator equipped with a TeeJet^{TM} 1/4 HH SS 14 WSQ nozzle (Spraying Systems Co., Wheaton, IL, USA) at 3 m above the soil surface. Simulated rainfall had the coefficient of uniformity >0·80 within the box placement area. Rainfall intensity of 2·66 cm h^{-1} was maintained for 1 h during the runoff experiments. Such rainfall has an approximate 1-year return frequency in south-central Pennsylvania. Water used in the rainfall simulations had total phosphorus (TP) of 0·02 mg l^{-1}, pH of 5·7, and electrical conductivity of 0·02 S m^{-1}.

Soil was first saturated using the rainfall for approximately 30 min, and allowed to drain for 30 min prior to manure application to minimize the hydrological variability related to antecedent moisture and to collect background runoff samples. To use Cl as a conservative tracer present only in liquid phase, $CaCl_2$ solution was added to the manure to obtain 2000 mg l^{-1} concentration, and manure was manually stirred for 3 min. Then manure was broadcast on the upper 30-cm section of each box at a rate of 7 kg m^{-2} (about five times higher than a typical PA agronomic rate), and rainfall was applied. Runoff samples were collected every 5 min after runoff initiation.

Table 1 Selected properties of the Hagerstown soil and dairy manure

Property	Hagerstown silt loam	Dairy manure
CEC (cmol kg^{-1})	16·3	ND
pH	6·75	6·95
Clay (%)	47	ND
Silt (%)	39	ND
Sand (%)	14	ND
Total P (mg kg^{-1})	475	6560
Total N (mg kg^{-1})	ND	39990
Solids (%)	ND	33·4
Faecal coliforms (CFU g^{-1})	ND	$2·1 \times 10^6$

ND, not determined; CEC, cation exchange capacity; CFU, colony-forming units.
All values are reported on the dry mass basis.

Soil, manure and water analysis

All soil analyses were conducted on air-dried and sieved (2 mm) samples. Soil sand, silt and clay contents were determined with the hydrometer method (Gee and Bauder 1986). A glass electrode was used to measure pH at a 1 : 1 soil : water ratio (w/w). Total soil P was determined following digestion with a semimicro-Kjeldahl procedure (Bremner and Mulvaney 1982).

Manure moisture content was determined by gravimetric analysis (35°basis). The pH of manure was measured by a glass electrode at a 5 : 1 water : material ratio (w : w). Organic C was determined by dry combustion (Nelson and Sommers 1996). Total P and N concentrations of the manure were determined by a modified semimicro-Kjeldahl procedure (Bremner and Mulvaney 1982). The procedure used a 1·133 g mixture of K_2SO_4 and $CuSO_4$ (100 : 3 weight ratio), *conc* H_2SO_4 (4 ml) and fresh manure (equivalent to 0·25 g of dry material), digested at 180°C for 60 min and 375°C for 120 min. Faecal coliform (FC) concentrations were determined by taking two 10-g manure samples, blending each of them with 90 ml of sterile distilled water, diluting the manure suspensions 100-fold with sterile distilled water, and finally plating (each in triplicate) on MacConkey Agar using an Autoplate 4000 spiral plater (50 μl) manufactured by Spiral Biotech (Bethesda, MD, USA).

Two unfiltered runoff subsamples were taken for FC analysis and were plated in <10 min (each in triplicate) on MacConkey Agar using an Autoplate 4000 spiral plater (50 μl). Total P in unfiltered runoff samples was determined no more than 7 days after the completion of the rainfall simulation with digestion with a semimicro-Kjeldahl procedure (Bremner and Mulvaney 1982). Phosphorus in neutralized digests was measured by the colorimetric method of Murphy and Riley (1962). Chloride concentration was measured with an ion-selective electrode (Orion; Thermo Electron Corporation, Woburn, MA, USA).

All plates were incubated overnight at 44·5°C. FC as colony-forming units (CFUs) were counted using a Protocol plate reader (Synoptics, Cambridge, UK). We report average counts.

The S-PLUS software (Mathsoft, Cambridge, MA, USA) was used to perform ANOVA tests, compute means, standard errors and regression slopes, and evaluate significance of differences at the 0·05 significance level with the *t*-test.

Mention of trade names or other proprietary information does not imply endorsement by the USDA.

RESULTS AND DISCUSSION

There were no statistically significant differences in runoff rates as a function of either box length or slope. Runoff rates quickly stabilized and after 10 min of rain remained constant with an average of 1·95 cm h^{-1} for all treatments. Infiltration rate averaged 0·65 cm over the 1-h rainfall. The average background concentrations of FC and TP in runoff prior the manure applications were $1{\cdot}9{\cdot}10^2$ CFU ml^{-1} and 0·261 μg ml^{-1} respectively.

The dynamics of FC, TP and Cl concentrations in runoff are shown in Fig. 1. Larger variability was observed at 0·7 m compared with 1·7 m or 2·7 m. Peak concentrations decreased as the distance increased and more dilution occurred as discussed by McDowell and Sharpley (2002).

Runoff data was analysed using three different approaches to determine if there was a consistent relationship between FC and TP in runoff. First, we computed the count of FC and mass of TP recovered in runoff as the percentage of applied amount (Table 2). Recovery percentages from individual plots varied from 26·6 to 45·8 for FC and from 22·9 to 59·7 for TP. There was a significant effect of the length on the FC recovery, which was not the case for TP. However, none of the differences among average values of the recovery percentages were significant ($P > 0{\cdot}05$), suggesting that rates of FC and TP release and transport were similar. The similarity in transport cannot be proven with the recovery data because theoretically the slow transport and fast release could cause the same recovery as the fast release and slow transport. However, the similarity of the recoveries from slopes of different lengths implies that the recoveries mostly represented bacteria release from manure so that during the experiments the same amount of rainfall fell on the manure patches, caused the same release, and released bacteria were then transported along the slope without substantial loss for surface retention and infiltration.

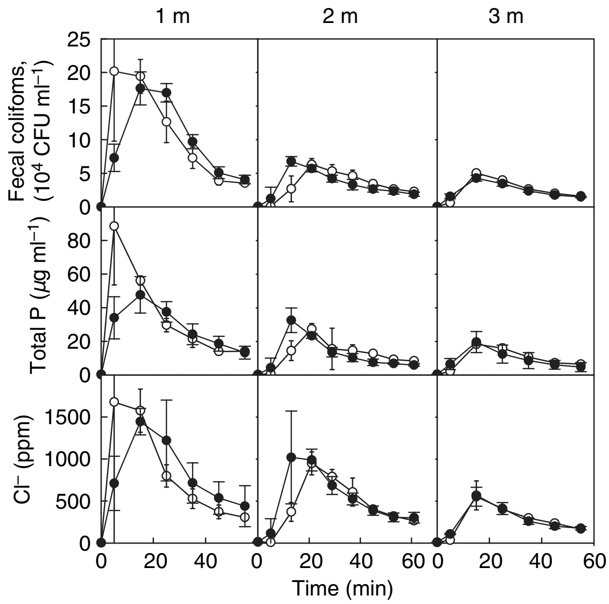

Fig. 1 Concentrations of FC, TP, and Cl in runoff averaged across three replications. Error bars show standard deviations; open symbols and closed symbols represent 2% and 4% slope, respectively; lengths of boxes are shown above graphs

Table 2 Recovery of faecal coliforms and total P in runoff

Distance (m)	Slope (%)	Recovered in runoff (% of applied in manure)	
		Faecal coliforms	Total *P*
0·7	2	38·6 ± 7·1	40·6 ± 4·5
	4	38·7 ± 1·6	34·8 ± 1·9
1·7	2	31·2 ± 3·7	39·9 ± 5·2
	4	34·0 ± 4·1	42·4 ± 12·3
2·7	2	32·7 ± 2·9	39·8 ± 6·6
	4	30·3 ± 3·3	39·1 ± 18·8

The second approach was to compare the decrease of concentrations FC and P in runoff. The tails of the breakthrough curves indicate that the decrease in concentrations with time was close to exponential (Fig. 1). The

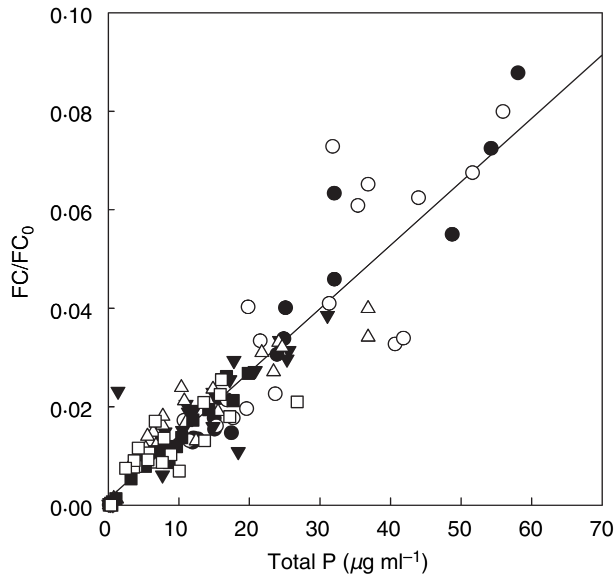

Fig. 2 Relationships between normalized faecal coliform (FC) concentration and total phosphorus (TP) concentrations in runoff; FC – the concentration of FC in runoff, FC_0 – the initial concentration of FC in manure; the linear regression line is also shown

equation $C = C_0\exp(-kt)$ was used to simulate the decrease of concentrations C of FC, P, and Cl in runoff with time t. Here C_0 is the 'maximum' concentration while k is the decrease rate constant. The decrease rate constants k were computed as the slopes of regressions in semi-logarithmic scale for each replication and for each monitored manure constituent separately. With the exception of FC at 0·7 m, there were no significant differences in slopes of the regression lines for FC, P, or Cl as a function of slope or distance. Similarly, there was no significant difference between FC, P and Cl regression line slopes across all experiments. Average values of the rate constants k for FC, P and Cl ranged from 0·01 min^{-1} to 0·02 min^{-1} and were higher than previously reported manure dissolution rates of 0·003–0·008 min^{-1} (Bradford and Schijven 2002; Shelton *et al.* 2003) for experiments in which only infiltration but not runoff was allowed to occur. The higher rate of manure dissolution with runoff, as compared with infiltration, has been previously suggested (Moore *et al.* 1988).

The third approach was to compare the ratio of FC and TP runoff concentrations after normalizing FC concentrations in runoff to initial concentrations in manure. Total P and normalized FC concentrations in runoff were related ($R^2 = 0.87$; Fig. 2). The scatter in Fig. 2 could partly be caused by the effects of time, distance and slope on the ratio $FC_{normalized}$: P. However, ANOVA did not reveal any significant effect of time, distance or slope on the ratio $FC_{normalized}$: P. This supports the assumption that, at least over short distances, TP and FC are transported similarly and their transport is not substantially affected by deposition of solid phase. Nonetheless, the average values of the ratio $FC_{normalized}$: P tended to increase as the distance and the slope increased (Fig. 2). One possible explanation is that an increase in distance and/or slope should increase the dispersion in the transport, i.e. differences in surface water velocity along different pathways along the slope. If there is a difference between manure particulates that carry FC and P, this difference can manifest itself in distributions of time intervals between particulate attachment and its release back to the flow. If those times are nonlinearly dependent on velocity, then the increase in dispersion may cause changes in the $FC_{normalized}$: P ratio. This hypothesis is also supported by the increase in standard deviations of P recoveries with distance as opposed to FC recoveries (Table 2).

Concentration of FC and Cl in runoff were related ($R^2 = 0.83$). Statistical analysis (ANOVA) demonstrated the ratio $FC_{normalized}$: Cl significantly decreased with box distance and slope. This may mean that Cl is transported solely in aqueous phase and is unaffected by particulate transport, sorption or deposition processes, which influence FC transport. Similar differences between FC and nitrate transport in VFS were reported by Fajardo *et al.* (2001).

Some of TP in runoff could be attributed to the FC cells as *Escherichia coli* contains about 3% P on dry mass basis (Neidhardt *et al.* 1990). However, P from FC can constitute only small part of the P found in manure as the dry mass of an *E. coli* cell varies mostly between $0{\cdot}1{\cdot}10^{-12}$ and $1{\cdot}10^{-12}$ g (Loferer-Krößbacher *et al.* 1998) and, with FC concentrations in manure of $2{\cdot}10^{6}$ CFU g^{-1} (Table 1), P from FC cells is <10^{-4}% of manure dry weight. We have not measured the total microbial biomass which is usually <30% of manure mass on dry mass basis (Van Kessel *et al.* 2000). Assuming that all microbial biomass is transported in runoff in same way as FCs and using an estimate of 3% TP in bacteria, we find that microbial P constitutes about 0·3% of manure dry weight which is almost half of 0·66% TP found in manure (Table 1). The correlation in TP and FC runoff concentrations may be related to large extent purely to the transport of P in bacterial cells. Other mechanisms, i.e. bacterial polyphosphate inclusion bodies, binding to external cell sites and may be salt complexes could contribute to the observed correlation.

Overall, our data demonstrate a similarity in transport of manure-borne FC and P through simulated vegetated buffer strips. This similarity was observed across relatively short distances and under saturated conditions close to saturation where infiltration is limited. Although these results may not represent FC and TP transport over longer distances or in unsaturated soils, they do indicate that it may be possible to use TP as a surrogate tracer of manure-borne FC in runoff. More work needs to be performed to verify the relationship.

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