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Transfer of *Escherichia coli* to Water from Drained and Undrained Grassland after Grazing

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ABSTRACT

The aim of this study was to determine the load of *Escherichia coli* transferred via drainage waters from drained and undrained pasture following a grazing period. Higher concentrations (ranging between 10^4 and 10^3 colony forming units [CFU] g^{-1}) of *E. coli* persisted in soil for up to 60 d beyond the point where cattle were removed from the plots, but these eventually declined in the early months of spring to concentrations less than 10^2 CFU g^{-1}. The decline reflects the combined effect of cell depletion from the soil store through both wash-out and die-off of *E. coli*. No difference ($P > 0.05$) was observed in *E. coli* loads exported from drained and undrained plots. Similarly, no difference ($P > 0.05$) was observed in *E. coli* concentrations in drainage waters of mole drain flow and overland plus subsurface interflow. Intermittent periods of elevated discharge associated with storm events mobilized *E. coli* at higher concentrations (e.g., in excess of 400 CFU mL^{-1}) than observed during low flow conditions (often <25 CFU mL^{-1}). The combination of high discharge and cell concentrations resulted in the export of *E. coli* loads from drained and undrained plots exceeding 10^6 CFU L^{-1} s^{-1}. The results highlight the potential for drained land to export *E. coli* loads comparable with those transferred from undrained pasture.

Fecal bacterial loading of surface waters can occur when runoff from agricultural land is coincident with land application of livestock wastes such as manures, slurries (liquid mix of excrement and urine produced by housed livestock), and excrement deposited by grazing animals (Aitken, 2003; Rodgers et al., 2003; Patni et al., 1984; McDonald et al., 1982). The detection of fecal indicator organisms (FIOs), such as *Escherichia coli*, in surface waters is indicative of fecal pollution and we can use these bacteria as "microbial trackers" to map potential pathogen delivery to water via surface and subsurface flows from agricultural land. The prevalence and proportions of FIOs exceed those of pathogenic strains but both are derivatives of livestock waste. Thus, using generic *E. coli* as a FIO is advantageous because concentrations of pathogenic *E. coli* may drop below limits of detection on dilution in receiving waters more readily than indicator organisms, but still be present in high enough numbers to severely debilitate vulnerable hosts on ingestion (Gerba, 1996). For example, fewer than 30 cells of *E. coli* serotype 0157 are thought to cause illness in humans (Strachan et al., 2002). The relative contribution of different transfer pathways that link the land store of fecally derived bacteria to receiving waters and provide hydrological connectivity during periods of high rainfall requires investigation at the field scale (Oliver et al., 2003, 2005). Understanding fecal bacteria persistence in soil after grazing and the subsequent transfer from land to water is important both in terms of minimizing the risk to public health and in developing and improving management strategies to achieve compliance with legislation designed to safeguard water quality such as the EU Water Framework Directive (Anonymous, 2000).

Evans and Owens (1972) assessed *E. coli* concentrations in drainage water from a 0.7-ha pasture following grazing combined with pig slurry application. However, the authors made no comparison of the different transfer pathways. Fecal bacteria concentrations have been measured in tile drain flow at the field scale (Scott et al., 1998) and in overland flow under laboratory conditions (Quinton et al., 2003) and in the field under natural conditions (e.g., Patni et al., 1985; Hunter et al., 1992). More recently, Abu-Ashour and Lee (2000) examined *E. coli* transport on sloping soils by runoff action using 10- × 10-m plots. They concluded that runoff was a dominant carrier of bacterial cells.

A series of field experiments in Scotland has detailed *E. coli* emergence in drainage water following cattle slurry applications to 600-m^2 plots and made comparisons between cell concentrations exported in both drained pathways and those carried by surface runoff (Vinten et al., 2002; Fenlon et al., 2000) and this marks an important move toward quantifying the relative contributions of different flow pathways. However, a simultaneous study of the relative contribution of different hydrological pathways exporting fecal bacteria on large plots (1-ha size) has not been reported. Furthermore, a gap in current understanding exists in terms of upscaling experiments to enable realistic routes of FIO transfer to be predicted in the field.

The aim of this study was to establish whether *E. coli* deposited via excreta from grazing steers could be transmitted to receiving waters from poorly drained pasture in undrained (near-surface flow to a 30-cm depth) or drained (artificial drainage to an 85-cm depth) conditions. A key objective was to determine *E. coli* loads transferred via different hydrological pathways and thus identify whether drained or undrained pastoral land had the potential to export more bacteria. It was hypothesized that during baseflow conditions, both drainage

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Abbreviations: CFU, colony forming units; FIO, fecal indicator organism; MTF, mole and tile drain flow; OSF, overland plus subsurface interflow.

treatments would export reduced cell concentrations (and as a result reduced loads) in contrast to storm events. Storm runoff was proposed as the main driver of FIO losses from agricultural land, during which near-surface and drained hydrological pathways were expected to export high *E. coli* loads to streams. To meet these objectives, temporal changes in *E. coli* concentrations and water discharge emerging from replicated drained and undrained 1-ha hydrologically isolated plots were monitored after a summer grazing season. *Escherichia coli* was chosen as it is the most specific microorganism used to confirm fecal contamination (Mendoza et al., 2004) and is easily enumerated.

MATERIALS AND METHODS

Experimental Site

The work reported here used the Rowden Experimental Research Platform (UK National Grid Reference [NGR]: SX 650 995) described in detail by Armstrong and Garwood (1991). The soil at the experimental site is classified as a clayey noncalcareous pelostagnogley (Avery, 1980), a Typic Haplaquept (USDA Soil Conservation Service, 1975) of the Hallsworth Series (UK Soil Survey; Harrod, 1981). The site was established in 1982 on old, unimproved pasture on slowly permeable sloping land (5–10%) (Scholefield et al., 1993). A total of eight 1-ha hillslope lysimeters were used: four plots with artificial drainage and four plots without. Details of the flow pathways can be found in Haygarth et al. (1998). Briefly, flow monitoring on the undrained plots amalgamates all overland plus subsurface interflow (OSF) to a depth of 30 cm. The OSF is collected in gravel-filled ditches installed at a 30-cm depth at the lower plot boundary. Artificial drains provide the drained plots with a secondary discharge route via mole and tile drain flow (MTF), in addition to the OSF pathway that is found in undrained plots. The OSF pathway is less important on drained plots as most water is rerouted through the mole and tile drains at 40- and 85-cm depths, respectively. Deep interceptor drains were installed on all plots at downslope boundaries with gravel backfill to the surface and similar drains were installed to divert extraneous water at upslope boundaries (Scholefield et al., 1993) thus isolating, hydrologically, each plot, except for deep seepage, which is negligible due to the impermeable clay layer at a 30-cm depth (Armstrong and Garwood, 1991). None of the plots had been grazed for more than one year before the experiments during the UK outbreak of Foot and Mouth Disease in 2001.

Sampling Procedures

All water samples were collected from October 2002 through to March 2003 in sterile polyethylene 500-mL bottles. Each of the four replicate drained and undrained plots had been grazed for 6 mo (May–October 2002) with 4 steers ha^{-1}. A two-tiered approach was used, whereby a manual systematic weekly baseline sampling program complemented a manual plus automated intensive storm runoff sampling procedure. The weekly systematic baseline sample included a series of three grab samples through the course of the day, each separated by at least 4 h. This enabled an observation of diurnal fluctuations in bacteria concentrations in addition to longer-term changes in cell numbers through several months. For each manual sample a flow measurement was taken, using a stop-watch and measuring cylinder, so that FIO flux could be calculated. Five of the sampling days had rainfall in excess of 8 mm d^{-1}. Soil cores to a depth of 7 cm were randomly taken to relate *E. coli* numbers emerging in drainage waters to those determined in the upper soil matrix. Each 1-ha plot was subdivided into a 6 × 6 grid and 12 cores bulked from each subsector sampled. An automatic weather station provided rainfall data. Detailed instrumentation was installed for one replicate of each drainage treatment and included a flow rate recording device (Talman, 1983) and automated water sampling units (1011 Epic; Buhler Montec, Dusseldorf Germany). Operation of all devices was controlled using a CR10 datalogger and wiring panel (Campbell Scientific, Logan, UT). Automated sampling was initiated whereby samples were collected with a designated sampling interval of 1 h to allow characterization of flow hydrograph signatures during storm events (see also Roser et al., 2002). Turbidity, pH, and total P were measured in drainage waters to test for relationships between *E. coli* and environmental variables. Turbidity readings for water samples were obtained via an LP 2000 bench turbidity meter (Hanna Instruments, Woonsocket, RI). Water pH was recorded using a pH microprobe with a silver/silver chloride reference electrode and measurements read from a Model 3320 pH meter (Jenway, Dunmow, UK). Total phosphorus was measured using a persulfate digestion method and the absorbance of the sample measured at 880 nm following addition of an acid–antimony–molybdate reagent.

Microbiological Analysis

All samples were dealt with within 4 h of collection. Fresh soil samples were crumbled and 10 g was added to 90 mL of sterile water before mixing for 40 min on a rotary agitator. The resulting soil suspensions, and water samples, were serially diluted in sterile water then spread-plated onto MacConkey agar and incubated at 37°C for 24 h. Those colonies characteristic of *E. coli* growing on MacConkey agar were enumerated, and seven random isolates were used to confirm their identity using both MicroPlate test panels (Biolog, Hayward, CA) and API 20E biochemical identification kits (bioMerieux Vitek, Hazelton, MO). Both these procedures rely on the biochemical profiles exhibited by the test isolates for confirmation of their identity through database comparison.

Statistical Analysis

Microbial data was log-transformed and then all statistical analysis was performed on these values. Comparison of soil data was made using two-tailed *t* test and analysis of variance (ANOVA). Pearson's product-moment correlation coefficients were derived to assess the association between biological and environmental variables such as turbidity, pH, and total P. The hydrological patterns of *E. coli* emergence in the drainage treatments were compared using ANOVA. When assessing diurnal fluctuations (i.e., when sampling more than once during the day of sampling), a split plot design was used. The use of a split plot in time (repeated measures) ANOVA allowed for comparison of diurnal variations in addition to differences between drainage treatments. To account for the correlation between repeated measures where, theoretically, the correlation should be zero, a correctional factor (Greenhouse–Geisser epsilon) was derived to scale down the degrees of freedom. A split plot type ANOVA using residual maximum likelihood (REML) was required in cases where there were missing data points. In all cases, Genstat 7.1 for Windows (VSN International, 2003) was used as the statistical package.

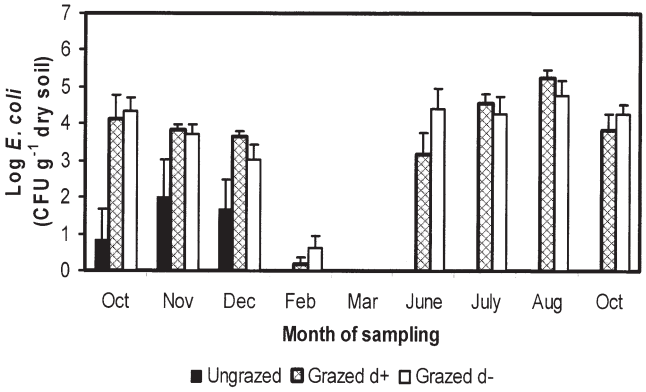

Fig. 1. *Escherichia coli* numbers in ungrazed and grazed (drained, d+; undrained, d−) plots, October 2002 to October 2003. Error bars represent 1 SE of logarithmic means. CFU, colony forming units.

RESULTS

Escherichia coli in Soil

The soil data in Fig. 1 suggest that introduced *E. coli* is able to persist in the soil for several months following the removal of cattle. However, there is a steady decline in cell numbers over time. No significant difference ($P > 0.05$) was observed in *E. coli* decline for drained and undrained treatments. By February, *E. coli* numbers were below the limit of detection (5×10^2 CFU g^{-1} dry soil). The results shown in Fig. 1 suggest that the *E. coli* detected in soil was sourced from grazing cattle because it was not detected in high numbers in the ungrazed control plot throughout the experiment in contrast to the grazed plots where *E. coli* concentrations were significantly higher ($P \leq 0.05$).

Drainage Results

Table 1 summarizes the relationships between biological and environmental variables for two periods: October to December 2002 and October 2002 to March 2003. Two periods are shown to indicate the effect of time since removal of the *E. coli* reservoir on the strength of the relationships shown. The results for the OSF pathway on the drained plot are not included in correlation analysis because flow in this pathway occurred on a few occasions only, hence data points were limited.

For October to December 2002, Table 1 shows the strength and significance of associations between bacteria concentrations and water quality variables within the undrained OSF pathway and MTF pathway, respectively. A strong relationship is shown between *E. coli* and total MacConkey bacterial colonies and *E. coli* and flow. However, the correlation coefficient of 0.739 between *E. coli* and flow for the MTF pathway ($P \leq 0.001$) is stronger than the association between these two variables within the undrained OSF pathway ($r = 0.568$, $P \leq 0.001$). There is a significant ($P \leq 0.001$) positive correlation between turbidity and *E. coli* (OSF: $r = 0.615$; MTF: $r = 0.674$). A strong relationship exists between total MacConkey colonies and both flow and total P.

For October 2002 to March 2003, examination of the relationships between biological and environmental variables through to March 2003 indicated that the strength of some of the relationships declined with time since

Table 1. Pearson's product-moment coefficients for correlation analysis between biological and environmental properties.

	E. coli	Flow	Total colonies	Turbidity	pH	Total P
			October 2002–December 2002, composite undrained pathway			
E. coli	–					
Flow	**0.568*** **	–				
Total colonies	**0.715*** **	**0.879*** **	–			
Turbidity	**0.615*** **	**0.875*** **	**0.881*** **	–		
pH	**NS**	**NS**	**NS**	**NS**	–	
Total P	**0.584*** **	**0.816*** **	**0.847*** **	**0.878*** **	**NS**	–
			October 2002–December 2002, mole drain pathway			
E. coli	–					
Flow	**0.739*** **	–				
Total colonies	**0.900*** **	**0.858*** **	–			
Turbidity	**0.674*** **	**0.872*** **	**0.807*** **	–		
pH	**NS**	**NS**	**NS**	**NS**	–	
Total P	**0.653*** **	**0.870*** **	**0.843*** **	**0.863*** **	**0.311** **	–
			October 2002–March 2003, composite undrained pathway			
E. coli	–					
Flow	**0.443*** **	–				
Total colonies	**0.658*** **	**0.831*** **	–			
Turbidity	**0.295***	**0.674*** **	**0.589*** **	–		
pH	**NS**	**NS**	**NS**	**NS**	–	
Total P	**0.516*** **	**0.821*** **	**0.840*** **	**0.853*** **	**NS**	–
			October 2002–March 2003, mole drain pathway			
E. coli	–					
Flow	**0.605*** **	–				
Total colonies	**0.815*** **	**0.842*** **	–			
Turbidity	**NS**	**0.614*** **	**0.631*** **	–		
pH	**NS**	**−0.312*** **	**−0.302*** **	**−0.258** **	–	
Total P	**0.636*** **	**0.863*** **	**0.861*** **	**0.842*** **	**−0.357*** **	–

*** Significant at the 0.05 probability level.**
**** Significant at the 0.01 probability level.**
***** Significant at the 0.001 probability level.**

the removal of cattle. Within both drainage treatments, correlation coefficients between *E. coli* and all other variables declined. This is most noticeable with the association between *E. coli* and turbidity, where a nonsignificant ($P > 0.05$) relationship is now observed in the MTF pathway. The association between total MacConkey colonies and turbidity within both transfer pathways also declines in strength, though remaining at the same level of statistical significance.

Rainfall and Flow Relationships

Instantaneous loads (discharge × cell concentration) were calculated for the three hydrological pathways. The load exported from drained plots was determined from sum of the load exported via both the OSF and MTF pathway whereas the load transferred from undrained plots was calculated for a single export pathway (OSF). The relationship between daily rainfall and *E. coli* loads exported from both drained and undrained plots is shown in Fig. 2. *Escherichia coli* loads in excess of 10^4 CFU mL^{-1} s^{-1} were frequent in the first six weeks after the removal of cattle. The greatest fluxes of *E. coli* from drained and undrained pasture were coincident with high rainfall. No significant differences ($P > 0.05$) in *E. coli* loads were identified between drainage treatments. The first storm event sampled on Day 24 exported average instantaneous loads of 1.25×10^6 and 1.05×10^6 CFU L^{-1} s^{-1} from drained and undrained plots, respectively. Instantaneous *E. coli* loads transferred during the second storm event (Day 38) averaged 1.15×10^6 CFU L^{-1} s^{-1} for drained and 5.51×10^5 CFU L^{-1} s^{-1} for undrained plots. The undrained OSF pathway exported higher concentrations of *E. coli* than the MTF pathway on the majority of sample dates, though the differences in concentrations were also nonsignificant ($p > 0.05$). Concentrations of *E. coli* in drainage discharge had declined by the end of December (see Fig. 3). However, fluxes of 1.2×10^4 and 5.6×10^3 CFU L^{-1} s^{-1} were recorded as late as Day 102 for undrained and drained plots, respectively.

The storm event on Day 24 increased the average instantaneous *E. coli* load exported from drained and undrained plots by more than 100- and 40-fold, respectively, relative to baseflow loads 24 h previous. The second storm event on Day 38, relative to the baseline sample collected the previous day, saw *E. coli* loads exported from drained and undrained plots increase by a factor in excess of 6×10^5 and 5×10^4, respectively. Diurnal fluctuations in the loads of *E. coli* exported in hydrological pathways correlated with fluctuations in discharge. For example, on Day 24, a significant increase ($P \leq 0.001$) in the diurnal load of *E. coli* corresponded with a significant increase ($P \leq 0.001$) in flow. On Day 38 a diurnal decline in cell load ($P \leq 0.001$) was accompanied by a decline in discharge ($P \leq 0.001$).

Time-Series Data

Figure 3A (MTF pathway) and Fig. 3B (undrained OSF pathway) show the concentration of *E. coli* in discharge from the continuously flowing pathways over time. Flow and *E. coli* concentration are positively correlated (Table 1). There is a marked decline in cell numbers with time for both near-surface and drained pathways. A storm event on 28 Feb. 2003 generated discharge of 2 and 2.7 L s^{-1} for the drained and undrained plots, respectively; the associated *E. coli* concentrations were 50 CFU mL^{-1} via MTF and <25 CFU mL^{-1} via undrained OSF. For the drained plot, it is interesting that samples collected at the beginning of December 2002 (Fig. 3A, Point X) experienced flow rates of approximately half that recorded at the end of February 2003, and yet the cell concentration exported was nearly five times higher at 236 CFU mL^{-1}, in December. Similarly, for the undrained OSF pathway, the December 2002 flow (Fig. 3B, Point X) was nearly a third of that at the end of February 2003, yet *E. coli* concentrations of 150 CFU mL^{-1} were recorded.

Figure 4 presents time-series data for a single storm event (27 Nov. 2002) for *E. coli* concentration, turbidity, and flow within MTF. A sample resolution of 1 h was used during the hydrograph to evaluate the pattern of *E. coli* emergence. The peak in cell numbers was observed approximately 2 h after peak discharge. A peak in turbidity is shown before both the discharge and microbial peaks.

DISCUSSION

The transfer of fecal bacteria to surface waters occurs as a function of both organism survival in both soil and drainage water, and transfer via different hydrological pathways. The decline in abundance of *E. coli* in soil, shown in Fig. 1, was anticipated because the surface source of cells was no longer replenished by supplies from cattle after their removal in October 2002. The ability of *E. coli* to persist for periods of several months highlights the potential for contamination of the surrounding environment and has been noted previously by Evans and Owens (1972) who concluded that *E. coli* isolated from drain discharge survived in or on pasture for at least 4 mo. In this study we found that drainage status had no impact on the decline of *E. coli* numbers in soil. However, the data are not a true reflection of die-off under different treatment conditions, but are instead the resultant *E. coli* population following water flux through and over the soil. It is possible that high discharges, at this scale, are the dominant factor dictating the observed decline in cell numbers in soil. The research reported here focused on cell concentrations in the upper 7 cm of soil, but cells may have also become trapped in soil pores through processes of straining, attachment, and sedimentation (Abu-Ashour et al., 1994) within deeper sections of the profile, been caught up in blocked pores (Kim and Corapcioglu, 2002) below 7 cm depth, or may have entered a viable but nonculturable (VBNC) state. The low concentrations of *E. coli* detected in October, November, and December within ungrazed plots may represent *E. coli* indigenous to the soil, or possibly be derived from wild animal feces (Wasteson et al., 1999) or be a function of surface runoff

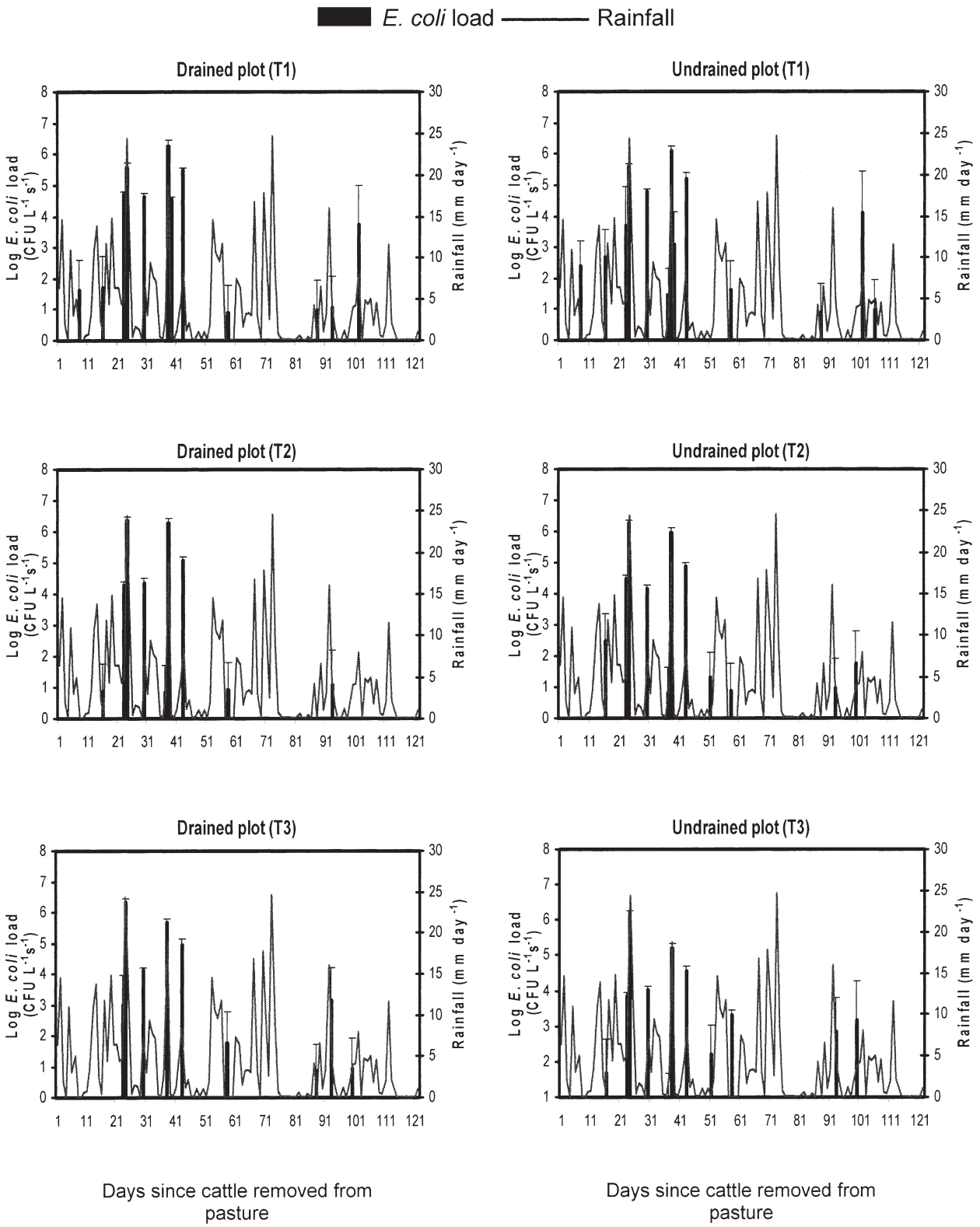

Fig. 2. Instantaneous *E. coli* loads exported from drained and undrained plots in relation to daily rainfall. The *x* axis represents day of sampling since cattle removed. The terms T1, T2, and T3 depict short-term changes on day of sampling. Error bars represent 1 SE. CFU, colony forming units.

from adjacent fields during a period associated with high rainfall (e.g., Abu-Ashour and Lee, 2000).

The lack of significant differences in *E. coli* loads exported from drained and undrained pasture suggests fecal bacteria, deposited on grasslands by grazing cattle, may be transferred via different pathways under varying

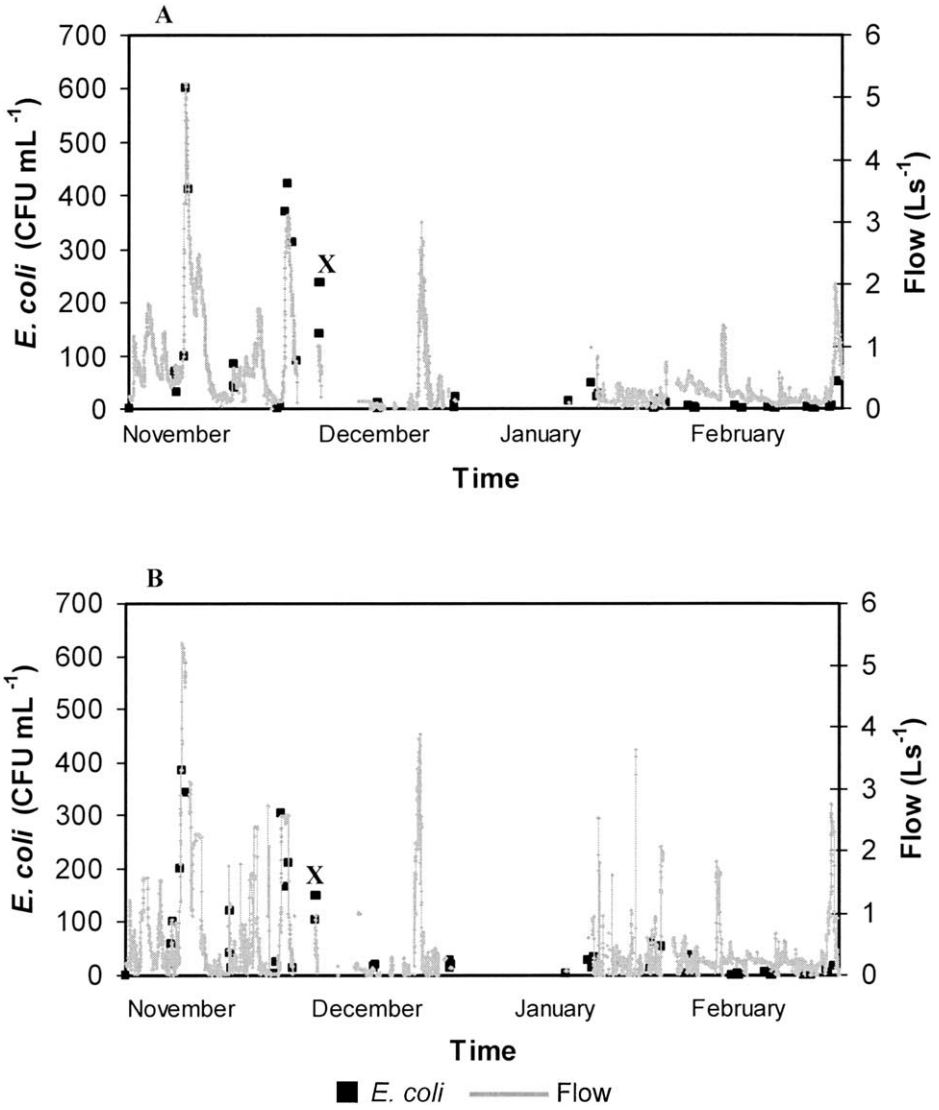

Fig. 3. Time series data illustrating *E. coli* concentration emergence with flow for (A) mole and tile drain pathway and (B) undrained composite pathway. CFU, colony forming units.

drainage conditions. For example, the installation of mole and tile drains, primarily to lower the soil water table, may be seen as advantageous in limiting the load of potential pollutants transferred rapidly via surface runoff or near-surface flow pathways. However, the pattern of *E. coli* concentrations and loads from drained and undrained plots demonstrates that cells may transfer through the soil profile, perhaps via fissures and preferential pathways, to reach the mole and tile drains. So although drainage may potentially reduce surface runoff and minimize "wash-off" of surface dwelling cells it may also reroute fecal bacteria through subsurface pathways. As noted by Smith et al. (1985), sufficient input of rainfall to a soil may initiate water flow in larger pores and create the potential for suspended bacteria to transfer rapidly through the profile of well structured soils. Other studies have observed the rapid transfer of bacteria via preferential flow pathways at the laboratory scale (e.g., Aislabie et al., 2001; Abu Ashour et al., 1998); this pathway may be important in carrying cells into the tile drain pathway of this study. Hunter et al. (1999) comment that better drained land is nonconducive for fecal bacterial survival and that transport opportunities are reduced for these bacteria. However, the present data suggest that under given rainfall conditions, *E. coli* loads comparable with those exported from poorly drained fields can be transferred from well-drained plots and emerge via subsurface routes. Furthermore, the data presented in Fig. 2 and 3 suggest that subsurface flow is a key driver of *E. coli* transfer from soil to water. These findings are similar to those of Vinten et al. (2002) who recorded high *E. coli* concentrations in drainage water following slurry application in association with periods of high rainfall and drain flow.

The large percentage increase in *E. coli* loads detected in the high flow conditions, linked to high rainfall, is evidence that discharge through pathways dictates *E. coli* emergence. A possible explanation is that increased water flux at the soil surface is required to release cells from the source material and drive their movement from

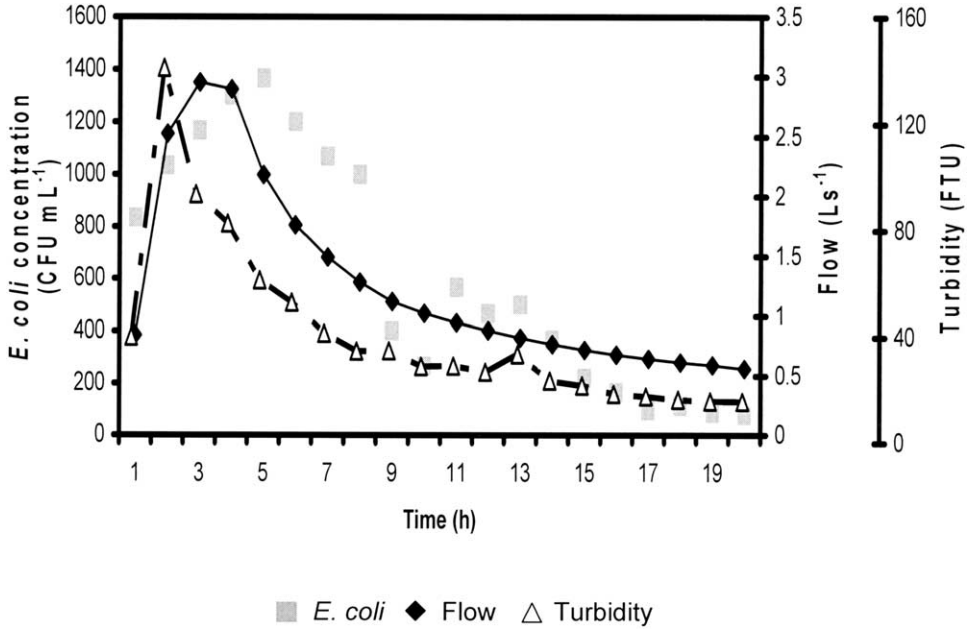

Fig. 4. Storm hydrograph (27 November) for mole drain pathway with associated *E. coli* concentrations and turbidity values at 1-h sampling resolution. CFU, colony forming units.

land to water. Low rainfall (<5 mm d^{-1}), as seen in Fig. 2, is insufficient to transfer *E. coli* loads at such high magnitudes as observed during storm events. High loads were exported from undrained plots 102 d after the removal of cattle despite the relatively low cell concentrations detected in drainage water when compared with the two storm events in November. However, the recorded discharge of water was sufficiently high to transfer an *E. coli* load comparable with that exported within 30 d of the cattle being removed from pasture. This again highlights the importance of flow volume in the export of high cell loads even after previous depletion of *E. coli* in soil through washout and cell death.

Relationships between *E. coli* and turbidity in water have been cited in the literature within river flow under flood conditions (Nagels et al., 2002) where positive associations between the two variables were noted, and within leachates (Gagliardi and Karns, 2000) where a negative correlation was observed. This field study recorded a positive correlation between *E. coli* and turbidity. This apparent relationship suggests a potential association of *E. coli* with soil colloids and particles that are dispersed during energetic flows, though the proportions of cells that are freely suspended or attached is unknown. This association with *E. coli* and flow suggests that some cells may behave similar to particulate contaminants such as colloidal P and sediment (Heathwaite et al., 2005). Tyrell and Quinton (2003) acknowledged that many authors assume that microbes behave like soil particles. Thus we can conclude that *E. coli* is physically mobilized by water, and not diluted by the increased discharge volume in a similar manner to nitrate. When calculating correlation coefficients for the data extending through to March it becomes clear that there is a decline of *E. coli* in the plots through wash out and cell death. This suggests that *E. coli* in the experiment is derived from the excrement deposited onto the soil during the grazing season of the same year and is not a legacy of the previous year's grazing.

During the storm event (Fig. 4), the peak in turbidity breakthrough occurs before *E. coli*, and in turn this occurs before flow, suggesting that soil particles and colloids are mobilized before bacterial cells. This may be because soil lines the transfer pathways through the soil profile, and it is possible that soil particulates are removed much more rapidly than surface dwelling cells located within the microhabitat of excrement and once removed, they free the pathways for the movement of cells. It is therefore proposed that cells lag behind flow because they need to be first mobilized from their protected location within the waste source at the soil surface. Vinten et al. (2002) note that *E. coli* transport to drains is likely to be more prevalent when the waste material (in their case slurry) remains at the soil surface rather than being transferred down into the soil profile during light rain. This complements the findings of our study, whereby fecal deposits remained at the soil surface during the post-grazing period and provided a source of high *E. coli* flux through drains during storm events.

CONCLUSIONS

Rainfall events, by triggering surface and subsurface runoff from grassland soils, may significantly impact the bacteriological quality of drainage waters following a typical grazing season in the UK. This is largely because *E. coli* is able to persist in soil at high concentrations extending into autumn and months typically associated with rainfall. Artificial drainage, which can reduce overland flow and potentially minimize wash-off of livestock excreta–derived contaminants from the soil surface, has

the potential to enhance *E. coli* export via subsurface routes during periods of high discharge. Thus, in terms of minimizing the risk of *E. coli* transfer from pasture to receiving waters, the installation of drainage does not necessarily offer improved protection from fecal bacteria inputs. Determining those areas of grazed pasture considered to be vulnerable to bacterial transfer should prove effective in reducing coliform loading of receiving waters. For example, the removal of cattle from steep slopes and areas prone to overland flow may limit rapid bacterial transfer during storm events. However, the high loads exported via artificial drainage pathways suggest that management options such as riparian buffers may have limited impact on reducing *E. coli* transfer from drained pasture.

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