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Impact of slurry management strategies on potential leaching of nutrients and pathogens in a sandy soil amended with cattle slurry

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A B S T R A C T

For farmers, management of cattle slurry (CS) is now a priority, in order to improve the fertilizer value of the slurry and simultaneously minimize its environmental impact. Several slurry pre-treatments and soil application methods to minimize ammonia emissions are now available to farmers, but the impact of such management strategies on groundwater is still unclear. A laboratory experiment was performed over 24 days in controlled conditions, with undisturbed soil columns (sandy soil) in PVC pipes (30 cm high and 5.7 cm in diameter). The treatments considered (4 replicates) were: a control with no amendment (CTR), injection of whole CS (WSI), and surface application of: whole CS (WSS), acidified (pH 5.5) whole CS (AWSS), the liquid fraction obtained by centrifugation of CS (LFS), and acidified (pH 5.5) liquid fraction (ALFS). An amount of CS equivalent to 240 kg N ha^{-1} was applied in all treatments. The first leaching event was performed 72 h after application of the treatments and then leaching events were performed weekly to give a total of four irrigation events (IEs). All the leachates obtained were analyzed for mineral and organic nitrogen, electrical conductivity (EC), pH, total carbon, and phosphorus. Total coliforms and *Escherichia coli* were also quantified in the leachates obtained in the first IE.

The results show that both acidification and separation had significant effects on the composition of the leachates: higher NO_3^- concentrations were observed for the LFS and ALFS relative to all the other treatments, throughout the experiment, and lower NO_3^- concentrations were observed for acidified relative to non-acidified treatments at IE2. Acidification of both the LF and WS led to higher NH_4^+ concentrations as well as an increase of EC for treatment ALFS relative to the control, in the first IE, and lower pH values in the AWSS. Furthermore, the *E. coli* and total coliform concentrations in AWSS, LFS, and ALFS were significantly higher than in WSI or WSS. In conclusion, none of the strategies generally used to minimize ammonia emissions impact positively on leaching potential relative to the traditional surface application of CS. Furthermore, some treatments, such as separation, might increase significantly the risk of leaching.

1. Introduction

Close to one billion tonnes of animal manure and slurry are produced within the EU each year (Marmo et al., 2009). Consequently, over the last few years, animal slurry management has become a central activity in intensive dairy, beef, and swine farms. Treatments such as solid–liquid separation and anaerobic digestion have been developed to increase the slurry value and improve management but, in most of these treatments, the final product cannot be discharged directly to water bodies and is generally applied to agricultural soils as fertilizer. However, it is well known that slurry application to soil can lead to high emissions of ammonia (NH_3) and greenhouse gases (Chadwick et al., 2011; Webb et al., 2010) and may also result in water pollution due to the leaching of nitrate (NO_3^-) or pathogenic bacteria (Amin et al., 2013; Lee et al., 2007; Mantovi et al., 2006).

Minimization of NH_3 emissions following animal slurry application to soil has been the priority over recent decades (Webb et al., 2005), since they represent not only an environmental problem (Carozzi et al., 2013; Oenema et al., 2012) but also a significant decrease of the fertilizer value of slurry (Sørensen and Amato, 2002). As a consequence, several mitigation measures have been

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proposed to minimize NH_3 emissions during and after slurry application to soil (Ndegwa et al., 2008). Animal slurry injection is considered as one of the most effective solutions to minimize NH_3 emissions at the field scale and is now compulsory in some European countries (Carozzi et al., 2013; Webb et al., 2010). Nevertheless, this technique presents several limitations (cost, not applicable in some arable soils or grassland) and band application of pre-treated slurry could be a good alternative to slurry injection. As slurry pre-treatment, acidification is considered an efficient way to minimize NH_3 emissions at the barn and field scales, although its application is still restricted to a few countries such as Denmark (Kai et al., 2008; Oenema et al., 2012). Solid–liquid separation is another possible pre-treatment for the minimization of NH_3 emissions. Indeed, some authors (Petersen et al., 2003; Sommer and Hutchings, 2001) suggested that the application of the liquid fraction (LF) obtained by solid–liquid separation instead of whole slurry (WS) may also be efficient with regard to minimizing NH_3 emissions, assuming that the LF quickly infiltrates the soil.

All three of these NH_3 abatement strategies have proved to be efficient with regard to the minimization of NH_3 emissions but little is known about their impact on leaching losses and potential water contamination. Several studies focused on the impact of animal production on the environment, in terms of water contamination (Unc and Goss, 2004), but the introduction of new tools for slurry management, namely treatments such as solid–liquid separation or acidification, may alter the leaching of the slurry elements that are affected by such treatments. Also, the leaching potentials of slurry elements will differ according to whether slurry injection or surface application is used. Hence, we believe that the impact of these new technologies needs to be evaluated, to accurately define the best option that minimizes total nutrient losses to the environment and avoids the so-called “pollution swapping”. Indeed, the main risk associated with the minimization of NH_3 emissions from slurry amended soil is the high ammonium (NH_4^+) content of the amended soil, that can be quickly nitrified by soil aerobic bacteria (Cavagnaro et al., 2008). If the NO_3^- produced exceeds crop requirements, it can leach down through the soil and into the groundwater.

Our hypotheses are: i) surface application of the LF rather than WS will increase the leaching potential of nutrients and pathogens due to their greater exposure to percolating water, ii) acidification of WS or the LF will increase the leaching potential of nutrients and pathogens due to their potential solubilization and the decrease of dry matter (Fangueiro et al., 2009), iii) injection of WS will increase the leaching potential of nutrients and pathogens relative to surface application due to the position of the slurry in the soil column and/or slurry-soil contact (Bech et al., 2011; Glaesner et al., 2011). We also considered the acidification of the LF since it might prevent NH_3 emissions during storage.

The main objective of our study was to compare the impact of five slurry management strategies on the potential release of nutrients and pathogens into water, in soils amended with cattle slurry. For this, we quantified the leaching potential (proportion of applied contaminant leached) of amended soil after four simulated rain events.

2. Materials and methods

2.1. Soil sampling

Intact columns of a sandy soil were collected from an arable field located at Palmela-Portugal (N 38.57957; W 8.82954) under a typical Mediterranean climate. The soil is classified as Haplic Arenosol (IUSS, 2006). The field has not received any animal slurry in the last 12 years and is used for double cropping maize/ryegrass. For soil sampling, PVC columns (30 cm long, internal diameter 5.7 cm) were pushed into the soil to a depth of 25 cm and were then excavated by removal of the surrounding soil. The soil column was sealed at the bottom by a glass wool layer and a PVC net. The top surface of the column remained undisturbed. The soil columns were taken to the laboratory and weighed. Three of the columns were destroyed for a full characterization of the soil; the main characteristics are shown in Table 1. The soil was analyzed following standard laboratory methods (van Reeuwijk, 2002); cation exchange capacity was determined following method described by Chapman (1965).

2.2. Slurry

The cattle slurry (CS) used was collected from a dairy farm near Palmela (Portugal) and preserved at 4 °C in plastic barrels. The liquid fraction (LF) was obtained by centrifugation of the whole slurry (WS) at 4000 rpm for 10 min. The acidification of both the whole slurry (AWS) and the liquid fraction (ALF), to pH 5.5, was performed by addition of concentrated sulfuric acid (96%) at a rate of 4.5 ml kg^{-1} slurry.

The WS, AWS, LF, and ALF were fully characterized by following procedures described by Fangueiro et al. (2009). Analysis of *Escherichia coli* and fecal coliforms was performed by following a standard procedure (ISO 9308-2, 2012). The main characteristics of the different fractions are presented in Table 2. All slurry samples were stored at 4 °C until soil application.

2.3. Experimental design

Six treatments were considered: injection of the whole CS (WSI), surface application of the whole CS (WSS), surface application of the acidified whole CS (AWS), surface application of the liquid fraction (LFS), surface application of the acidified liquid fraction (ALFS), and a control without slurry application (CTR). Four replicates of each treatment were considered.

The soil columns were placed on a shelf equipped at the bottom with a funnel that allowed the recovery of the leachates. The amount of CS applied to each column was calculated in order to apply 240 kg N ha^{-1} (the maximum allowed legally in Portugal). This application rate is equivalent to 48 kg P ha^{-1} in WSS and AWS, 12 kg P ha^{-1} in LFS and 15 kg P ha^{-1} in ALFS. Slurry injection was simulated by placing the slurry in a slit (10 cm deep, 5 cm long and 2 cm wide) located at the center of the soil column. After slurry application, the slit was covered with the soil removed previously. For surface application, the different fractions were applied in a

Table 1
Soil characteristics – mean values of three replicates.

Characteristic	Unit	Value
Soil composition		
Clay	%	3.3
Silt	%	4.5
Sand	%	92.2
Porosity	$m^3\ m^{-3}$	0.45
Bulk density	$g\ cm^{-3}$	1.457
Cation exchange capacity	$cmol_c\ kg^{-1}$	2.938
Total C	$g\ kg^{-1}$	8.8
pH (H_2O)		5.7
EC	$\mu S\ cm^{-1}$	74.56
NO_3^-	$mg\ kg^{-1}$	43.4
NH_4^+	$mg\ kg^{-1}$	7.5
Total N	$mg\ kg^{-1}$	510.4
Total P	$mg\ kg^{-1}$	287.2
Available P	$mg\ kg^{-1}$	43.3

Table 2
Slurry characteristics on a fresh weight basis – mean values of three replicates.

Characteristic	WS	AWS	LF	ALF
Amount applied (g)	18	18	28	31
Dry matter (g kg^{-1})	119.2	164.3	27.8	31.9
pH	7.4	5.6	7.3	5.5
Conductivity (mS/cm)	19.1	22.4	18.2	28.0
Total C (g kg^{-1})	39.7	45.8	9.5	9.9
Total N (g kg^{-1})	3.5	3.4	2.2	2.0
NH_4^+ (g kg^{-1})	1.4	1.5	1.2	1.2
Total P (mg kg^{-1})	700.0	683.0	108.0	123.0
E. coli (MPN mL^{-1})	2.8E+05	7.0E+05	6.8E+04	7.8E+04
Fecal coliforms (MPN mL^{-1})	2.8E+05	1.1E+06	1.7E+06	2.4E+07

band without disturbing the soil surface. After application, a plastic ring was placed on top of the soil to minimize sidewall flow (Corwin, 2000; Lewis and Sjöstrom, 2010). The soil columns were weighed regularly to check the moisture content. Soil columns were kept at 20 °C during all the experiment.

2.4. Irrigation events

Rainfall was simulated by applying the same amount of distilled water to all columns. The first irrigation event (IE) was performed three days after slurry application and then IEs were performed weekly to give a total of four. The initial three-day delay between slurry application and the first IE was chosen bearing in mind the field situation, where slurry is not applied prior to rainfall events predicted by the three-day meteorological forecast.

At each IE, three pulses of 100 mL of distilled water were gently applied to the top of each soil column, following the procedure used by other authors (O'Flynn et al., 2013; Matlou and Haynes, 2006). A time interval of 15 min was kept between each pulse. The total amount of water added at each IE was equivalent to one pore volume.

For each IE, the total volume of the leachate was measured and a sample was analyzed for pH, electrical conductivity (EC), NO_3^-, ammoniacal N, total N, total C, and total P. For the first leaching event, the leachate was analyzed for fecal coliforms and *E. coli*.

The EC and pH were measured with a Metrohm 680 conductivity meter (Switzerland) and Russell RL 150 pH meter (Thermedics, USA), respectively. The $NH_4^+ - N$ and $NO_3^- - N$ were analyzed by spectrophotometry, using the Berthelot and sulfanilamide methods for NH_4^+ and NO_3^-, respectively, in an automatic segmented flow analyzer (Autoanalyzer Skalar, Germany). The total N and total C were analyzed in an elemental C and N analyzer (Formacs, Skalar Analytical B.V., Breda, The Netherlands), by combustion at 850 °C followed by NIR detection for C and chemiluminescence detection for N. The total P was determined in a segmented flow auto-analyzer (Skalar Analytical B.V., Breda, Netherlands) with a previous dialyzer step (Coutinho, 1996). The *E. coli* and fecal coliforms were analyzed according to a standard procedure (ISO9308-2, 2012).

2.5. Statistical analysis

The results were analyzed by analysis of variance (two-way ANOVA) to test the effects of slurry acidification, slurry type, and the acidification × slurry type interaction. A one-way ANOVA was performed to test the effect of each treatment relative to the control and time, independently. The statistical significance of the mean differences was determined by the Tukey test at a probability level of 0.05. The error bars shown in the figures represent the standard errors used for comparison in the Tukey test. The statistical software package used was Statistix.

3. Results

3.1. Potential leaching of nitrogen

The amounts of NO_3^- leached at IE1 represent less than 5% of the mineral N applied in all treatments and were significantly higher in LFS and ALFS than in WSS and AWSS (Fig. 1a). A strong increase of the NO_3^- concentration in the leachates was observed over the first week, for all treatments, and NO_3^- peaked at IE2. In addition, for the treatments involving acidification the peaks were prolonged until day 17, whereas in LFS and WSS the peaks were clearly defined on day 10.

The NO_3^- losses from WSI were significantly higher than those from WSS at IE2 (10 d) and IE4 (23 d), whereas at the other two IEs the values were not statistically different.

The effect of separation was significant at the first three IEs, with greater NO_3^- leaching from LFS than from WSS. Acidification had a significant effect at IE2 (lower NO_3^- leaching from acidified material) and IE4 (greater leaching from acidified material). It is worth

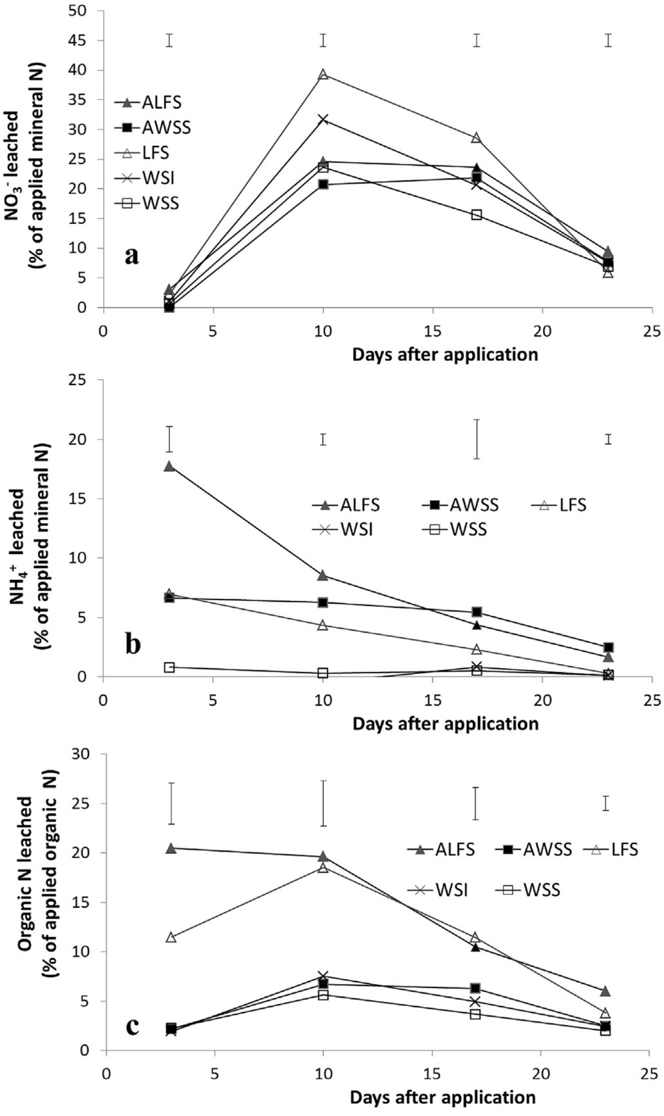

Fig. 1. NO_3^- (a), NH_4^+ (b), and N_{org} (c) leached in the five treatments considered during the four irrigation events, over a 23-day period. Mean of four replicates. Error bars represent the standard error values used for comparison in the Tukey test at each irrigation event.

noting that the interaction acidification × separation was significant at the first three IEs.

Considering the total amount of NO_3^- leached, the highest losses were observed in LFS (Table 3). Furthermore, treatment WSI induced higher losses of NO_3^- than WSS. Acidification had no effect on the loss (as NO_3^-) of the total N applied when comparing AWSS and WSS or ALFS and LFS, but the loss (as NO_3^-) of the mineral N applied was significantly lower in ALFS than in LFS. Less than 2.5 mg NO_3-N was lost by leaching in the control treatment over the 4 IEs (data not shown).

Leaching of NH_4^+ was observed in significant amounts only in treatments ALFS, AWSS, and LFS, whereas in WSS and WSI values remained similar to those of the control (<0.5 mg $NH_4^+ - N$) in all leaching events (Fig. 1b). The effect of separation on the leaching of NH_4^+ was significant at IE1 and IE2, with higher losses from LFS and ALFS relative to WSS and AWSS, respectively. For WS, the application method had no significant effect on the leaching of NH_4^+: for both WSI and WSS, less than 2% of the NH_4^+ applied was released over the whole experiment (Table 3). The interaction acidification × separation was significant for the first three IEs only. Nevertheless, the ALFS treatment led to the highest total NH_4^+ losses over the whole experiment, more than 32% of the NH_4^+ applied (Table 3).

For the LFS and ALFS treatments, a significantly higher amount of N_{org} was leached, relative to the other treatments, at the two first IEs (Fig. 1c). Similar amounts of N_{org} were leached for the other amendment treatments over the entire experiment. The effect of acidification on the leaching of N_{org} is not clear: no significant differences between treatments WSS and AWSS were observed, while differences between LFS and ALFS were significant only at IE1. However, the effect of solid–liquid separation was significant over the whole experiment, with higher amounts of N_{org} being lost from LFS and ALFS than from WSS and AWSS. Close to 35% of the N_{org} applied was leached during the whole experiment for LFS and ALFS, against 19–24% for the other amendment treatments (Table 3). The application method had no significant effect on N_{org} leaching.

As mentioned previously, the cattle slurry treatments considered here affect not only the amount of N released but also the N species that are leached (Fig. 2). Indeed, considering the proportion of total N applied, the potential leaching was highest for ALFS and LFS at the four IEs and these two treatments also led to the highest potential leaching of N_{org}. The acidification of CS, which kept higher amounts of mineral N in the form of NH_4^+, induced greater potential leaching of NH_4^+: 80% and 70% of the total N applied was potentially lost by leaching in soil receiving ALF and LF, respectively, against 28% in WSS, 37% in WSI, and 41% in AWSS (Table 3). The fractions of NO_3^- and NH_4^+ in the leachate indicate that, 23 days after slurry application, complete nitrification of NH_4^+ had occurred in all treatments except ALFS and AWSS. Furthermore, the N_{org} fraction in the leachates tended to decrease over time in all treatments.

Table 3
Amounts of NO_3^-, NH_4^+, N_{org}, total N, total C and total P lost by leaching over the four IEs in the treatments considered – means of four replicates.

Nutrient	Unit	WSS	WSI	AWSS	LFS	ALFS
Nitrate	% of total N applied	[a]19.27^{c}	25.14^{b}	22.05^{bc}	41.73^{a}	37.59^{a}
	% of N mineral applied	46.77^{c}	61.01^{b}	50.34^{c}	76.11^{a}	60.69^{b}
Ammonium	% of total N applied	0.72^{c}	0.15^{c}	9.13^{b}	7.63^{b}	20.02^{a}
	% of N mineral applied	1.76^{d}	0.37^{d}	20.85^{b}	13.91^{c}	32.33^{a}
Organic N	% of total N applied	7.97^{b}	9.89^{b}	9.93^{b}	20.41^{a}	21.55^{a}
	% of organic N applied	19.33^{b}	24.00^{b}	22.68^{b}	37.22^{a}	34.80^{a}
Total N	% applied	28.02^{d}	37.3^{c}	41.7^{c}	70.3^{b}	79.7^{a}
Total C	% of total C applied	2.64^{c}	2.28^{c}	0.11^{c}	22.05^{a}	12.25^{b}
Total P	% applied	2.40^{b}	1.45^{b}	0^{b}	0.02^{b}	22.04^{a}

[a] In each row, values followed by the same letter are not statistically different ($P < 0.05$).

3.2. Carbon and phosphorus leaching

Soil application of LF or ALF led to a significant increase of the TC concentration in the leachate, relative to the control values, over the whole experiment except in the case of ALF at IE3 (Fig. 3a). Less leaching of TC was observed in LFS relative to ALFS, but no differences were observed between WSS and AWSS. The slurry application method had no effect on the potential leaching of C. 22% and 12% of the total C applied was lost in treatments LFS and ALFS, respectively, over the whole experiment, whereas in all other treatments less than 3% was lost (Table 3).

During the first three IEs, the amount of P leached from the amended soil was lower than or equal to the amount leached from the control soil (Fig. 3b). However, at IE4, treatments ALFS and WSI led to significantly greater leaching of P than in the control. Slurry acidification or separation had no significant effect on P leaching, nor did the slurry application method. It is noteworthy that 22% of the P applied was leached in treatment ALFS over the four IEs, against less than 3% in all the other amendment treatments (Table 3).

3.3. pH and EC of leachates

The pH of the leachate was not affected by any treatment and remained constant over the four IEs for treatments WSI, FLS, and AFLS, whereas a significant decrease was observed for WSS, AWSS, and CTR (Fig. 4a) although this decrease cannot be attributed directly to slurry application.

An increase of the leachate EC, relative to the control, was observed at IE1 in the soils receiving ALFS, LFS, or AWS (Fig. 4b). It is of note that for ALFS an EC value of 1600 mS cm^{-1} was registered at IE1, whereas for all the other treatments the values were below 800 mS cm^{-1}. At the remaining IEs, the leachate EC of the amendment treatments was always significantly higher than for the control. Acidification of WS led to a significant increase of leachate EC over the four IEs, but this effect was observed only at IE1 in the case of LF. The effect of separation was significant only at IE1 and the slurry application method had no effect on leachate EC.

3.4. Leaching of pathogens

Higher populations of fecal coliforms were observed in the initial LF, relative to WS, and this trend was also observed in ALF, relative to AWS (Table 2). However, the *E. coli* population was greater in WS and AWS relative to LF and ALF, respectively. Acidification had a significant effect on fecal coliforms and *E. coli*: higher populations were observed in AWS and ALF relative to WS and LF, respectively. Interestingly, *E. coli* represented 100% and 65% of fecal coliforms in WS and AWS, respectively, but only 4% and 0.33% of fecal coliforms in LF and ALF, respectively.

The leaching of fecal coliforms was greatest in the soil amended with AWS (28.8%), whereas in ALFS less than 0.5% was leached (Fig. 5). Acidification of WS increased the leaching of fecal coliforms but the opposite effect was observed with LF acidification. No significant differences were observed between LFS and WSS or between WSI and WSS in terms of fecal coliforms. More than 44% of the *E. coli* applied was leached in AWSS and ALFS, against less than 24% in LFS and 1% in WSS. The potential leaching of *E. coli* was higher in WSI than in WSS, although the differences were not statistically significant.

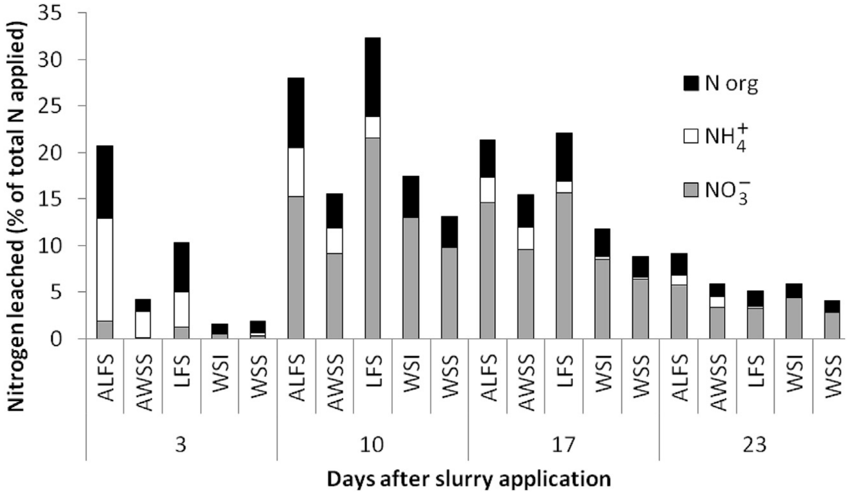

Fig. 2. Speciation of the nitrogen leached in the five treatments considered during the four irrigation events, over a 23-day period. Mean of four replicates. Error bars were removed for clarity.

4. Discussion

All the strategies considered here, known to minimize efficiently NH_3 emissions, increased the potential NO_3^- leaching relative to treatment WSS, except AWSS that gave results similar to WSS (Table 3). Delayed nitrification in soils amended with acidified cattle slurry has been reported previously (Fangueiro et al., 2010, 2013); consequently, NO_3^- leaching in AWSS was expected to be lower than in WSS. However, in the present study, the differences in the nitrification rate between acidified and non-acidified slurry were not significant, even if the peak of NO_3^- leaching was delayed by acidification. Our results indicate that separation increases the potential leaching of NO_3^- in sandy soils. Indeed, the higher NH_4^+ and labile carbon contents of the LF relative to the WS should have contributed to higher nitrification, followed by NO_3^- accumulation in soil and consequent leaching. Since the NO_3^- concentration of all the amendments was null before soil application, all the NO_3^- leached came from the nitrification process. Indeed, it is generally

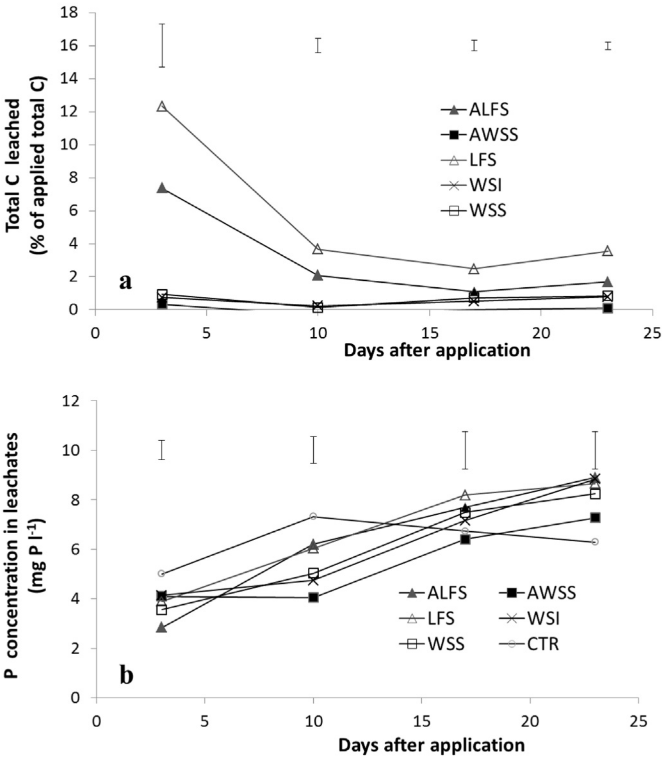

Fig. 3. Total C (a) and P (b) leached in the five treatments considered during the four irrigation events, over a 23-day period. Mean of four replicates. Error bars represent the standard error values used for comparison in the Tukey test at each irrigation event.

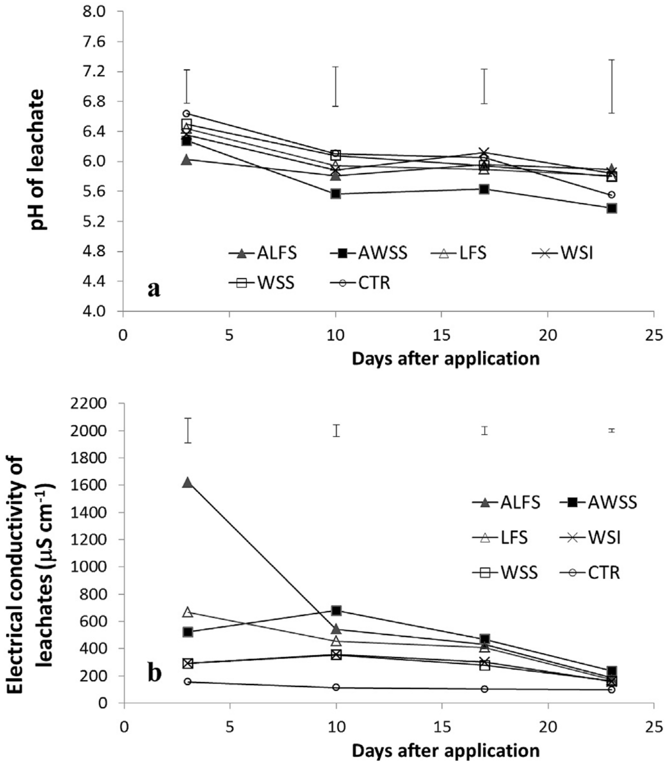

Fig. 4. pH (a) and EC (b) of the leachates obtained in the five treatments considered during the four irrigation events, over a 23-day period. Mean of four replicates. Error bars represent the standard error values used for comparison in the Tukey test at each irrigation event.

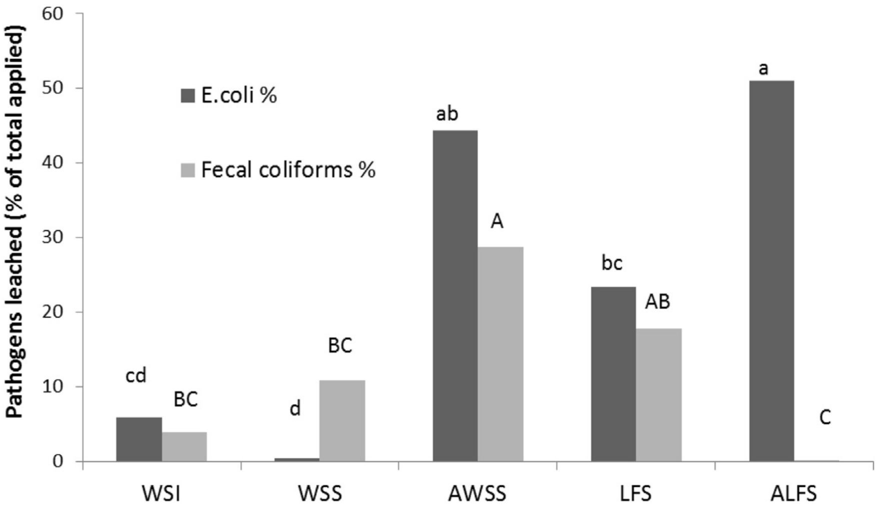

Fig. 5. Leaching of *E. coli* and fecal coliforms in the five treatments considered during the first irrigation event. Mean of four replicates. Similar letters indicate no statistical differences between values (normal letter for *E. coli* and CAPITAL for fecal coliform).

accepted that the average time for maximal nitrification to be reached ranges from 7 to 14 days (Addiscott, 1983) and, in our study, the NO_3^- peaks in the amendment treatments occurred on days 10 and 17 (for acidified amendments only).

Ammonium is less prone to leaching, due to its binding to soil particles (Eriksen et al., 2006), and can be rapidly converted into NO_3^-. Previous studies (O'Flynn et al., 2013; Svoboda et al., 2013) reported very low concentrations of NH_4^+ in the leachates from slurry amended soils, as occurred in some treatments of the present study, and these authors concluded that NH_4^+ contributed only marginally to nitrogen leaching. However, in the present study, close to 17% of the NH_4^+ applied was lost from the ALF at the first IE, which may be greater than the amount of NH_4^+ lost by NH_3 volatilization over the same period (Sommer and Hutchings, 2001). Leaching of NH_4^+ was observed here, due mainly to the sandy texture of the soil and its low cation exchange capacity. Furthermore, the small soil column length (25 cm) may have improved an unusual NH_4^+ leaching.

Treatments involving acidification led to higher potential leaching of NH_4^+, in agreement with the NO_3^- leaching potential and our hypothesis that acidification would delay nitrification. Furthermore, some NH_3 may have been released in the LFS treatments, since LF infiltration was not as fast as expected. So, it can be concluded that CS separation and/or acidification might increase the risk of NH_4^+ leaching, although one has to bear in mind that the present results are only indicative of what may occur in field conditions prior to plant growth. As occurred with NO_3^-, none of the strategies proposed here decreased the potential NH_4^+ leaching relative to WSS, but it is of note that injection did not induce an increase relative to WSS.

The main problem associated with N_{org} leaching is the subsequent potential for bacterial growth, phytoplankton growth, photochemical decomposition, and abiotic adsorption (Berman and Bronk, 2003). In the present study, the effect of acidification on N_{org} leaching is not clear, but the effect of solid–liquid separation was significant over the entire experiment with higher amounts of N_{org} lost from LFS and ALFS than from WSS and AWSS. As occurred with NH_4^+, the amount of N_{org} leached relative to NO_3^- was relatively small, in agreement with results from Svoboda et al. (2013). Nevertheless, in the case of LF and ALF, a large part of the N_{org} was in soluble forms and highly prone to leaching. Therefore, N_{org} leaching should also be considered in future experiments dealing with acidified slurries at the field scale.

More than 70% of the total N applied could be lost by leaching in soils receiving ALF or LF. Therefore, LF or ALF application to soil should be restricted to the spring, when the leaching potential is lower than in the autumn.

Cattle slurry separation increased the risk of TC leaching, whereas the effect of acidification was observed only with ALFS, probably because of the high C losses that occur during acidification (Fangueiro et al., 2013) It is of note that an increase in TC leaching was observed for WSS and AWSS at the last two IEs. Indeed, TC leaching can be delayed due to soil adsorption, namely for WSS (Dunnivant et al., 1992). As observed here, Amin et al. (2013) also found that TC leaching from soil amended with LF was higher than from soil receiving WS. An effect of acidification on P leaching was expected since previous studies (Daumer et al., 2010; Fangueiro et al., 2009) showed a strong dissolution of slurry P when the pH was lowered. However, P leaching in soil amended with animal slurry relies mainly on the soil properties (Glaesner et al., 2011) and indirectly on the manure application history (Koopmans et al., 2007). Indeed, Liu et al. (2012a) compared P leaching from two soils, a loamy sand and a clay loam, following pig slurry application and observed significant effects of slurry application on P leaching only in the clay loam soil, whereas in the loamy sand soil the leaching was similar in amended and non-amended soils, as occurred here. The P concentrations in the leachates obtained here are comparable with those reported by Liu et al. (2012b) in a similar soil and the weak effect of slurry application can be attributed mainly to the high baseline of P leaching, a result of the high available P content in the soil used here. Even after the application of 900 ml of distilled water, equivalent to three pore volumes, the P concentration in leachates from the amended soils was not significantly higher than in the control. Hence, it might be hypothesized that strong sorption of slurry P occurs after soil application. Furthermore, the effect of slurry injection (versus surface application) was probably masked by this strong soil sorption. Our results show that the application to a sandy soil of 50 kg P ha^{-1} as WS or AWS or 15 kg P ha^{-1} as LF or ALF should not be problematic in terms of P losses to water. This is in agreement with results obtained by other authors (Liu et al., 2012a; Sørensen and Rubæk, 2012) at the field scale, with similar soils.

A strong increase of the EC was observed in the leachates from the acidified treatments, relative to the non-acidified treatments. Indeed, as occurs with P, acidification induces the dissolution of some metal complexes that can be easily released into the soil water. We also observed a significant influence of CS separation on leachate EC, as reported previously by Amin et al. (2013) who also found higher EC values in leachates from soil receiving LF rather than WS.

In the present study, higher fecal coliform and *E. coli* populations were observed in acidified materials, in agreement with previous studies that indicated higher survival of *E. coli* and fecal coliforms at low pH (close to 6) than at high pH (close to 8) (Lin et al., 1996; Franz et al., 2005). Both LF and WS had a pH close to 7.5, higher than the optimum range of 6–7 for pathogens survival. A decrease of CS pH to 5.5 may have promoted the multiplication of pathogens since, after a few hours, the ALF and AWS pH values were close to 6. By contrast, a lower *E. coli* population was observed in the fractions obtained by separation, as reported also by Amin et al. (2013). This decrease may have been due to stronger attachment of *E. coli* to slurry particles, as suggested by Forslund et al. (2011). The potential leaching of pathogens was considered here only at the first IE. Indeed, according to Semenov et al. (2009), the highest risk of pathogen leaching is immediately after CS application. Nevertheless, as previously explained, weather forecasts have a significant confidence interval of almost three days, preventing slurry application when rainfall is expected. Therefore, we performed the first IE only three days after slurry application. Three aspects have to be considered here: the survival (or even multiplication) of the pathogens after acidification or separation, then after soil application, and finally the movement of the pathogens in the soil column. The effect of acidification on the potential leaching of fecal coliforms is not clear but it clearly increased the potential leaching of *E. coli*. By maintaining more mineral N in the slurry, acidification may prolong *E. coli* activity. Similarly, significantly greater *E. coli* leaching was observed for treatment LFS than for WSS, for the same reason (LF was richer in mineral N than WSS). Our results indicate that a higher amount of pathogens leached from the soils amended with materials obtained by separation, in agreement with results reported by Amin et al. (2013). The effect of the slurry application method was not significant, even though the mean potential leaching values of the pathogens following slurry injection were always higher than after surface application. Similar results were obtained by Bech et al. (2011) in a silt loam soil and previous studies showed that the slurry application technique (broadcast or injection) does not affect the potential leaching of *E. coli* (Amin et al., 2013; Forslund et al., 2011). Semenov et al. (2009) indicated that the leaching potential of *E. coli* in soil is affected by the type of manure (solid manure or slurry) and the method of its application (e.g., spreading on the soil surface or injection into the soil). They recommended surface application of slurry, to minimize the risk of water contamination.

5. Conclusions

The main objective of this study was to evaluate the impact of five cattle slurry management strategies on the potential leaching of nutrients and pathogens.

None of the strategies proposed decreased the potential leaching of slurry nutrients and pathogens relative to the surface application of WS, except AWS that decreased the potential leaching of TC (Table 4). Nevertheless, surface application of acidified cattle slurry increased the leaching risk for NH_4^+, salts, and pathogens. Injection was equivalent to surface application regarding several parameters but led to a worsening of NO_3^-, total P, and pathogens leaching. However, surface application of LF or ALF had a negative impact on the potential leaching and therefore should be recommended only in cases where the leaching potential is low. One has still to bear in mind that an efficient slurry management should minimize environmental impacts as well as human community impacts, namely in terms of odors and landscape, what give a strong advantage to slurry injection relative to other options tested here.

Table 4
Summary of the effect of separation, acidification, combined separation and acidification, and slurry injection on nutrients and pathogens leaching, relative to the surface application of whole slurry.

Parameter	Separation	Acidification	Separation + acidification	Injection
NO_3^-	↗	↔	↗	↗
NH_4^+	↗	↗	↗	↔
Organic N	↗	↔	↗	↔
Total C	↗	↘	↗	↔
Total P	↔	↔	↗	↗
Pathogens	↗	↗	↗	↗
pH	↔	↔	↔	↔
EC	↗	↗	↗	↔

Our results need to be confirmed with other soil types and then validated at the field scale since our soil columns may not be representative of field conditions, due to the influence of soil cracks and wall effects on water flow.

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