

Lactic acid fermentation of human urine to improve its fertilizing value and reduce odour emissions

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Abstract: During storage of urine, urea is biologically decomposed to ammonia, which can be lost through volatilization and in turn causes significant unpleasant smell. In response, lactic acid fermentation of urine is a cost-effective technique to decrease nitrogen volatilization and reduce odour emissions. Fresh urine (pH 5.2 ± 0.3 and $\text{NH}_4\text{-N}$ 1.2 ± 0.3 g L⁻¹) was lacto-fermented for 36 days in closed glass jars with a lactic acid bacterial inoculum from sauerkraut juice and compared to untreated, stored urine. In the lacto-fermented urine, the pH was reduced to 3.8 ± 0.7 and the ammonium content by 22–30%, while the pH of the untreated urine rose to 6.1 and its ammonium content increased by 32% due to urea hydrolysis. The concentration of lactic acid bacteria in lacto-fermented urine was 7.3×10^8 CFU ml⁻¹, suggesting that urine is a suitable growth medium for lactic acid bacteria. The odour of the stored urine was subjectively perceived by four people to be twice as strong as that of lacto-fermented samples. Lacto-fermented urine induced increased radish germination compared to stored urine (74–86% versus 2–31%). Adding a lactic acid bacterial inoculum to one week old urine in the storage tanks in a urine-diverting dry toilet reduced the pH from 8.9 to 7.7 after one month, while the ammonium content increased by 35%, probably due to the high initial pH of the urine. Given that the hydrolyzed stale urine has a high buffering capacity, the lactic acid bacterial inoculum should be added to the urine storage tank of a UDDT before urine starts to accumulate there to increase the efficiency of the lactic acid fermentation.

1. Introduction

Over the last decade, concern has grown in the world for efficient application of artificial nitrogenous fertilizers in order to reduce adverse environmental impacts including eutrophication, greenhouse gas effects and acid rain ([Gastal and Lemaire, 2002](#)). Alternative natural fertilizers capable of replacing or complementing mineral fertilizers need to be considered. Source-separated human urine is an excellent fertilizer that could be applied more widely owing to its nutrient content and universal availability. Fertilization of okra, cabbage, tomatoes and cucumber with urine generated similar or even higher yields compared to chemical fertilizers ([Akpan-Idiok et al., 2012](#); [Heinonen-Tanski et al., 2007](#); [Pradhan et al., 2007, 2009](#)). In fresh human urine, most of the nitrogen (75–90%) is present as urea [$\text{CO}(\text{NH}_2)_2$], with smaller amounts of uric acid, amino acids and other substances. When urine leaves the body ([Kirchmann and Pettersson, 1994](#)), only 7% is in the form of ammonia. In addition to nitrogen, urine contains phosphorous (H_2PO_4^- and HPO_4^{2-}) and potassium (K.) in ionic forms, calcium (Ca^{2+}), sulphate (SO_4^{2-}) and soluble organic matter ([Maurer et al., 2006](#)), which have a fertilizing effect as well. Urine-diverting dry toilets (UDDT) and waterless urinals are ideal systems for harvesting urine for use as fertilizer. However, misuse of UDDT systems may lead to cross-contamination of urine

with faeces and thus expose it to pathogens. To reduce the pathogen content to a safe level, it is recommended that urine be stored for 1e6 months (Jaatinen et al., 2016; WHO, 2006).

During storage, under the influence of bacterial activity and particularly the enzyme urease, urea is degraded into ammonia and carbon dioxide, which raises the pH these being the crucial factors for reinforcing pathogen inactivation (Nordin, 2010). At pH 8.9e9.0, 95% of the nitrogen in the stored urine is constituted of ammoniacal nitrogen (Kirchmann and Pettersson, 1994). As a result of becoming more alkaline and increasing the bicarbonate and ammonia concentration, the buffering capacity of urine increases as well (Udert et al., 2006). Once nitrogen is in form of ammonia, it can evaporate into the air (Udert et al., 2006). Aside from impacting the recovery efficiency of nitrogen, this triggers undesirable odour emissions which are intensified by other malodorous components, e.g. volatile fatty acids which are released by bacteria (Zhang et al., 2013). Odour emissions are a nuisance for toilet users, as well as for the neighbours of the agricultural fields where urine is applied. Different methods have been proposed to reduce ammonia volatilization and inhibit urea decomposition in urine. This can be successfully achieved by adding strong acetic or sulphuric acids (2.9 g L⁻¹) to keep the pH below 4 (Hellström et al., 1999), but this is not widely applied due to the cost of the acids and health risks involved in their handling (Maurer et al., 2006). There is also limited knowledge about the impacts these acids may have on soils and crops. An alternative method for stabilizing nitrogen is biological nitrification with the use of ammonia and nitrite-oxidizing bacteria, however sustaining bacterial activity in high strength ammonia solutions as urine is challenging and requires skilled staff (Udert and Weachter, 2012). Thus, there is a need to develop a cost-effective method to acidify urine that safeguards its fertilizer value and land applicability, minimizes energy input, and does not require sophisticated technical skills. Lactic acid fermentation (LAF) of urine is a simple and economical technique that can be carried out with different homemade fermented products that contain lactic acid bacteria (LAB), e.g. sauerkraut juice (Beganovic et al., 2014), and a source of carbohydrates. Moreover, there are no major health or environmental risks during urine treatment and there is no need for large investments or highly skilled staff. This study, therefore, focused on the efficiency of LAF for treating urine. The change in pH, chemical oxygen demand (COD) and ammonium concentration, buffering capacity and odour as well as the potential biological effects on plant germination of LAF treated urine was compared to that of (untreated) stored urine.

This research expands the knowledge regarding the sensitive practicalities of resource-oriented sanitation, which is currently limited. Maurer et al. (2006) suggest that focusing on the prevention of urea hydrolysis as the ultimate goal of urine stabilization, as it prevents nitrogen loss via ammonia volatilization and organic matter degradation (the main causes of odour), together with the precipitation of phosphorous compounds (the main cause of pipe clogging). This research benefits urine management of UDDTs since LAF of urine can reduce ammonia loss and unpleasant odours, while improving the fertilizing value of urine. The LAF process can also contribute to the potential of urine for fertigation (incorporation of fertilizer into irrigation water), by decreasing the risks of blocking the drip irrigation sets due to phosphorus precipitation.

2. Material and methods

2.1. Experimental set-up

Storage and LAF of fresh urine was performed under laboratory conditions in two trials, each with three replicates, over two periods of 36 days each time (in this period the pH is lowered to 3.8e4.7, thus hampering the urea hydrolyzation). The experiment was carried out from

December 2015 to January 2016 and from April to May 2016. The urine was kept at room temperature ($20 \pm \text{ }^\circ\text{C}$), and shielded from direct sunlight exposure. Urine samples were collected from 2 donors (a female 44-year old and a 7-year boy) for a period of three days and stored in 1 L glass jars tightly closed with a plastic lid. At the end of the collection period, all urine of both donors was thoroughly mixed, chemical analysis was performed and then the urine was separated into two parts.

The first part was mixed with a LAB solution (1:1) and LAF proceeded in the glass containers for a period of 36 days. The second part was stored in parallel in tightly closed glass containers, without any additions for the same period of time. The LAB solution was obtained by fermenting chopped cabbage over a period of one month, after which sauerkraut juice was extracted, collected, then mixed with sugar beet molasses and water at a proportion of 1:1:9 and kept in a closed plastic jar until the pH was reduced to below 5. After the treatments, chemical analysis was performed in both the LAF and stored urine samples. In addition, the LAB solution as well as the LAF urine was analysed for their *E. coli* and LAB concentration. No analysis of *E. coli* was performed in the untreated, simply stored urine.

The efficiency of LAF was also evaluated at field conditions in a functioning household UDDT in the vicinity of Chisinau (Moldova), used by a family of two adults (45-year-old male and 44-year-old female) and one seven-year-old boy. In this test, the 300 L plastic urine storage tank and the urine pipes were thoroughly washed and rinsed with vinegar prior to the experiments. Then, urine was collected in the tank for a period of one week (for pH and ammonia analysis) after which the LAB inoculum and molasses were added to the tank at the same ratio as in the laboratory experiments, which reduced the pH below 4.5. Each time the toilet was used, the urinal and urine compartment were sprayed with the inoculated LAB solution.

2.2. Odour evaluation

The intensity of the odour of LAF and stored urine was evaluated by four people (2 men and 2 women) independent from each other. Perception of the strength of the perceived odour was evaluated according to a rank scale from 0 (no odour) to 6 (extremely strong odour) as described in [Table 1 \(Misselbrook et al., 1993\)](#).

2.3. Germination tests

To evaluate if the urine samples stimulate germination, they were firstly diluted 1:10 with distilled water and then 3 ml was added to Petri dishes on Whatman filter pads together with twenty seeds of radish *Raphanus sativus*. As control, 3 ml of distilled water was used. The urine dilution rate (1:10) was obtained after a few germination tests and was required to adjust for the low pH of the LAF urine. After 72 h, germination was terminated by adding 3ml of 50% alcohol to each of the Petri dishes ([Tiqua et al., 1996](#)). The germination index was calculated according to [Mitelut and Popa \(2011\)](#) and [Tiqua et al. \(1996\)](#) (Eq. (1)):
$$GI = \frac{G}{G_{RRG}} \cdot 100$$
 (1) where G = number of germinated seeds in the sample/number of germinated seeds in the control and RRG = relative root growth = mean root length in the treated sample/mean root length in the control ([Mitelut and Popa, 2011](#); [Tiqua et al., 1996](#)).

2.4. Microbiological analysis

The LAB solution and LAF urine were thoroughly mixed and serially diluted 8 times. Then, 1ml of the solution was inoculated on agar plates. *E. coli* incubation was done on HiChrome Coliform agar at $43 \text{ }^\circ\text{C}$ for 24 h, while LAB incubation was done on M.R.S agar

(CMO361) at 36 °C for three days under anaerobic conditions (ISO, 1998; MHRF, 2005). The analysis was performed in duplicates.

2.5. Chemical analysis

The ammonium (NH₄-N) concentration was determined by a UVeVIS Analytik Jena Specord 210 spectrophotometer at 400 nm using cuvettes of 10mm according to the standard SM-SR-ISO7150-1:2005 (Zubcov et al., 2015). Urine was diluted 1000 times (0.5 ml/500ml of distilled water) from which 50 ml was taken and mixed with Seignette salt (C₄H₄KNaO₆) and Nessler's reagent. Chemical oxygen demand (COD) was analysed by dichromate oxidation, using the closed reflux method (Standardinform, 2014). Buffer capacity was determined by measuring the initial pH in urine with a Hanna portable EC/pH meter and titrating with 0.1 mol NaOH until the pH changed by one unit (Kirchmann and Pettersson, 1994). All chemical analyses were conducted in triplicates.

3. Results

3.1. Concentration of *Lactobacillus* and *E. coli* in LAB solution and LAF urine

The microbiological analysis indicated high bacterial counts of LAB both in the LAB solution and the LAF urine (Fig. 1). The LAB concentrations were, respectively, 7.5 and 7.3 log CFU ml⁻¹. Fresh non-hydrolyzed urine offered favourable growth conditions for LAB. *E. coli* was not detected in the LAB solution or in the LAF urine. *E. coli* may appear in urine in the case of urinary tract infection or potential faecal contamination during urine collection (Höglund et al., 2002; Kunin et al., 1992). Under the influence of LAB, *E. coli* growth is inhibited because of the low pH and excretion of inhibitory substances, such as bacteriocins, lactic acid or hydrogen peroxide (Saranraj, 2014). Nevertheless, some of the acid-tolerant strains of *E. coli* can survive in fermented sauerkraut (Niksic et al., 2005).

3.2. Changes in the chemical composition of urine during LAF

In LAF urine, the buffering capacity remained unchanged, while in the stored urine it approximately doubled (Table 2). The pH of the urine during LAF decreased to 4.7 and 3.8 due to the formation of lactic acid. At this pH value, the bacterial urease is normally inhibited and urea hydrolysis stops (Schneider and Kaltwasser, 1984). It is important to note that this is only effective with fresh urine; the hydrolysis process is irreversible, so the LAF needs to take place before ureolysis sets in. The ammonium content in LAF urine decreased by approximately 22-30% compared to the fresh urine (Table 2), from 1.2 to 0.9 g L⁻¹. In the untreated urine, the hydrolysis was not inhibited and the ammonium content increased by 32% compared to the fresh urine, from 1.2 to 1.6 g L⁻¹. In the urine tank of the UDDT, urea hydrolysis in the urine tank occurred at a much faster rate compared to that in the tightly sealed glass containers, with the pH increasing rapidly to 8.9 in only one week. The addition of the LAB inoculum and molasses in the urine tank of a UDDT contributed to a reduction of the pH of the urine by 1.25 units; however, it did not stop the hydrolysis. Even though the pH was reduced to 7.7 after 36 days of LAF, the ammonium content continued to increase and was 1.5 times higher than the initial value (Table 2). Compared to a previous analysis of one-month-old stored urine, the ammonium concentration was still only about half as much (Table 2). Additional research is required to test if the LAF technique can be more effective in preventing urea hydrolysis if the tank is inoculated before any urine enters it. During LAF, the soluble carbohydrates are converted to lactic acid leading to a pH decline, which inhibits the further decomposition of organic compounds (Murphy et al., 2007). Therefore, the COD value in LAF urine was expected to remain similar to that of fresh urine. In contrast, it was 15% higher (Table 3). Also in the

untreated, simple stored urine there was no reduction, but a slight increase of the COD concentration compared to the fresh urine.

3.3. The effect of LAF urine on seed germination

LAF removes the germination-inhibiting substances on the seed coat and decreases the number of seeds infected with fungi, thus promoting the health of seedlings and the vigor of mature plants (Szopińska, 2013). LAF urine had beneficial effects on the germination of seeds of *Raphanus sativus* relative to the control, with a germination index of 74e86%. One batch of stored urine had an ammonium content of 1.6 g L⁻¹ and resulted in a GI of 31% (Table 2, Fig. 1 A). In contrast, the other batch had a higher ammonium content (1.9 g L⁻¹) and a much greater inhibitory effect on germination, with a GI of 2% (Table 2, Fig. 2 B). In the urine tank, with an ammonium content of 4.6 g L⁻¹, the germination was almost entirely inhibited and the GI was 0.009% of the control (Table 2), probably due to toxicity of ammonia.

Another factor that might have influenced the germination index was the pH. LAF urine had a higher germination index in the first experimental run (pH . 4.7) than in the second run (pH . 3.8) (Table 2). The pH of the LAF urine with a GI of 74% after the dilution of LAF urine with water (1:10) was increased to only 4, such low pH values are usually unfavourable to germination, while the pH of the stored urine was 6.3 (1st run), which was beneficial to germination 3.4. Odour reduction during LAF of urine.

The odour of fresh urine was perceived as faint to distinct, that of the LAF urine was also faint to distinct and that of the stored urine was judged to be very strong to extremely strong (Table 4). In the toilet room, the urine odour was reduced compared to when no LAB solution was used for rinsing the urine compartment. In addition to the evaluation by these 4 persons, the variations were clearly apparent to the researchers during laboratory analysis. When diluted with distilled water, the fresh and LAF urine did not produce any nuisance. The odour almost dissipated after dilution, but the odour of stored urine, particularly which came from the urine tank, was more pungent and smelled noticeably even after dilution. Furthermore, after 36 days of LAF, the pH of LAF urine had decreased to 3.5 and the typical urine odour was replaced then by a medicinal or ester one.

4. Discussion

4.1. Urine as a suitable growth medium for LAB

This study showed that fresh urine can serve as a suitable growth medium for LAB, since their number in both the bacterial inoculum and LAF urine was higher than 7 log CFU ml⁻¹ after 36 days of incubation. The complex proteolytic system of LAB (Savijoki et al., 2006) probably allowed them to adapt to grow in the urine. Non-hydrolyzed urine is rich in important components, including peptides, urea, hippuric acid, amino acids, citric acid and minerals such as K., Na., Mg., PO₄³⁻, SO₄²⁻ and Cl⁻, which support or can stimulate the growth of LAB (MacLeod and Snell, 1947; Strong et al., 2005; Udert et al., 2006). Urine from healthy people also contains small quantities of carbohydrates such as glucose, lactose, xylose and arabinose (Ji Jin and Sam, 1999).

As LAB are not able to synthesize all amino acids by themselves, they need to receive certain essential amino acids and peptides in the growth medium (Niamsiru and Batt, 2000).

Conveniently, both non-hydrolyzed urine and molasses (added as a carbohydrate source for LAB) contain these (Dunn et al., 1947; Mee et al., 1979; Stein and Carey, 1953).

Amino acids and peptides can also serve as a nitrogen source, since LAB cannot catabolize mineral nitrogen (Saeed and Salam, 2013). During the current study, the $\text{NH}_4\text{-N}$ concentration decreased as a result of LAF. This might be due to the immobilization of free NH_3 into the bacterial cell wall (Ikawa and Snell, 1960) or $\text{NH}_4\text{-N}$ precipitation as ammonium lactate (Kuromiya et al., 2010), thus decreasing the $\text{NH}_4\text{-N}$ concentration.

4.2. Benefits of conducting LAF of urine

4.2.1. The fertilizer value of LAF urine

The ammonia and bicarbonate that are formed during urea hydrolysis by urease-positive bacteria contribute to an increase in the buffering capacity (Udert et al., 2006). Due to this high buffering capacity, it is not economical to treat urine with acids to decrease NH_3 volatilization (Udert et al., 2006). A combination of high buffering capacity, increased pH and high $\text{NH}_4\text{-N}$ concentration might also have a negative impact on soil bacterial nitrification, due to inhibition of nitrite oxidation and its accumulation in the soil (Burns et al., 1995). Nitrite accumulation is undesirable due to its potential phytotoxicity (Beauchamp, 1988). Another negative effect is the loss of ammonia following urine application, particularly in soils with high pH, high buffering capacity or low cation exchange capacity (Sherlock and Goh, 1984). The low cation exchange capacity of a soil reduces the amount of $\text{NH}_4\text{-N}$ cations bound on the exchange sites.

As a way to resolve these problems, this study showed that LAF can reduce the pH and ammonium content of urine. Studies have shown that a drop of pH below 5 decreases the urease activity (Schneider and Kaltwasser, 1984). The formation of free NH_3 and its loss via volatilization is also pH dependent. The highest NH_3 concentrations are present at a pH between 7 and 10 (Hartung and Phillips, 1994), which was obtained in the UDDT urine tank. In contrast at low pH (4.5 and below) as obtained with LAF of urine, no free ammonia is formed (Williams et al., 2011). Even though the addition of the LAB inoculum in the urine tank of a UDDT decreased the pH from 8.9 to 7.7, it could not stop urea hydrolysis and the accompanied increase in $\text{NH}_4\text{-N}$ concentration. Proteinaceous compounds, e.g. amino acids and peptides, from urine and molasses are more efficiently used by the proteolytic system of LAB (Niamsiru and Batt, 2000) than urea (Carvalho et al., 2011). Therefore, nitrogen in urine, after its LAF, will remain available mainly as urea. Urea based fertilizers are frequently used nitrogen fertilizers, own their popularity to low production costs, high nitrogen content and widespread availability (Glibert et al., 2006; Hawke and Baldock, 2010). However, urea applied to the soil can undergo hydrolysis to $\text{NH}_4\text{-N}$ under the influence of urease and is subsequently partly lost as NH_3 (Hawke and Baldock, 2010). It can be assumed that applying LAF urine will not cause such increased ammonia volatilization, since LAB and their associated low pH may act as urease inhibitors in the soil.

This study showed that the stored urine with the highest ammonium content completely inhibited germination of radish (Table 2). This is probably related to the toxicity of the increased NH_3 and $\text{NH}_4\text{-N}$ concentrations (Table 2). The stored urine kept in the glass jars with an ammonium content of 1.9 g L^{-1} contributed to a low germination index of 2% (Table 2), while that with a lower ammonium content (1.6 g L^{-1}) had a higher germination index of 31% (Table 2). Such a GI is still below the acceptable level of 80% when no toxic effect is encountered (Zucconi et al., 1981). Uptake of high concentrations of $\text{NH}_4\text{-N}$ by plants may alter the intracellular pH, reduce the synthesis of plant growth hormones, e.g. cytokinin (Britto and Kronzucker, 2002) and impair carbohydrate metabolism or photosynthetic and respiratory pathways of plants (WHO, 1986). The activity of LAB in LAF urine and the metabolites they produce may bring additional benefits to the plants and soil. Therefore,

LAB contributed to a 2e4 fold increase in tomato harvest weight of fruits compared to plants grown in unamended soil (Hoda et al., 2011). LAB may also contribute to the solubilization of the water insoluble phosphate compounds present in the soil, thus increasing their availability to plants (Zlotnikov et al., 2013). Additionally, LAB can suppress soil pathogens, e.g. bacteria and fungi (Hoda et al., 2011; Murthy et al., 2012), by producing different compounds with antagonistic activity including organic acids, hydrogen peroxide, cyclic peptides, and phenolic or proteinaceous compounds (Fhoula et al., 2013; Hoda et al., 2011). A better understanding of the effects of LAF of urine on nitrogen volatilization, plant uptake, nitrification and denitrification together with the benefits of LAB themselves on biological, physical and chemical components of the soil can be achieved via long-term field studies that are currently missing. One potential application of the findings from this study is the use of LAF urine in irrigation, particularly considering the need to increase water security in regions with limited water supply, e.g. mountainous regions (Yannopoulos et al., 2015) or arid areas. Climate change also increases the need to irrigate crops, in which approximately 46% of the cultivated areas in the world are not suitable for rain-fed agriculture (Valipour, 2016). The use of LAF urine in irrigation may be advantageous over simple stored urine due to lower risks of blocking the emitters of drip irrigation sets as its low pH prevents struvite precipitation. For example, struvite precipitation occurs at pH values between 7 and 11 (Doyle and Parsons, 2002). Moreover, a low pH also strongly reduces the risk of ammonia loss. In most UDDT systems it is recommended to store urine up to six months for hygienisation reasons (Welemaker et al., 2016). The WHO guidelines even recommend extreme pH and a high NH₄-N concentration, preferably in combination with an elevated temperature to kill off pathogens. Considering the value of LAF urine for fertigation, it is worth to further investigate the capacity of LAB to degrade micropollutants, such as hormones, pharmaceuticals and pesticides, which are excreted via urine and enter the aquatic environment upon urine application. An example is published by Caswell et al. (1977), who studied LAF of mixed corn grain and chicken manure bedding for a period of 80 days. During this period, the concentration of the veterinary drug sulfoquinoxaline, used in coccidiosis treatment, was significantly ($p < 0.01$) reduced by 39%. Owing to their specific enzyme systems, some LAB strains can also break down organophosphate pesticides, utilizing them as a carbon and phosphorus source (Cho et al., 2009; Zhang et al., 2014). The use of LAF urine for nutrients charging of biochar towards forming a slow-release fertilizer also requires further studies (Andreev et al., 2016; Schmidt et al., 2015). Ammonia and phosphate absorption onto biochar is more effective at a lower pH, with a maximal phosphate absorption in the pH range of 2.0 and 4.1, while this process is least effective at a pH higher than 6 (Spokas et al., 2012; Yao et al., 2011). The organic compounds present in urine form a type of coating onto the biochar, onto which anions and cations such as phosphorus and ammonium can bind (Schmidt et al., 2015). 2001). During LAF, LAB can metabolize amino acids via their enzymatic system into flavour compounds such as alcohols, aldehydes, esters or sulphur compounds (Savijoki et al., 2006). Therefore, the reduction of odour emissions during urine LAF was probably caused by the decrease in ammonia emissions and the synthesis of flavor compounds by LAB. For example, LAB can metabolize citrate from urine into diacetyl, acetoin and butanediol, which are important flavour compounds (Hassan et al., 2012). Also, hippuric acid, which is present in urine, can be used during LAF as a precursor for the synthesis of benzoic acid, which is another flavour compound (Güzel-Seydim et al., 2000). In contrast to the glass containers, where odour was considerably reduced after LAF, the odour was stronger in the UDDT urine tank (Table 4). This was probably due to the fact that free urease and urease positive bacteria prevailed in the pipes and urine tank. Therefore, urea hydrolysis and anaerobic decomposition of organic

matter occurred and the LAB could likely not dominate the processes in the urine tank, even though they contributed to a reduction in pH (Table 2). Another factor that might have hampered urine LAF was that the urine tank was not completely anaerobic, thus other microorganisms besides LAB could compete for organic matter and nutrients contained in the urine. This study showed that COD does not accurately reflect the decomposition of organic compounds from urine (Table 4). The increase in COD in the LAF urine, instead of an expected decrease, was due to the nitrogenous organic compound urea, which has no COD value, i.e. it is not oxidised in the standard dichromate method, explaining the increased value in LAF urine (Li et al., 2012). Besides, hydrogen peroxide generated by LAB (Kang et al., 2005) consumes the oxidation agent, potassium dichromate, leading to an overestimation of the COD concentration. For example, hydrogen peroxide present in anaerobically digested livestockwastewater led to a 9e14% overestimation of the theoretical COD values (Lee et al., 2011). Therefore, in addition to COD analysis, also the BOD, which is more specifically reflecting the biodegradable organic matter content, should be determined in further research on LAF of urine.

5. Conclusions

Urine LAF can be an effective low-cost technique that may lower ammonia volatilization and reduce odour emissions, thus increasing the sustainability and viability of UDDT systems. The addition of a LAB solution of sauerkraut containing molasses to fresh urine led to effective acidification to pH < 4.5 and a reduction by 1/3 of the ammonium content, while maintaining a high concentration of viable LAB (7.3 CFU ml⁻¹) compared to stored urine. In addition, LAF of urine reduced drastically the perceived odour strength and beneficially affected seed germination, showing a potentially higher fertilizing value than untreated, stored urine. Applying this technique to urine tanks may reduce odour and ammonia emissions, however additional research is required to transfer these research findings to practical applications. Urine LAF is also important when considering the increasing global demand for irrigated agriculture and the potential for reducing the risk of blocked drip irrigation systems due to phosphorus precipitation as is often the case with untreated, stored urine.

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