Urea stabilisation and dehydration for urine-diverting toilets

System and hygiene evaluation

Jenna Senecal
Faculty of Natural Resources and Agricultural Sciences
Department of Energy and Technology
Uppsala

Licentiate thesis
Swedish University of Agricultural Sciences
Uppsala 2017

Cover illustration by Donna-Lee Smith

Licentiate thesis/Report 096
ISSN 1654-9406
ISBN (print version) 978-91-576-9523-9
ISBN (electronic version) 978-91-576-9524-6

Print: SLU Service/Repro, Uppsala 2017

Urea stabilisation and dehydration for urine-diverting toilets: System and hygiene evaluation

Abstract

Over four billion people are discharging untreated human excreta into the environment without any prior treatment, causing eutrophication and spreading disease. This eutrophication is caused by nutrients found predominantly in urine. If managed adequately, urine can be used as a fertiliser because it contains the same plant nutrients as the fertilisers used to produce the food that people eat. Currently to replace the nutrients removed from fields during harvesting, more fertilisers are being manufactured and applied and ultimately more are being leached into the environment.

The use of human urine as a fertiliser is limited by its low nutrient concentration compared with commercial fertilisers. This study sought to increase the nitrogen (N) concentration (from 0.6 % to >6 %) through dehydration to produce a dry fertiliser of monetary value and where no liquid disposal from the toilet is required. The objective of this thesis was to evaluate a treatment that could stabilise urea and concentrate the urine while retaining >80 % of the NKP. Fresh human urine was added at various intervals to wood ash or biochar to first alkalise and thus inhibit the enzyme urease which catalyses the hydrolysis of urea. The urine was then dehydrated at temperatures of between 35 and 65 °C. A hygiene assessment was undertaken to observe the inactivation of five microorganisms (three indicators: *Enterococcus faecalis*, MS2 bacteriophage and ΦX 174 bacteriophage; and two pathogens: *Ascaris suum* and *Salmonella enterica* sub *enterica* Typhimurium) at the end of the alkaline dehydration process.

Urine mass was reduced by 95 % during dehydration, while preserving up to 90 % of the N and all the P and K. Ascaris inactivation data was fitted to a non-linear regression model, which estimated that 325 days of storage would be required for a 3 $\log_{10}$ reduction at 20 °C and 9.2 days of storage at 42 °C. The bacteria and bacteriophages were below the detection limit within four days at 20 °C. Just collecting urine separately from faeces provides a 5.2 $\log_{10}$ reduction. The material is concentrated during dehydration which results in a 3.5 log10 reduction overall just from urine-diversion.

This alkaline dehydration system installed in new or already existing toilets would greatly simplify the logistics and costs of storing, transporting and applying urine as a fertiliser. The truly innovative feature is the final product, a dry powder with 7.8 % N, 2.5 % P and 10.9 % K on dry weight, *i.e.* equivalent to commercial fertilisers. After just four days of storage, the dehydrated medium would meet WHO (2006) and USEPA (1994) guidelines for unrestricted fertiliser use.

Keywords: Alkaline disinfection, ash, dehydration, hygiene, pathogen, response surface optimisation, source separation, urea stabilisation, urine diversion, volume reduction.

Author's address: Jenna Senecal, SLU, Department of Energy and Technology, P.O. Box 7032, 750 07 Uppsala, Sweden

Ureastabilisering och torkning för urinsorterande toaletter: System- och hygienutvärdering

Sammanfattning

Mer än fyra miljarder människor har toalettlösningar som släpper ut obehandlad latrin i miljön utan någon behandling, vilket orsakar övergödning och sjukdomsspridning. Övergödning orsakas av växtnäring som främst kommer från urinen. Om urinen hanteras på rätt sätt kan den användas som gödningsmedel eftersom den innehåller samma växtnäringsämnen som kommersiella gödselmedel som används för att producera den mat vi äter. För att ersätta de näringsämnen som tas bort från åkern med skörden måste mer gödningsmedel produceras och användas, vilka i slutändan riskerar att läcka ut i miljön.

En huvudsaklig begränsningen för användandet av urin som gödningsmedel är den låga näringskoncentrationen jämfört med konstgödsel. I denna studie avsågs att öka kvävekoncentrationen i urin (från 0,6 % till > 6 %) genom torkning. Målet med denna process är att producera ett torrt gödningsmedel som har ett ekonomiskt värde. Fördelen med en torr produkt är att man inte behöver transportera och lagra vatten. Syftet med denna avhandling var att utvärdera en behandling för att stabilisera urea och därmed kunna koncentrera urinen bevarande >80% av näringen i den torra fraktionen. Färsk urin tillsattes till en torkbädd bestående av träaska eller biokol med högt pH vilket hämmar enzymet ureas från atthydrolysera av urea. Torkningsprocessen genomfördes sedan vid temperaturer mellan 35 och 65 °C. Inaktiveringen av fem mikroorganismer (tre indikatorer: *Enterococcus faecalis*, MS2 bakteriofag och ΦX 174 bakteriofag och två patogener: *Ascaris suum* och *Salmonella enterica sub enterica* Typhimurium) i slutet av torkningsprocessen studerades för att bedöma den hygieniska slutkvaliteten.

Under torkningen reducerades urinevolymen med 95 % samtidigt som upp till 90 % av kvävet behölls i torkbädden. Ascarisinaktiveringen anpassades till en icke-linjär regressionsmodell, med vilken det uppskattades att 325 och 9,2 dagars lagring krävs för en 3 $\log_{10}$ reduktion vid 20 °C respective 42 °C. Bakterierna och bakteriofagerna reducerades till under detektionsgränsen inom fyra dagar vid 20 °C.

Torksystemet som utvecklats i denna studie skulle i hög grad förenkla logistik och kostnader för lagring, transport och applicering av urin som gödningsmedel. Innovationen är slutprodukten, ett torrt pulver med 7,8 % N, 2,5 % P och 10,9 % K på torrvikts basis, dvs. motsvarande kommersiella gödningsmedel. Efter bara fyra dagars förvaring skulle den torkade produkten uppfylla WHO (2006) och USEPAs (1994) riktlinjer för obegränsad användning som gödselmedel.

Nyckelord: Alkalisk desinfektion, aska, hygien, källsortering, patogen, responsyteoptimering, torkning, urinsortering, urea-stabilisering, volymminskning.

Dedication

To Håkan, for starting this research group and keeping nutrient cycling at the discussion table. And to Sam, for always believing in me.

We are stuck with technology when what we really want is just stuff that works.
Douglas Adams

Contents

List of publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

I **Senecal, J.*** and Vinnerås, B. (2017). Urea stabilisation and concentration for urine-diverting dry toilets: Urine dehydration in ash. *Science of The Total Environment* **586**: 650–657.

II Simha, P.*, **Senecal, J**. and Vinnerås, B. (2017). Alkaline dehydration of anion-exchanged human urine: Volume minimisation, nutrient recovery & process optimisation (manuscript).

III **Senecal, J.*,** Nordin, A., Simha, P. and Vinnerås, B. (2017). Hygiene aspect of treating human urine by alkaline dehydration (manuscript).

Paper I is reproduced with the permission of the publishers.

* Corresponding author

The contribution of Jenna Senecal to the papers included in this thesis was as follows:

I Senecal and Vinnerås planned the study and Senecal performed the laboratory work. Senecal and Vinnerås wrote the paper.

II Simha, Senecal and Vinnerås planned the study. Simha performed the laboratory work and modelling and Senecal the urea loss calculations. Simha was the main author and Senecal contributed sections on urea and anion exchange, with revisions by Vinnerås.

III Senecal, Simha and Vinnerås planned the study. Senecal performed the laboratory work. Senecal and Nordin wrote the paper with revisions by the co-authors.

Abbreviations

dw	Dry weight (g)
NPK	Percentage of nitrogen, phosphorous, and potassium in elementary form a given product
RH	Relative humidity (%)
TS	Total solids (g)
VS	Volatile solids (g)
ww	Weight wet (g)
Compounds	
H_2CO_3	Carbonic acid
H_2N-COOH	Carbamate
$Ca(OH)_2$	Calcium hydroxide
$Ca_{10}(PO_4)_6(OH)_2$	Hydroxyapatite
Cd	Cadmium
Cl	Chloride
$CO(NH_2)_2$	Urea
K	Potassium
KOH	Potassium hydroxide
K_2SO_4	Potassium sulphate
KCl	Potassium chloride
$Mg(OH)_2$	Magnesium hydroxide
$MgNH_4PO_4 \cdot 6H_2O$	Struvite
$MgSO_4$	Magnesium sulphate
N	Nitrogen
N_2	Dinitrogen (nitrogen gas)
NH_3	Ammonia
NH_4^+	Ammonium
NO_3^-	Nitrate
P	Phosphorous
PO_4^{3-}	Phosphate

1 Introduction

The saying ‘we are what we eat’ is only part of the story. What we eat is what we excrete, at least in terms of plant nutrients. Human excreta contain the same nitrogen, phosphorus and potassium (NPK) minerals as the plant fertilisers used to grow the food humans consume (Winker et al., 2009). However, human excreta are considered unwanted waste throughout the world, creating humanitarian and environmental problems (Baum et al., 2013). In order to replace the nutrients removed from the fields during harvesting, more fertilisers are manufactured and applied to the fields, releasing even more nutrients into the environment and further contributing to environmental changes at a global level (Smil, 2002, Steffen et al., 2015). Recycling human excreta back to agricultural fields would reduce the current dependence on fertilisers derived from non-renewable resources (Ramírez and Worrell, 2006). It could also improve crop yields, for example in sub-Saharan Africa where fertiliser application is low (FAO, 2015), and protect marine ecosystems in many places by limiting the flow of excess nutrients to surface waters (Steffen et al., 2015). With 4.1 billion people currently lacking access to improved sanitation systems (Baum et al., 2013), there is an immense need for better sanitation technologies that do not require complex installation.

Urine, rather than faeces, contains the majority of the NPK excreted. Each person annually excretes in their urine: 80-90 % of the total 4 kg of N, 50-80 % of the 0.4 kg of P and 80-90 % of the 1 kg of K (Vinnerås et al., 2006). The main limitation with using urine as a fertiliser is that it is mostly water (97 %), meaning that its concentration of nutrients is low. For example, the N concentration in urine (without flush water) is about 0.6 % (Vinnerås et al., 2006), whereas that of the manufactured fertiliser urea is 46 %. Lower nutrient concentrations therefore require larger quantities of urine to be applied per hectare as fertiliser, which creates logistical problems in terms of storage (since approximately 500 L of urine are produced per person per year) and increases the costs of transportation and application. Hence urine, as excreted,

is not a competitive fertiliser. To improve its competitiveness, the excess water in urine needs to be removed.

Removing the excess water without losing the nitrogen is a challenge that several technologies have been designed to overcome (Section 3.3) (Maurer et al., 2006). However, the implementation and acceptance of such technologies is still limited. Furthermore the technologies generally collect only one or two of the elements in the concentrated fraction, *e.g.* N by ammonia (NH_3) stripping, or P and a small fraction of N in struvite.

2 Aims and structure

2.1 Overall aim

The overall aim of this study was to identify an appropriate way to circulate plant nutrients from human excreta back to the field to cultivate food again (Figure 1). The focus of this thesis was on circulating urine with the aim to evaluate a treatment to stabilise the urea in urine in a way that specifically retains the N during dehydration, along with all the P and K.

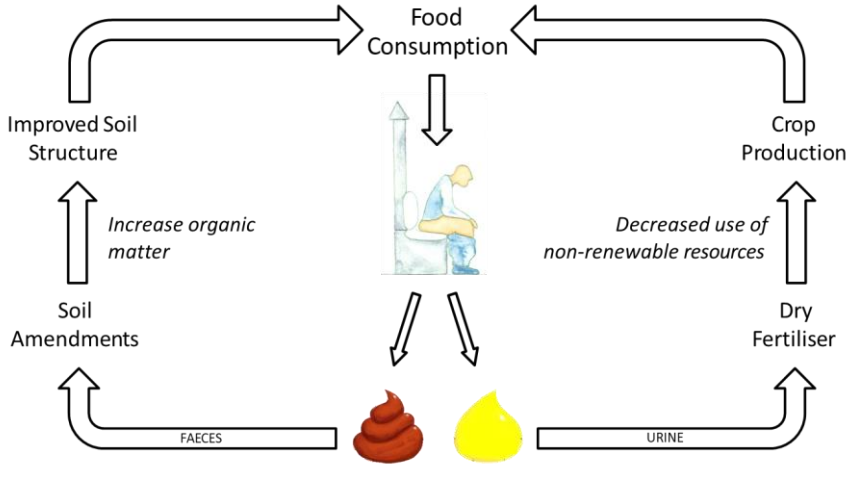

Figure 1. Circulating plant nutrients from the toilet back to fields to more grow food to be consumed and excreted once again.

The detailed aims were to provide responses to the following questions:

- Is it possible to dehydrate human urine in an alkaline medium, while retaining >80 % of the total nitrogen? (Paper I & II)
- Will the use of an initial anion exchanger increase the pH of the urine to a level that allows unlimited amounts of urine to be dehydrated in the same dehydration medium? (Paper II)
- How is the hygiene affected by dehydrating urine in an open, ammonia-free, alkaline system? (Paper III)

2.2 Structure of the thesis

This thesis is based on three papers (Figure 2). Paper I evaluated the effectiveness of a stabilisation treatment in ash to enable urine dehydration while retaining the nitrogen. The proof of concept in Paper I led to Paper II, which sought to optimise stabilisation and dehydration. The use of an anion exchanger was examined as a method for stabilising urine before being added to the dehydration medium, with the intention of increasing the dehydration capacity of the medium. Paper II explored the effect of different the

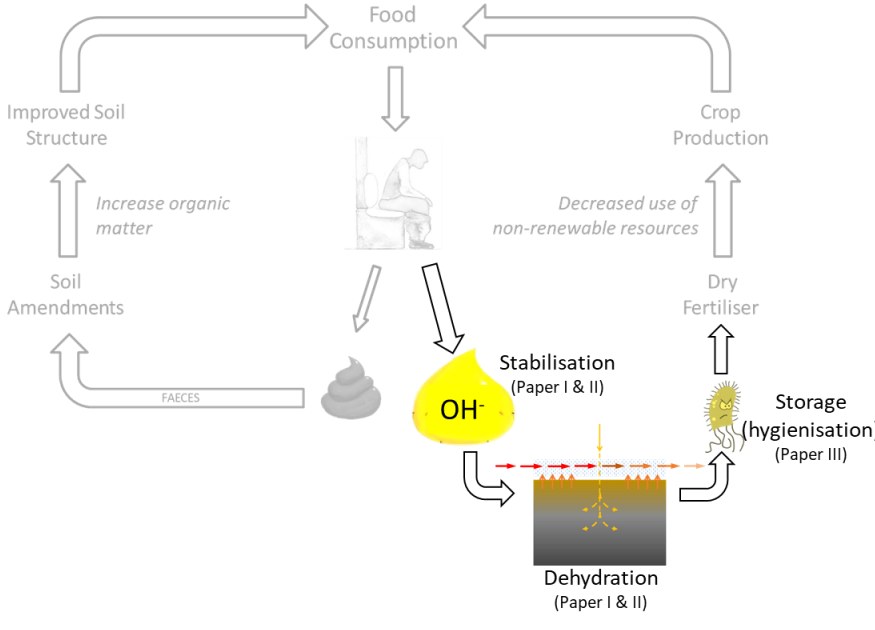

Figure 2. This thesis was focused on three processes to dehydrate urine: stabilisation of the urea in urine (Paper I-II); dehydration of urine to concentrate the nutrients (Paper I-II); and the effect of storage on indicator and pathogenic organisms (Paper III).

dehydrating parameters to decrease the surface area and dehydration time required. In urine-diverting toilets, there is a risk of cross-contamination from faeces and Paper III assessed how indicator and pathogenic microorganisms fared in the dehydrated medium. The ideal dehydration conditions found in Paper II were used to dehydrate the urine in Paper III.

3 Background

Plants take up minerals while they grow and then humans consume these minerals through various forms of food and excrete the minerals in their urine and faeces. The plants' productivity is dependent on having good access to these minerals, yet urine, which contains most of the excreted minerals, is not a valued resource. As we are what we eat and what we eat is what we excrete, the minerals needed for plant growth can be found in excreta, and the most of important plant nutrients (N, P, K and S) are found in the urine. While dehydrating urine, all P, K, S etc. will be retained. The big challenge however is N, as it is easily lost during the dehydration. Hence this background focuses on the challenge of retaining N during the dehydration process.

3.1 Nitrogen's boundaries

Nitrogen (N) is required by plants more than any other nutrient (based on the number of atoms, excluding C from CO_2 and H from H_2O) and is often a limiting nutrient for non-leguminous plants (Camberato, 2011, Jönsson et al., 2004). An increase in N fertilisation levels often results in more plant growth than an increase of any other nutrient (Camberato, 2011), and hence N in various forms is the most widely produced and used fertiliser (FAO, 2015). N is the most abundant element in the atmosphere and 99 % of it is in the inert gaseous form, dinitrogen (N_2) (Camberato, 2011). However plants are unable to utilise this form of N. Plants mainly use two inorganic forms of N: ammonium (NH_4^+) and nitrate (NO_3^-), the latter being favoured by most crops (Jönsson et al., 2004, Lasa et al., 2000).

There are three dominant paths to fix atmospheric N_2 into a form that is more usable in the biosphere: lightning (5×10^9 kg N $year^{-1}$), biological N_2

fixation by an enzyme in prokaryotic microbes called nitrogenase (1.0-1.4 x 10^{11} kg N $year^{-1}$), and the Haber-Bosch N_2 fixation process (1.1 x 10^{11} kg N $year^{-1}$) (Camberato, 2011). Lightning fixation is too uncontrollable to be used as a regular production method and there are only a few species of bacteria and algae that utilise the N_2-fixing enzyme (Prosser, 2011); which means that this fixed N_2 in only available to a few types of crops in symbiosis with bacteria utilising the N_2-fixing enzyme. The Haber-Bosh process is the method mainly used for extracting inert N_2 from the atmosphere and converting it into a reactive form that plants can use.

Before the invention of the Haber-Bosch process in the early twentieth century, human excreta had been used in agriculture for thousands of years (King, 1911). However the practice of using human excreta as fertiliser had been falling out of favour in some countries even before the Haber-Bosch invention due to the introduction of water closets that limited access to excrement (in that excrement was diluted with water) and the increase in knowledge regarding bacteria and the spread of disease in connection with faeces (Höglund, 2001). The increase in animal husbandry and the increased use of synthetic fertilisers during the green revolution further decreased the desire to use human excreta as a fertiliser (Shiming, 2002).

A problem with this shift is that the mass of reactive N in the biospheric cycle has doubled since the pre-industrial era, causing aquatic, terrestrial and atmospheric changes at a global level (Smil, 2002, Steffen et al., 2015). This increase in reactive N in the biospheric cycle is largely caused by the plants not taking up all of the reactive N that is applied as fertiliser (as well as the increase in population and the lack of wastewater treatment). Due to leaching and volatilisation, much of it ends up in the biosphere as harmful (reactive) forms of N (Figure 3).

To understand the implications of human activities, Rockström et al. (2009b) presented a new approach of planetary boundaries for defining biophysical thresholds of the Earth. The N cycle is one of nine planetary boundaries and its boundary threshold has been overstepped more than twofold (Steffen et al., 2015). The proposed boundary is to limit the industrial and agricultural fixation of N_2 to 62 x 10^9 kg N $year^{-1}$ (or approximately 41 % of N_2 fixed in the terrestrial ecosystem per year) (Steffen et al., 2015).

Fixed N_2 is used inefficiently today. N fertiliser is manufactured and applied to the fields where the plants take up about 50 % of N applied, with the rest entering the environment, largely as reactive N and some as N_2 through nitrification and denitrification processes in the soil (Smil, 2002). The harvested crops are used for human and animal consumption (Figure 3), where

the N from the consumed food is then excreted, predominantly in urine (Kirchmann and Pettersson, 1994). As worldwide an estimated 55 % of human excreta is not treated at all before entering the environment (Baum et al., 2013), most of the N flows into aquatic ecosystems where it is either denitrified into N_2 (desired) or contaminates the aquifer with high nitrate levels, causing eutrophication (not desired) (Smil, 2002). The N lost from the field through the harvested crops, leaching *etc.* is then replaced with new manufactured N fertiliser – further pushing the N planetary balance above the planet's threshold as more reactive N is released into the environment (Rockström et al., 2009a). Human use of N could decrease drastically with

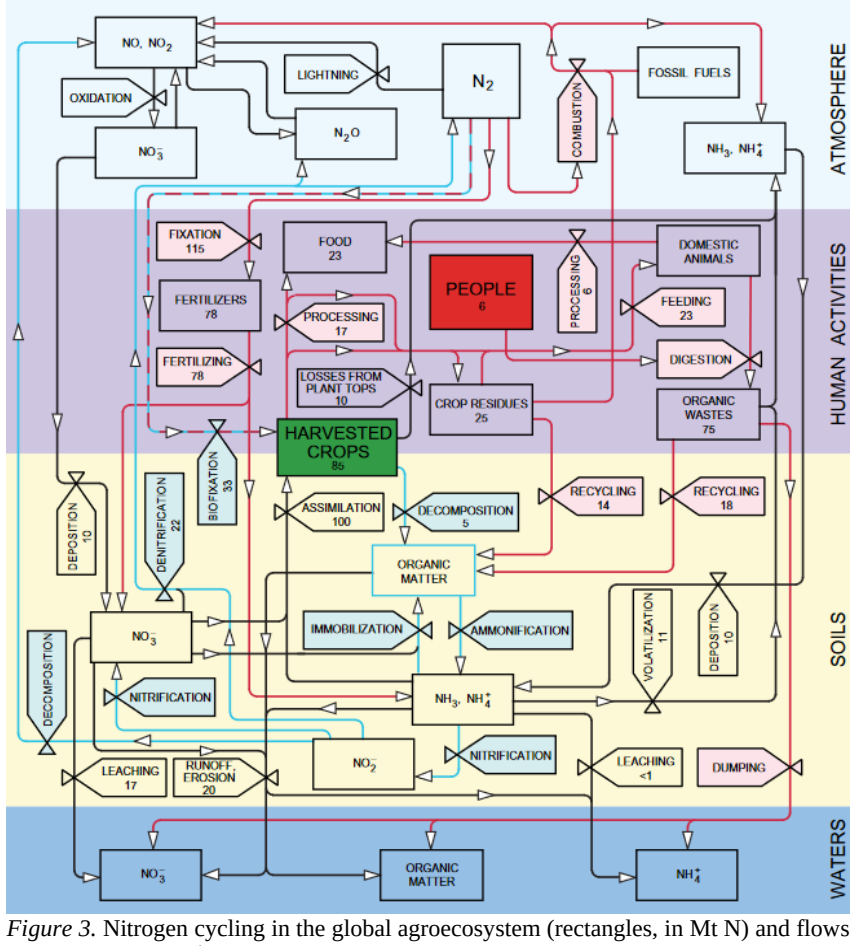

Figure 3. Nitrogen cycling in the global agroecosystem (rectangles, in Mt N) and flows (valves, in Mt N yr^{-1}). Flows impacted by human activity (red) and bacterial metabolism (blue) are highlighted. Image used with the permission of Smil (2002).

more efficient nutrient use and management, such as recycling human excreta as fertiliser (Smil, 2002, Rockström et al., 2009a, Steffen et al., 2015).

3.2 Urease enzyme and urea stability

The N in freshly excreted urine is predominately found as urea (~85 %), NH_3/NH_4^+ and uric acid; which together account for 90-95 % of total N in urine (Kirchmann and Pettersson, 1994). The urea in fresh human urine is stable in the absence of active urease enzymes (urea amidohydrolase, EC 3.5.1.5) (Feng et al., 2008). Urease enzymes are most commonly known for their role in soil fertilised with urea but, unknown to most toilet users, human faeces contain large amounts of urease-forming bacteria (Wozny et al., 1977). Even in urine-diverting dry toilets, urease enzymes are found in urine piping systems due to cross-contamination from faeces and biofilm formation on the pipe surface and will cause rapid hydrolysis of urea to NH_3 (Vinnerås and Jönsson, 2002).

Once the urine is excreted into the toilet, the urease enzymes already present in the toilet quickly hydrolyse urea ($CO(NH_2)_2$) into its volatile form, ammonia (NH_3) (Eq. 1). The carbamic acid (H_2NCOOH) produced in this reaction then spontaneously hydrolyses into carbonic acid (H_2CO_3) and releases a second NH_3 molecule (Krajewska, 2009). NH_3 is highly soluble in water, but the concentration is dependent on temperature and pH and lost as a gas to the air above the water at a rate proportional to the partial pressure of $NH_{3(g)}$ above the solution (Nordin, 2010). Hence in a dehydration system with a large air flow, the NH_3 would be lost. It is hard to prevent the urea hydrolysis reaction (Eq. 1) since the enzymes are found everywhere.

$$CO(NH_2) + H_2O \xrightarrow{\text{Urease}} H_2NCOOH + NH_3 \xrightarrow{H_2O} H_2CO_3 + 2NH_3 \quad \text{(Eq. 1)}$$

Ureases are a group of highly efficient natural enzymes used in plants, algae, fungi and several microorganisms to catalyse the hydrolysis of urea (Ciurli et al., 1999). Urease, as an extracellular enzyme, can be immobilised on particles, continuing its degradation of urea there (Ciurli et al., 1996). The structure of the urease enzymes varies between different urease-forming bacteria (enzyme molar mass range 190-300 kDa), the rate of activity and optimal pH (Krajewska, 2009), but all the enzymes have a common feature of two nickel ions at the active site (Blakeley et al., 1982).

To limit the urease enzymes in agricultural soils (slow the release of N fertiliser so that the plants have more time to take it up), inhibitors such as N-(n-butyl) thiophosphoric triamide have been developed (Parker et al., 2012).

However, due to the potential risks to human and environmental health (Ciurli et al., 1999), they cannot be considered a viable option for use in household toilets. The urease enzymes can also be inhibited by pH (> 10) and temperature (>80 °C) (Hotta and Funamizu, 2008, Huang and Chen, 1991, Sizer, 1940, Geinzer, 2017, Kabdaşlı et al., 2006). At an elevated pH (>12), the urease enzymes activity may be inhibited, but urea starts to degrade by an elimination reaction with OH^- as the base catalysis (Eq. 2 and 3) (Blakeley et al., 1982). With regard to temperature, urea is very stable at 25 °C in neutral pH with a half-life of 40 years, but as the temperature increases the half-life decreases (Table 1). However, the elimination reaction and the thermal degradation of urea takes $>10^7$ times longer than enzyme-catalysed hydrolysis (Table 1). Hence the priority is to limit the urease enzyme by elevating the pH to enable dehydration of the urine with minimal N loss.

$$CO(NH_2) + \mathbf{OH^-} \rightleftharpoons CON_2H_3 + H_2O \qquad \text{(Eq. 2)}$$

$$CON_2H_3 + H_2O \rightarrow HNCO + NH_3 + \mathbf{OH^-} \qquad \text{(Eq. 3)}$$

Table 1. Examples of the half-life of urea depending on the temperature, pH and catalyst

Conditions	t½
Uncatalysed	
25 °C (neutral pH)[z]	40 y
38 °C (2<pH<12)[y]	3.6 y
65 °C (2<pH<12)[x]	15.3 days
Base catalysis	
20 °C (pH>10) [w]	Negligable at 32 days
65 °C (pH>12.5)[x]	14.1 days
Enzymatic catalysis	
Jack-bean (neutral pH, 25 °C)[z]	0.02 s

[z](Callahan et al., 2005)

[y](Zerner, 1991)

[x]Derived from Warner (1942)

[w](Kabdaşlı et al., 2006)

3.3 Concentrating and stabilising nitrogen in fresh urine

Human urine as a potential source of nutrients has led to various trials to concentrate urine where the urea has already been hydrolysed. Examples include NH_3 stripping (Antonini et al., 2012), nitrification (Udert et al., 2015, Feng et al., 2008), electrolysis (Udert et al., 2015, Ledezma et al., 2015), struvite formation by the addition of magnesium (Etter et al., 2011), osmosis (Liu et al., 2016) and many others that are reviewed by Maurer et al. (2006). However, the implementation and acceptance of such technologies is limited due to the sensitivity around them, high requirements for chemical inputs or the complexity of the technology. Furthermore, most of the concentrating technologies collect only some of the elements in the concentrated fraction, *e.g.* N in NH_3 stripping, or P and some of the N in urine by struvite precipitation.

Another approach is to first stabilise urea in the urine by a pre-treatment to limit urease activity. Keeping the urea as urea enables dehydration of the excess water in urine with limited N loss. Examples of stabilisation techniques include acidification (Hellström et al., 1999), alkalisation (Randall et al., 2016) and salinisation (Pahore et al., 2010). When the urease enzyme is limited by elevated pH and/or elevated temperature, urea degradation still occurs, but at a much slower rate (Table 1) (Jespersen, 1975).

Stabilising urine by alkalisation is an attractive option as there are several sources of strong bases available with low solubility, which means that small volumes would be required to increase the pH. The end product (fertiliser) would still be alkaline, which could be beneficial as alkalisation is often needed due to acidification from fertiliser application (in particular urea), legumes fixing N_2, and mineralisation (Goulding, 2016). Slaked lime or calcium hydroxide ($Ca(OH)_2$) has been demonstrated to effectively increase the pH to 12.46 and inhibit urease activity in a chemical reactor prior to drying in a separate system (Randall et al., 2016). Other alkali salts (magnesium hydroxide ($Mg(OH)_2$), potassium hydroxide (KOH), and sodium hydroxide (NaOH)) are available and contain nutrients required by plants.

Wood ash is also alkaline (pH > 12, Paper I). During combustion, the biomass is oxidised and the inorganic species freed and transformed into oxides, hydroxides, chlorides, sulphates, carbonates (such as $CaCO_3$) and crystalline potassium salt (such as K_2SO_4, KCl and $K_2Ca(CO_3)_2$) (Steenari, 1998). The final composition of the elements and the pH depend not just on the source of wood, but on the temperature and method of combustion as well.

The pH of urine can also be increased by an anion exchanger. Ion exchanging is a method commonly used to soften water or sometimes in wastewater treatment plants to remove nitrogen, sulphates and heavy metals

(Tchobanoglous et al., 2003). Anions present in urine can be exchanged for OH^- when passed through a resin, such as the strong basic resin Amberlite™ IRA 410 type 2 (dimethyl ethanol ammonium). Amberlite has a molar equivalent of 1.25 eq L^{-1} where hydroxide anions are exchanged with the anions in the urine, increasing the pH. In human urine, the anions present, in order of strength of charge, are $HCO_3^- < Cl^- < NO_3^- < CO_3^{2-} < SO_4^{2-} < PO_4^{3-}$ (Ferslew et al., 2001, Putnam, 1971). The anion present in the highest concentration is Cl^-. Theoretically, the average exchange capacity of urine is 0.23 eq L^{-1} (Table 2), giving a volume ratio (urine to resin) of 5.4:1.

Table 2. Anions present in human urine in decreasing order of concentration in human urine and the molar equivalent used to calculate the volume of resin required

Anion	Urine[z] (mg L^{-1})	Ionic mass (g M^{-1})	Equivalent mass[y] (g eq^{-1})	Mean Equiv. (eq L^{-1})
Cl^-	5137	35.5	35.5	0.145
PO_4^{3-}	1649	95	32	0.052
SO_4^{2-}	1115	96	48	0.023
HCO_3^-	560	61	61	0.009
CO_3^{2-}	150	60	60	0.004
NO_3^-	29	62	62	0.000
Total	**8055**		**Total =**	**0.233**

[z] (Ferslew et al., 2001, Putnam, 1971)
[y] is the molar mass divided by the charge

3.4 Dehydrating urine

The surface tension of urine (42.4-67.5 mN m^{-1}, at 21 °C, varying with total solids (Putnam, 1971)) is lower than that of water (72.8 mN m^{-1}, at 21 °C (Tchobanoglous et al., 2003)) due to the surfactant activity from amphiphilic and hydrophobic solutes such as bile salts (Mills et al., 1988). This suggests that the evaporation rate of urine is higher than that of water, however in reality this is not the case. Over time, a visible film forms on the surface of urine that greatly decreases the evaporation rate. This layer is probably composed of amino acids (such as glycine) that over time are being released from the organic compounds (606 mg L^{-1} (Putnam, 1971)) in the urine. Amino acids have been shown to increase the surface tension (Matubayasi et al., 2003). Glycine increases the surface tension of a solution even more as the pH increases from 7 to 11 (Al-Husseini, 2015). Hence, a method to disrupt the surface is needed to increase the efficiency of dehydrating urine.

The system in the present study used ash and biochar were used as a dehydrating medium. Both are porous material and the urine would either be unbound or bound during dehydration. The characteristics of the unbound urine during dehydration would be like freestanding water as the liquid is not adsorbed to the medium. The bound urine would require additional energy (heat) for the phase-change from liquid to gas.

There are four stages in the dehydration of liquid from a porous medium (NG et al., 2006). Stage one of dehydration is the transient period (Figure 4), *i.e.* the length of time required for the medium to reach the circulating air temperature. This is dependent on the rate of heat transfer from the air to the medium. Stage two is the constant rate period: as the medium reaches the heated temperature, water starts to evaporate and a steady state is reached with a constant rate of evaporation. The surface of the medium behaves like the wet-bulb temperature – the temperature varies with changes in the relative humidity of the surrounding air. The dehydration during stages one and two removes unbound water. Stage three is the falling rate period: the dehydration rate decreases linearly since all the unbound liquid has been removed and the bound liquid is more difficult to remove. As the medium continues to dehydrate, the surface area of evaporation decreases, thus decreasing the dehydration rate. The final stage (four) is the second falling rate period in which the dehydration falls at a second-order rate as the easily accessible liquid is depleted.

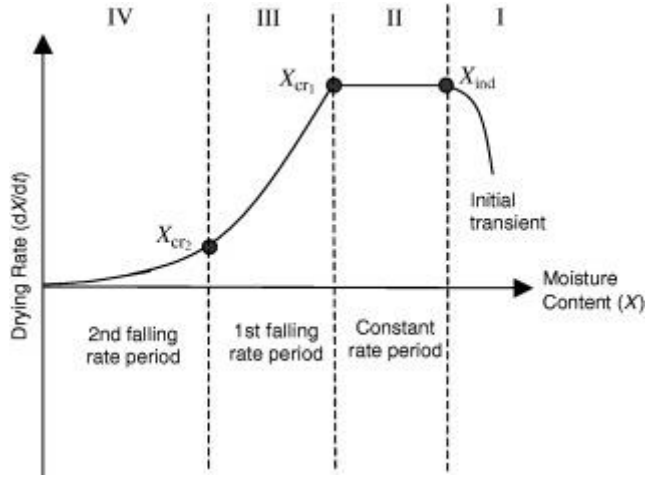

Figure 4. The four stages of the drying curve going from right to left (image from NG et al. (2006))

3.5 Pathogens in human urine

Urine in the bladder of a healthy person is typically pathogen free (Willey et al., 2009), however during excretion and the collection of the urine in the toilet bowl, cross-contamination from faeces occurs, even in urine-diverting toilets (Höglund et al., 2000). Diseases that can be emitted *via* the urine are considered insignificant due to the limited risk: transmission of a disease in the environment is low (such as *Salmonella typhi*, *Salmonella paratyphi*, *leptospira* and some viruses) or the disease is isolated to specific areas (such as schistosomiasis and *Mycobacterium tuberculosis*) (Feachem et al., 1983, WHO, 2006, Peter et al., 2010). Cross-contamination from faeces does present a risk, assessed by faecal sterols to an average rate of 9.1 mg of faeces per litre of urine (Schönning et al., 2002, WHO, 2006). One gram of faeces can contain 10^9 infectious virus particles without the human host necessarily exhibiting clinical signs (Feachem et al., 1983). As faeces potentially contain pathogenic viruses, bacteria, protozoa and helminths (WHO, 2006), the risk of cross-contamination of faeces into the urine needs to be considered.

4 Materials and methods

4.1 Urine and faeces collection (Papers I-III)

Human urine and faeces (Paper I-III) were collected anonymously in sterile containers from between five to ten people aged between 24 and 65. The urine and faeces were stored in the collection containers at 3 and -18 °C respectively until use.

4.2 Stabilisation and dehydration (Paper I-II)

Urine stabilisation and dehydration were assessed at various temperatures between 35 and 65 °C (Table 3). Three dehydration media were used: ash from wood pellets (Paper I), ash from birch trees (Papers I-II) and biochar from chopped willow trees (Paper II). Wood ash had a high initial pH (>12) whereas the pH of biochar (initially < 7) was increased to >12.5 with KOH. The dehydration trials were performed in incubators with two DC 12V computer fans for ventilation (total air flow of 750 L min^{-1}). In Paper II, for three of the trials (Table 3), the urine was pre-treated with an anion exchanger, Amberlite™ IRA410 type-2 resin, to increase the pH of urine to ≥ 11.5 before being added to the medium to assess whether this could help maintain an elevated pH in the dehydration media.

In the 65 °C trial (Paper I), the N concentration in the ash treatments was measured three times (on days 20, 33 and 41). The ash was first incubated with urease enzymes to convert the urea to NH_4^+, and then N was measured as NH_3-N by an ammonia electrode probe (Metrohm AG, Switzerland). In the other trials (Papers I-II), the N concentration was measured as total N dry combustion (LECO TruMac® CN, USA) at the end of the experiment. The initial and final P and K concentrations were analysed as phosphate-P (PO_4-P),

and K concentrations using ICP (ICP Optima 7300 DV Swedish Standard: SS 02 83 11).

Table 3. Dehydration medium, temperatures and loading method and rate for Papers I-II. Static loading had all the dehydration medium added at the beginning. Dose loading had the medium added with each urine application at 5 % (ash/urine, w/w)

Medium	Pre-Treatment	Temp. (°C)	Loading method
Birch ash	-	35	Static
Birch ash	-	35	Dose
Birch ash	Ion-exchange	40	Static
Birch ash	Ion-exchange	50	Static
Birch ash	-	65	Static
Birch ash	-	65	Dose
Wood pellet ash	-	65	Static
Wood pellet ash	-	65	Dose
Biochar	Ion-exchange	45	Static

In Paper II, a response surface methodology (RSM) was followed to optimise the urine dehydration rate in wood ash. A four factorial-five level central composite rotary design (CCRD) was set up with the independent variables of incubator temperature (38-42 °C), air flow rate (0.5-1.5 L min^{-1}), ash loading (100-200 g) and urine dosing (40-80 mL per addition till > 90 % is evaporated), with the response variable being the dehydration rate, *i.e.* the volume of water dehydrated per day in one square metre of ash (target was 90 % water removal). Design Expert software (V.7, Stat–Ease Inc., USA) formulated 30 experiments that varied the independent variables. The 30 experiments were run and the data collected were regressed against the third-order polynomial equation to identify the optimal variable values for maximised dehydration.

4.3 Calculating urea loss (Papers I-II)

During the dehydration process it was expected that some urea would be lost due to degradation influenced by the elevated pH and temperature. To estimate the theoretical amount of urea contained in the saturated medium at the end of the dehydration process, Equation 4 was derived.

$$Y_i = X_i + Y_{i-1} - \frac{X_i + Y_{i-1}}{2 * h_i} \qquad \text{(Eq. 4)}$$

where Y_i is the remaining amount of urea (g) on day i, X_i is the amount of urea (g) added on day i, Y_{i-1} is the amount of urea (g) that remains from the day before. The last term takes the amount of urea currently in the medium and calculates the amount that will be lost on day i based on the half-life of urea (h_i) under the conditions on day i (h_i values found in Table 1). This equation differs from Equation 3 in Paper I as here it is calculated as the urea remaining in the dehydrating medium, whereas the equation in Paper I calculated urea as a loss.

4.4 Hygiene assessment (Paper III)

In the saturated ash (final pH 10.5), five microorganisms were studied during storage. To isolate the effect of temperature and pH on the microorganisms, four controls were used (Table 4): two temperatures (20 and 42 °C), pH (10.5) and stored urine. Of the organisms studied, three were indicators – *Enterococcus faecalis* (ATCC 29212), MS2 ATCC15597-B1 bacteriophage, and ΦX 174 ATCC13706-B1 bacteriophage – and two were pathogens – *Ascaris suum* and *Salmonella enterica* subspecies *enterica* serovar Typhimurium (hereafter referred to as *S*. Typhimurium).

Table 4. Treatments studied for inactivation of five microorganisms in saturated ash with four controls: temperature (20 and 42 °C), stored urine and pH (10.5)

Treatment	Temp.	Microorganism
Saturated ash	20 °C	*A. suum*, *S*. Typhimurium, *E. faecalis*, MS2, ϕx174
	42 °C	*A. suum*
Temperature	20 °C	*A. suum*, *S*. Typhimurium, *E. faecalis*, MS2, ϕx174
	42 °C	*A. suum*
Stored urine	20 °C	*A. suum*, *S*. Typhimurium, *E. faecalis*, MS2, ϕx174
pH control	20 °C	*A. suum*, *S*. Typhimurium, *E. faecalis*, MS2, ϕx174

4.4.1 Ascaris extraction and data analyses

To prepare the saturated ash treatment, 10 ml of fresh urine was mixed with 0.1 g of the inoculated faeces (high contamination rate to ensure enough eggs were present). The saturated ash samples were left open to the air to simulate ash stockpiling until its use as a fertiliser (once or twice a year). For the control, faeces (0.1 g) inoculated with *A. suum* was mixed with 5 ml of either: M/15 phosphate buffer solution (pH 7.2) as the temperature control; glycine/NaOH buffer solution (pH 10.5) as the pH control; or freshly collected (not yet hydrolysed) urine as the stored urine control. All the controls were sealed.

The extraction procedure for the *A. suum* eggs was a modification of the USEPA (2003) method. Magnesium sulphate ($MgSO_4$, specific gravity 1.20) was used to float the eggs and Tyler sieves (⌀ 38 µm, Cat. No. L3-400 and ⌀ 300 µm, Cat. No. L3-50) were used to filter the eggs out. After extraction of the eggs, a subsample of 100 eggs was observed under the microscope to assess the condition of the eggs directly after extraction. The remaining eggs were incubated at 28 °C in 0.1 N H_2SO_4 for 28 days to enhance larval development.

The data were normalised according to the initial viability and the zero value adjusted to the number of eggs counted that were viable. The *A. suum* egg viability data was then fitted against a non-linear inactivation model (Eq. 5) from Harm (1980) and variable values found by statistical analysis using Minitab 17 (Minitab Inc., US):

$$N = N_0\left[1 - (1 - 10^{-k*t})^{10^n}\right] \qquad \text{(Eq. 5)}$$

where N_o is the initial viability of the eggs at time 0 and N is the predicted viability of the eggs at time t (in days). The parameter determining lag phase duration (n) and the inactivation rate (k) were calculated by fitting the model to the data by non-linear regression using the Gauss-Newton algorithm (Minitab 17, Minitab Inc., US). The lag phase (l, Eq. 6) is the initial period with no significant inactivation.

$$Lag\ phase = l = \frac{n}{k} \qquad \text{(Eq. 6)}$$

4.4.2 Bacteria and viruses

The bacteria (*Salmonella* and *Enterococci*) were cultivated in nutrient broth and concentrated by centrifuging. The bacteriophages were propagated according to standard ISO10705-1 (1995). The bacterial host strains S. Typhimurium WG49 (ATCC 700730) and *E. coli* ATCC 1370 were used for the enumeration of bacteriophages MS2 and ΦX174 respectively. Five millilitres of the stock solution of microorganisms (*S.* Typhimurium, *E. faecalis*, MS2 bacteriophage and ΦX bacteriophage) were added to 5 g of the saturated ash and the containers left open to the air at 20 °C without further mixing (as there is no mixing in the system). For the temperature, pH and stored urine control, the same solutions were used as described in Section 4.4.1. The bacteria were sampled on days 1-4 and 14, and the viruses were sampled on days 1-4, 8 and 15. All were enumerated as described in Paper III.

5 Results

5.1 Dehydration (Papers I-II)

With the urea stabilised by high pH (method depending on the treatment applied) and the surface of urine broken by being absorbed into ash or biochar, the excess liquid in urine was removed by aeration. The lower the dehydration temperature, the greater the N retention (Table 5).

Table 5. Loading rate, ratio of urine to dehydration medium (ww/dw) and % N retained. Static loading had all the dehydration medium added at the beginning. Dose loading had the medium added with each urine application at 5 % (ash/urine)

Medium	Temp. (°C)	Loading method	Loading rate (L m^{-2} d^{-1})	Ratio[z]	N retained
Birch ash	35	Static	3.8	8	90 %
Birch ash	35	Dose	3.8	20	82 %
Birch ash	40	Static	7.3	10	76 %[y]
Birch ash	50	Static	12.2	16	74 %[y]
Birch ash	65	Static	5.1	16	64 %
Birch ash	65	Dose	5.1	20	66 %
Wood pellet ash	65	Static	5.1	16	64 %
Wood pellet ash	65	Dose	5.1	20	66 %
Biochar	45	Static	6.4	11	72 %[y]

[z] Urine:medium

[y] Urine was pre-treated with anion exchanger and the 5-10 % N lost due to this process is not included in this N retained

At 35 °C (Paper I), the static bed retained the most N at 90 % of the N added with urine. At 65 °C, N retention was the lowest at 64 %. In Paper II, the dehydration conditions were optimised and a dehydration rate of

6.68 L day^{-1} m^{-2} was achieved with N retention peaking at 77 %. Dehydrating urine in the alkaline medium achieved a dry powdery (5-7 % moisture) end product while retaining the majority of N. Figure 5 gives an example of the measured mass balance of the dehydration process. The excess water in the urine was evaporated, reducing the mass by 92 % (including the mass of medium). The N loss shown in the diagram is included in the total volatile solids (VS) lost.

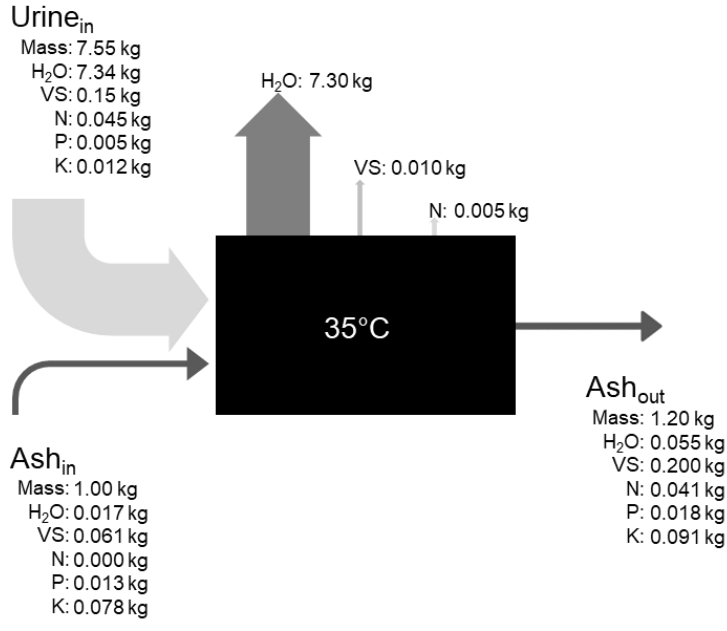

Figure 5. Measured mass balance for dehydrating 20 L of urine in 1 kg of ash at 35 °C for the static dehydration bed where all the dehydrating medium was added at the beginning and urine was added at regular intervals.

5.1.1 Anion Exchange

Different portion of urine was mixed with the anion exchanger (Figure 6) to assess how much of the urine needed to pass the anion exchanger to reach a pH > 10. The pH varied from 8.1 (20 % mixed) to 11.2 (100 % mixed) and 60 % of all urine would need to pass though the anion exchanger to reach a pH of 10.

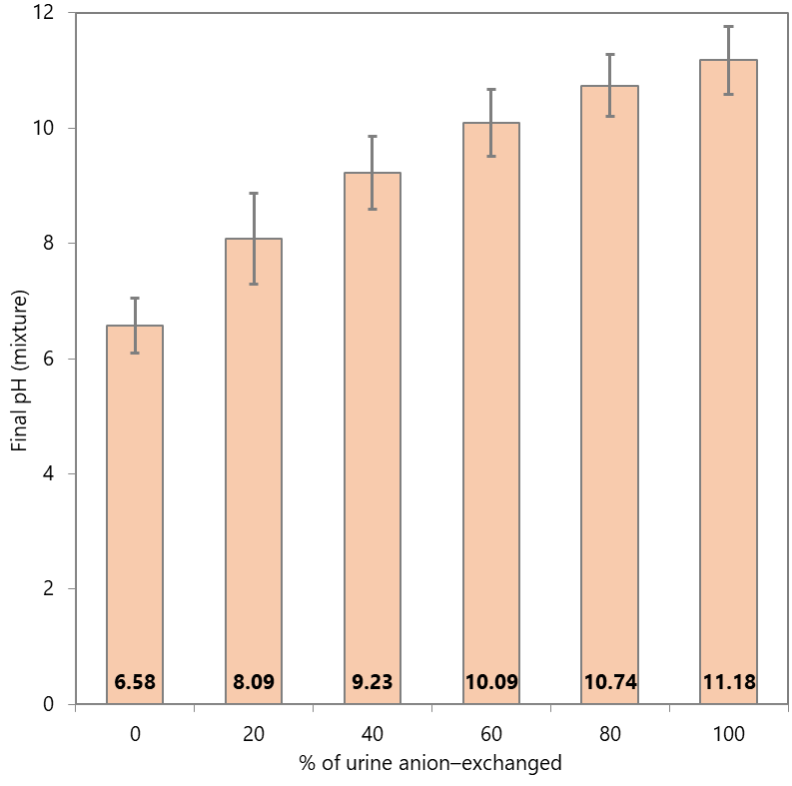

Figure 6. The final pH of urine after a portion (x-axis) was mixed with the anion exchanger. Values in bold specify average (n = 50) pH and y–axis error bars represent standard deviation of the average.

5.1.2 Optimisation (Paper II)

The loading rate increased from 3.8 L day^{-1} m^{-2} at 35 °C or 5.1 L day^{-1} m^{-2} at 65 °C (Paper I) to as high as 12.2 L $day^{-1}m^{-2}$ at 42 °C (Paper II). The optimal operating conditions, based on the response surface methodology and a surface area of 0.03 m^2, were a temperature of 41 °C, an air flow rate of 0.5 L min^{-1}, initial wood ash loading of 100 g and 80 mL of urine at a time (time till 90 % of the water is removed, then more urine would be added). This achieved a dehydration rate of 6.7 L $day^{-1}m^{-2}$ at 41 °C. The relationship seen in Figure 7 shows that a lower initial loading of ash with a higher loading rate of urine yields better dehydration rates. These dehydration conditions were then used to produce the saturated medium for the hygiene assessment (Paper III).

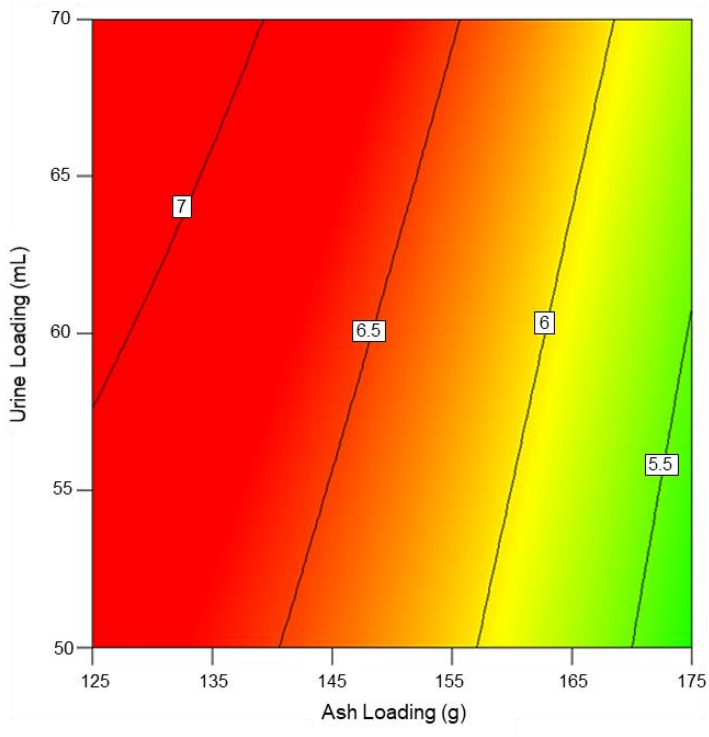

Figure 7. The relationship between urine dosing (mL) and ash loading (g) on the urine dehydration rate with the lines indicating average rates (L day^{-1} m^{-2}).

5.1.3 Nitrogen retention

The process concentrated the nutrients into a dry NPK fertiliser with concentrations as high as 7.8, 2.5 and 10.9 % of N, P and K respectively (dry weight). Paper II optimised the dehydration rate, which decreased the time the urine was exposed to heat, yet there were still high N losses. As the fresh urine was mixed with the anion exchanger to increase the pH, the NH_3 initially present (~ 6 % of N) was lost by volatilisation due to the elevated pH. Based on the Tot-N concentration after the urine passed through the anion exchanger, the majority of this N was retained: 76.5 % in ash at 40 °C, 73.8 % in ash at 50 °C and 71.8 % in biochar/KOH at 45 °C.

It was assumed that there would be some N lost due to the initial high pH of the media (> 12.8) and the elevated temperatures (40-50 °C). Based on the formula and the half-life of urea derived in Section 4.3, only ≤ 0.5 % would have theoretically been lost because the urine was not exposed to the elevated

pH (> 12) or temperatures (≥ 40 °C) for long (all treatments ≤ 8 days). However there was 24-28 % N loss (theories as to why are discussed in Section 6.1.3). There was complete recovery of P and K within the media since these elements are non-volatile at the investigated temperatures.

5.2 Hygiene assessment (Paper III)

The *A. suum* eggs in the saturated ash treatment at 20 °C (pH=10.5) were dead at the last sampling on day 346 (Figure 8). Fitting the data to the model (Eq. 5) gave a lag phase of 72 days and 3 $\log_{10}$ reduction of 325 days (Paper III, Table 2). The moisture content decreased from 43 % to 1 % by day 102. The pre-check at sampling (prior to incubation) showed that in the saturated ash treatment, eggs had started to develop during the treatment after 46 days and 7 % (9 out of 124) developed to larval stage after 74 days. At the last sampling (day 346), 9 % of the eggs had developed into larvae during the treatment, however all the larvae were dead and none developed during incubation.

At 42 °C, *A. suum* in the saturated ash treatment (open) had a short lag phase of 1.4 days and a 2 log inactivation after 9 days (Figure 9). The last sample extracted (day 14, data not shown) from the saturated ash treatment had disintegrated with only 436 eggs found (out of >2000 initially added). During incubation, 1.3 % developed into larvae during incubation.

The bacteria and virus indicators in the saturated ash were below the detection limit within four days. *E. faecalis* was inactivated and could no longer be detected at day 3 in the saturated ash treatment (TS: 61.1 %; electric conductivity$_{1:5}$ 35 dS m^{-1}; ionic strength: 0.55 M at 20 °C) (Paper III, Figure 3). *S.* Typhimurium was inactivated most quickly in the saturated ash treatment (Table 6), with a > 6 log reduction during day 1 and complete inactivation during day 2 (Paper III, Figure 3). For *S.* Typhimurium, the sampling from the saturated ash treatment was at too low a frequency to have sufficient data points for calculating the standard error (SE) of the reduction rate. The MS2 phage was inactivated rapidly in the saturated ash treatment, with 7.7 log reduction (detection limit) by day 4 (Paper III, Figure 3). The ϕX174 phage was also inactivated rapidly in the saturated ash treatment, with 5.3 log reduction (detection limit) at day 3 (Paper III, Figure 3).

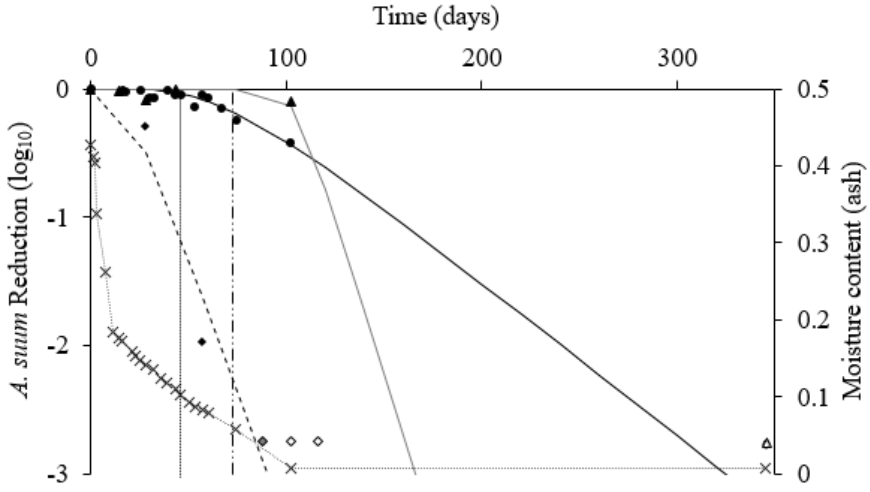

Figure 8. A. suum reduction at 20 °C in saturated ash (● / –), stored urine (◆ / - - -) and pH control of 10.5 (▲ / –) with lines modelling the inactivation. The vertical black dotted line indicates when the eggs started to develop in the treatment and the vertical black dashed line indicates when fully developed larvae were found in the treatment. The right vertical axis is the moisture content (×) of the ash samples throughout the treatment. Grey filled symbols indicate that the detection limit was reached and white filled symbols indicate data points not used in the model.

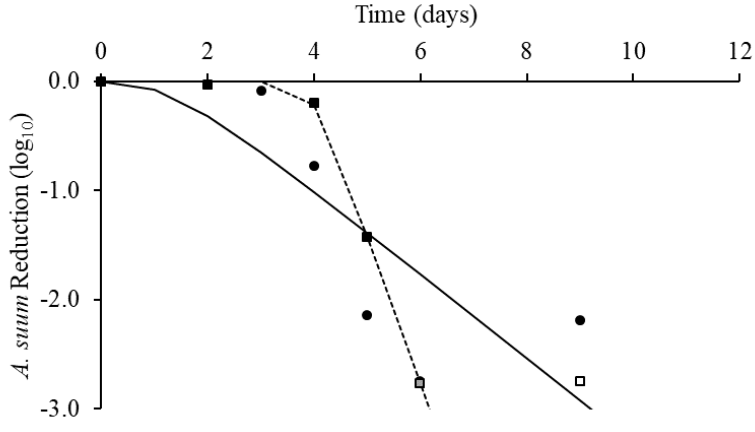

Figure 9. A. suum egg inactivation data (symbols) and inactivation model (lines) at 42 °C in saturated ash (● / –) and the temperature control (■ / - - -). Grey filled symbols indicate samples where the detection limit was reached and the white filled symbol indicates that the data point was not used in the model.

Table 6. The inactivation rate constants (k), standard error in brackets, decimal reduction time t_{90} (days) and calculated treatment time required for a 6 $\log_{10}$ reduction. Asterisk (*) indicates significant inactivation ($p < 0.05$)

				Reduction (days)	
	k (d^{-1})			t_{90}	6 $\log_{10}$
E. *faecalis*					
Saturated ash	-1.89	*	(0.24)	<1	3
Stored urine	-0.04		(0.04)	26	155
pH 10.5 control	-0.09		(0.05)	11	66
Control (T = 20 °C)	-0.03		(0.04)	33	199
***S.* Typhimurium**					
Saturated ash	-6.19	*	-[z]	<1	1
Stored urine	-0.11	*	(0.03)	9	53
pH 10.5 control	-0.40	*	(0.20)	2	15
Control (T = 20 °C)	-0.03		(0.03)	37	225
MS2					
Saturated ash	-1.79	*	(0.47)	<1	3
Stored urine	-1.04	*	(0.26)	1	6
pH 10.5 control	-0.81	*	(0.24)	1	7
Control (T = 20 °C)	-0.03		(0.01)	39	235
ϕX174					
Saturated ash	-2.09	*	(0.10)	<1	3
Stored urine	-0.03		(0.01)	29	175
pH 10.5 control	-0.07		(0.02)	14	85
Control (T = 20 °C)	-0.03		(0.02)	29	173

[z] The die-off was so quick that not enough measurements were made before the *S.* Typhimurium was below the detection limit to calculate the standard error, which also means that the significance of the k_d value could not be tested.

6 Discussion

6.1 Nitrogen retention

6.1.1 Fertiliser potential

The end product of the dehydration process was urine-based fertiliser with an NPK value of 7.4-2.3-10.3 (ww) and 5 % moisture content (Paper I). NH_3 stripping (another technology for concentrating N in from urine) results in a 10 % NH_3 solution, however the initial volume of liquid remains to be treated and disposed of (Maurer et al., 2006). The concentration of nutrients was higher than that found in cattle or chicken manure (ww) (Penhallegon, 2003) and similar to some mineral fertilisers, such as combined potassium nitrate and ground rock phosphate (FAO, 1991). This end product has a potential monetary value of approximately US$ 115 tonne^{-1} on the global commodity market based on the nutrient content, compared with the December 2016 price of urea (US$ 216 tonne^{-1}; 46 % N), triple superphosphate (US$ 267 tonne^{-1}; 20 % P) and potash (US$ 215 tonne^{-1}; 52 % K) (World Bank, 2017).

A benefit to using wood ash and biochar is their content of P and K, further enriching the urine as a complete fertiliser. In this dried product the specific forms in which P and K are retained were not investigated in this study. P from the urine was likely to occur mainly in the form of precipitated metal phosphates such as struvite ($MgNH_4PO_4 \cdot 6H_2O$) and hydroxyapatite (HAP, $Ca_{10}(PO_4)_6(OH)_2$) due to the high pH and the added Mg and Ca from the ash (Udert et al., 2003). The K from the urine can be assumed mainly to remain its original salt forms, such as KCl, K_2SO_4, $KHCO_3$ and K_3PO_4 (Putnam, 1971). Heavy metals found in human urine are far below the Swedish limit for land application (Jönsson et al., 1997), but the concentrations in different wood ash sources needs to be considered.

The salinity concentration of the end product will also need to be considered for this technology and urine as a fertiliser in general. Salts present, such as sodium (Na < 9 % dw (Putnam, 1971)), could increase the electric conductivity of the soil, which may negatively impact plant growth (Power and Prasad, 1997). The chloride (Cl^- <17 % dw (Putnam, 1971)) added to the soil could increase the cadmium (Cd) uptake in plants to levels above the WHO recommendations because Cl^- mobilises Cd in soils (Dahlin et al., 2016). As the dehydration media become efficient and more urine is concentrated into the same medium – the limitation of the system might be the salinity concentration. The use of an ion exchanger for removing part of the Cl^- and other salts could be used to reduce this risk.

6.1.2 Anion exchanger

The rationale of using the anion exchanger (resin) is that it can increase the pH of the urine within the toilet, which would enable pipe transport with minimal risk of enzymatic hydrolysing of urea. The resin (Paper II) functioned to increase the initial pH of urine (< 7) to > 10, which is high enough to keep the urease enzyme inactive. However to have the urine anion-exchanged did not result in a higher pH over time in the dehydration media (Paper II). As seen in Table 5, the ratio of urine to dehydration media was not increased with the addition of the anion-exchanged urine – further analysis are required to understand why this had little to no effect.

The factor influencing the capacity of the anion exchanger is the initial concentration of the anions in the urine. The concentration of anions in human urine can vary greatly on a daily basis, for example Cl^- has been measured to vary between 750 and 11 456 mg/L (Ferslew et al., 2001), making it difficult to predict what pH the urine will reach after passing the anion exchanger. Differences in the initial anion concentration in urine are why different amounts of anion exchanger were required to raise the pH to >10 during the experiment (Paper II).

An additional benefit to using the anion exchanger is the removal of salts, especially Cl^- as this has been shown to mobilise Cd in soils (Dahlin et al., 2016). The concentration of Cl^- in pre-trials decreased by 45-55 % (data not shown). The downside was that P also decreased by 35-70 % (a portion of this loss was probably due to precipitation of P where the visible solids remained within the resin matrix). However it is possible to recover the P at a later stage, *e.g.* by acid washing.

The practical application of using the tested anion exchanger (Amberlite resin) to increase the pH of urine was less functional than expected. The mass

of resin required in a toilet to stabilise the urea in urine before pipe transport is considerable: 200 ml for every litre of urine. The resin would need to be frequently changed or regenerated. One person can urinate 1 to 3 L day^{-1}, meaning 5.6 to 17 L of resin would be needed per week for a family of four. To the best of our knowledge, anion–exchange resins have not previously been used alkalise human urine, but cation-exchangers have been tested to capture the NH_4^+ in hydrolysed urine (Tarpeh et al., 2017). In Tarpeh et al. (2017), the volume of resin needed on a weekly bases ranged from 5.3 to 19 L.

With all the complications that occurred with using the anion exchanger, *i.e.* high consumption of resin, blockages due to precipitation, no lasting alkalising effect in the medium and risk of bacterial growth in the resin, it was concluded that it would not be convenient to have an anion exchanger in a household or public toilet for stabilising the urine. However, the removal of Cl^- may be advantageous from a fertiliser point of view.

6.1.3 Nitrogen loss

The elevated pH stabilised the urea in the urine during dehydration. This is supported by Geinzer (2017), who showed that urease enzymes are inactivated at pH > 10. However N losses were still experienced during the dehydration process (Table 5). Degradation due to the elevated pH should have been minimal (Table 5) as the pH decreased to <12 within the first 12 days of the experiment. In Paper I, thermal degradation was the main source of N loss. The dehydration medium exposed to an elevated temperature (65 °C) for 42 days lost 35 % of N (however Eq. 4 predicted >50 % loss). In Paper II, the duration of the trial was shorter and dehydration rate was optimised and the urine/ash mix exposed to 40-50 °C for < 8 days. It was modelled to lose <0.5 % N during this time, yet the system still lost 24-28 % N (this is the amount lost after the anion exchanger, which was approximately a 6 % loss from the initial ammonia excreted in the urine).

The differences in losses could have been due to localised pockets within the media that had a lower pH (< 10), especially towards the end of the experiment. These localised pockets might have had more of an impact in the experiments of Paper II as the trials were bigger (Paper I used max 50 g ash while Paper II used max 175 g ash). Another explanation for the larger N losses in Paper II might be that the urine and ash were dried in sieves (250 µm mesh) and it was observed that when the urine was applied to the sieve, the urine would flow through to the bottom and dry on the mesh (the surface tension was strong enough that the urine did not drip through). As a result, the urea could have dried on the mesh and not been recovered at the end of the

experiment. N losses may have also been elevated where the urine was in direct contact with the sieve. The differences in the modelled and measured values is important to understand for using the dehydration system with varying parameters.

The N retention during dehydration was similar or higher (64-89.9%) than in other mass-reducing urine treatments such as solar drying (68%; (Antonini et al., 2012)); struvite and nitrification (actual N recovery rate not specified due to N losses during storage) (Udert et al., 2015); NH_3 adsorption on zeolite and wollastonite (65-80%; (Lind et al., 2000)) and evaporation on gauze sheeting with high salt concentrations (85%; (Pahore et al., 2011)). The alkaline treatment retained N and had no liquid disposal.

6.2 Liquid reduction by dehydration

The dehydration process had an overall mass reduction of 92 % (including the medium used for hydration), which along with the N retention is a novel achievement. The ratio of urine to dehydration medium ranged from 8:1 to 20:1. The volume reduction of urine achieved in this system for the urine 95 % and 75 – 90 % including the dehydration medium (range depending on the ratio of medium used). This is similar or better than what has been achieved by many other urine treatment technologies which still have liquid fractions to manage (% = volume reduction of liquid): vapour-compression evaporation (90 %), freeze-thaw (75 %), electrodialysis (35 %), struvite precipitation (0 %), ion-exchange for N removal (0 %), acidification (0 %), NH_3 stripping (0 %), partial nitrification (0 %), (Maurer et al., 2006).

The saturation of the ash bed was not limited by the mass of urine added, but rather by the mass of carbon dioxide absorbed from the ventilation. As seen from the controls (Paper I, Figure 1) where only water was added, the pH decreased at the same rate as in the urine treatments. A plausible explanation for the drop in pH over time is that the urine–filled medium absorbs the CO_2 from the air passing over. The dissolved CO_2 results in an increase in H+ concentration (Paper I-II). The CO_2 from the incoming air could be captured by a lime filter to prolong the life of the medium (optimising the urine:medium ratio is further discussed in Section 6.4).

Temperature was the driving force for mass transfer of steam from the ash-urine mixture to the ambient air. Increasing the temperature allows for more rapid removal of moisture from the surface, which decreases the dehydration time (Menzies and O'Callaghan, 1971). The optimisation model experimented with a narrow temperature rage (38-42 °C) however a higher dehydration rate was achieved at 50 °C with minimal N losses – the model could be extend to

higher temperatures, with lower initial medium loading and higher urine dosing. High initial relative humidity (RH) and high inlet temperature might diminish the ratio. However, with increased temperature, the water-holding capacity of the air increases exponentially, *e.g.* from 40 g/kg dry air at 35 °C to 200 g/kg dry air at 65 °C. Warming the inlet air, either by solar or other means, would help greatly with dehydration (optimising the dehydration rate is further discussed in Section 6.4).

6.3 Hygiene assessment

The results in Paper III support previous work that has also seen rapid inactivation of indicator bacteria (Nyberg et al., 2011) and viruses (Decrey et al., 2016) in alkaline medium. To reach a 6 $\log_{10}$ reduction for the bacteria and viruses, the saturated ash would only need to be stored for four days at 20 °C. For the re-use guideline standards, such as WHO (2006), that specify a maximum concentration in the end product, diverting urine away from faeces is actually an effective 'treatment' for *A. suum*. For example, with a high value of 735 eggs g^{-1} TS of faecal sludge (Jiménez, 2006) and daily excretion of 200 g faeces and 1.5 L urine a day (Vinnerås et al., 2006) with an average cross-contamination of 9.1 mg of faeces L^{-1} of urine (Schönning et al., 2002), there is already a 5.2 $\log_{10}$ reduction (about 1 egg L^{-1} or 0.001 eggs g^{-1}) just from urine diversion. In the present system, where 1.5 L of urine is concentrated into 30 g of ash, the final concentration would be 0.05 eggs g^{-1} ash or an overall $\log_{10}$ reduction of 3.5. This would meet the WHO (2006) (< 1 egg in 1 g^{-1} TS) and USEPA (1994) (< 1 egg in 4 g^{-1} TS) standards for *A. suum* without further treatment being required.

Bacteria and viruses would also be reduced by 3.5 $\log_{10}$ from urine being diverted away from faeces, but depending on their initial concentration this may not be sufficient to meet the WHO (2006) and USEPA (1994) guideline of <1000 *E. coli* or faecal coliforms per g of TS. However after only four days of storage, the indicator bacteria and viruses in the saturated ash reached a 6 $\log_{10}$ reduction, which with the dilution adding a further 3.5 $\log_{10}$ reduction would, based on bacterial and viral numbers found in faeces around the world (Feachem et al., 1983), be more than sufficient to meet the guidelines.

One advantage of using high pH for inhibiting urease activity is the long-term effect: the remaining elevated pH would continue to inhibit contamination or regrowth of pathogens, including the inhibition of urease-producing bacteria (Nyberg et al., 2011). In other treatments, such as oxidation, that have good initial inhibition but no protection against regrowth of urease-producing bacteria or pathogens, post-oxidation leads to hydrolysis

of urea and/or increased risk of disease transmission (Zhang et al., 2013). Using the alkaline dehydration system enables the concentration of the nutrients in urine and eliminates the need for liquid disposal and the end product is a safe dry fertiliser after just four days of storage. During the whole storage (until used as fertiliser), the saturated ash can be kept open to the air as the medium continues to dehydrate (and not absorb moisture from the air (Figure 8)). This is much simpler than storing the liquid urine for six month in a sealed container (the recommendation for stored liquid urine) that is 20 times the size of the saturated ash container.

6.4 System development

The energy required to dehydrate the liquid in the urine is estimated to be 2600 MJ m^{-3} but could be decreased to < 400 MJ m^{-3} by using a vapour-compression evaporation that recovers the heat (Maurer et al., 2006). Vapour-compression evaporation is quite complex. A simpler approach could be to use a dehumidifier. The dehydration would be performed in a closed gas loop with a heat pump. The heated air would pass over the dehydration medium and then would be cooled to remove the water vapour by condensation, the same as what a dehumidifier does. The energy requirements could be minimised with a heat exchanger and/or using solar radiation during the day. Such a system could decrease the energy required as the evaporation energy could be recovered when the water is condensed from the hot gas. The closed gas loop would have no new CO_2 would be entering the system and could decreases the risk of odours spreading from the drying (thus far not a noticeable issue). Condensing the water vapours could also capture any ammonia formation, which would greatly decrease the acidifying and eutrofying emissions from the system. The condensed water would need to be treat, *e.g.* with the greywater. The energy requirement would need to be assessed. In such a system, the pH may no longer be the limiting factor of the dehydration medium, however the salt content might become more of an issue as ever more urine is concentrated.

The dehydration rate could be further improved by optimisation of the dehydration media. The dehydration process could be evaluated using a non-porous medium, such as sand alkalised with KOH. All liquid would then be freestanding, which should increase the dehydration rate, however more medium would be required to prevent pooling and oils forming on the surface.

6.5 Implementation potential

The evaluated urine dehydration system is flexible enough to be added to existing sanitation systems. The installation is not complex and no extra piping is required, but a service provider would be needed to transport and process the end product. The system is flexible to expand as a city is growing – one does not need to wait for a sewage network to have a hygienic sanitation system, instead the treatment is on-site and supports the polluter pays principle.

Urine-diverting toilets collect urine separately from faeces during excretion and such toilets are already available on the market as urine-diverting *flush toilets* or urine-diverting *dry toilets*. In the present study's system, either type of these user interfaces could be used. Following excretion, urine would be diverted into a containment unit located close to the seat, within the toilet room. The function of the containment unit is based on the principle of alkaline dehydration whereby the mass of urine is reduced by 95 % and nutrients are simultaneously retained in an alkaline dehydration medium.

Once the dehydrating medium is saturated (pH < 10.5), the unit (potentially a plastic bag or box) would be removed and enter the recycling system in a similar way that household organic waste is removed and entered into a recycling system. A service provider could operate this system in a similar way as for municipal solid waste collection. The quantity of the saturated medium per person per year would be approximately 40 kg, which is minimal compared to municipal solid waste production, 480 kg $person^{-1}yr^{-1}$, (Hoornweg and Bhada–Tata, 2012) and the same size as the food waste collected in Swedish municipalities with separate food waste collection (Avfall Sverige, 2017).

Faecal management was not the focus of this thesis, however its management should briefly be considered. In the urine-diverting flush toilet, faeces along with flush water could be transported by pipes for treatment at the wastewater treatment plant. The wiping material and/or anal cleansing water would be added to the faeces through the same user interface. In the urine-diverting dry toilet, faecal dehydration in another containment unit beside the urine dehydration unit could be used (Tilley et al., 2014). Diverting urine and faeces away from the wastewater treatment plant could greatly reduce the complexity and costs of operation since approximately 88 % N and 67 % P would already be removed (Jönsson et al., 2005). The saturated medium from the urine dehydration (and dried faeces) process would be taken to a treatment and conversion plant. The saturated medium (urine) would then be pelletised and packaged to be sold as a fertiliser that is easy to transport and store until use.

7 Conclusions

- The alkaline dehydration system enables the concentration of nutrients in urine, eliminates the need for liquid disposal and produces a safe dry fertiliser within four days of storage at 20 °C after the last use.

- For the variants of media and dehydration conditions investigated, 95 % of the liquid was reduced by volume. Including the ash, the volume reduction ranged from 75 to 90 % depending on the ratio of dehydration medium used.

- N retention ranged from 64 to 90 % and complete recovery of P and K were achieved.

- Increasing the urine dosing while decreasing the initial medium load yielded higher dehydration rates. The optimal conditions found for dehydration were an initial medium load of 100 g ash and loading urine of 80 ml (repeated after >90% would be evaporated) at 41 °C with an air flow rate of 0.5 L min^{-1}.

- Dehydrating urine at higher temperatures (35 to 65 °C) offers flexibility to either increase capacity or decrease the required surface area of ash.

- Sustaining alkalinity is necessary to ensure the effectiveness of the process, hence the pH of the dehydration medium is what determines the time range within which dehydration can be performed.

- For guideline standards on re-use, such as from WHO (2006) that specify a maximum acceptable concentration in the end-product, urine diversion is actually an effective treatment in its own right for helminths (Ascaris).

- Installing such a dehydration system in new urine-diverting and existing dry toilets could greatly simplify transportation and fertiliser application since there would be no liquid disposal.

- The urine can be collected, contained, treated and reduced within the same collection container to produce a dry end product with a high N-P-K concentration (up to 7.8–2.5–10.9 % of N, P and K), *i.e.* a fertiliser with similar concentration as existing mineral fertiliser and thus have a potential monetary value of US$ 115 tonne^{-1}.

8 Future research

The performance of this system in terms of efficiency and hygiene safety should be assessed under the following conditions:

- cold (4 °C) and extremely cold (-20 °C) conditions
- on a larger scale
    - households of four to six people
    - public toilets with high peak loads
- confirmation of urease inactivation with faecal contamination in the dehydration medium
- assessment of the inactivation of microorganisms in saturated ash in a more humid environment (> 70 %) and at lower temperatures (< 20 °C)
- assessment of the inactivation of other pathogens, such as *Vibrio cholera* and *Cryptosporidium* oocysts

Further research and development to be explored:

- how much could the urine:medium ratio increase if the dehydration is done with closed gas loop, where the gas is first dried and then heated by a dehumidifier
- whether ion exchange resin can be used effectively
- whether there are other resins or chemicals that can increase the pH of urine efficiently to enable pipe transport
- energy requirements compared to other technologies
- user acceptance of this kind of toilet technology
- how faeces could be managed alongside this urine treatment technology
- fertiliser potential and possible effects of the fertilisers salinity (should the salinity be decreased with ion-exchanger?)
- how the dry fertiliser product can be turned into strong granules similar to present commercial mineral fertilisers.

References

AL-HUSSEINI, D. 2015. *Effects of Anions and Amino acids on Surface Tension of Water*. Senior Honors Projects, Paper 67, James Madison University.

ANTONINI, S., NGUYEN, P. T., ARNOLD, U., EICHERT, T. & CLEMENS, J. 2012. Solar thermal evaporation of human urine for nitrogen and phosphorus recovery in Vietnam. *Science of The Total Environment,* 414**,** 592-599.

AVFALL SVERIGE 2017. Hushållsavfall i siffror - Kommun- och länsstatistik 2016. Rapport 2017:26. Malmö, Sweden: Avfall Sveriges Utvecklingssatsning ISSN 1103-4092.

BAUM, R., LUH, J. & BARTRAM, J. 2013. Sanitation: A Global Estimate of Sewerage Connections without Treatment and the Resulting Impact on MDG Progress. *Environmental Science & Technology,* 47**,** 1994-2000.

BLAKELEY, R. L., TRESTON, A., ANDREWS, R. K. & ZERNER, B. 1982. Nickel(II)-promoted ethanolysis and hydrolysis of N-(2-pyridylmethyl)urea. A model for urease. *Journal of the American Chemical Society,* 104**,** 612-614.

CALLAHAN, B. P., YUAN, Y. & WOLFENDEN, R. 2005. The Burden Borne by Urease. *Journal of the American Chemical Society,* 127**,** 10828-10829.

CAMBERATO, J. J. 2011. Bioavailability of nitrogen. *In:* HUANG, P. M., LI, Y. & SUMMER, M. E. (eds.) *Handbook of Soil Science: Resource Management and Environmental Impacts.* 2 ed. Boca Raton, FL: CRC Press.

CIURLI, S., BENINI, S., RYPNIEWSKI, W. R., WILSON, K. S., MILETTI, S. & MANGANI, S. 1999. Structural properties of the nickel ions in urease: novel insights into the catalytic and inhibition mechanisms. *Coordination Chemistry Reviews,* 190–192**,** 331-355.

CIURLI, S., MARZADORI, C., BENINI, S., DEIANA, S. & GESSA, C. 1996. Urease from the soil bacterium Bacillus pasteurii: Immobilization on Ca-polygalacturonate. *Soil Biology and Biochemistry,* 28**,** 811-817.

DAHLIN, A. S., ERIKSSON, J., CAMPBELL, C. D. & ÖBORN, I. 2016. Soil amendment affects Cd uptake by wheat — are we underestimating the risks from chloride inputs? *Science of The Total Environment,* 554**,** 349-357.

DECREY, L., KAZAMA, S. & KOHN, T. 2016. Ammonia as an in–situ sanitizer: influence of virus genome type on inactivation. *Applied and Environmental Microbiology,* 82**,** 4909–4920.

ETTER, B., TILLEY, E., KHADKA, R. & UDERT, K. M. 2011. Low-cost struvite production using source-separated urine in Nepal. *Water Research,* 45**,** 852-862.

FAO 1991. Manual on fertilizer statistics. *Economic and social development paper.* Rome: Food and Agriculture Organization of the United Nations.
FAO 2015. World fertilizer trends and outlook to 2018. Rome: Food and Agriculture Organization of the United Nations.
FEACHEM, R., BRADLEY, D., GARELICK, H. & MARA, D. 1983. *Sanitation and disease - health aspects of excreta and wastewater management,* Chichester, UK, John Wiley and Sons.
FENG, D., WUB, Z. & XU, S. 2008. Nitrification of human urine for its stabilization and nutrient recycling. *Bioresource Technology,* 99**,** 6299-6304.
FERSLEW, K., HAGARDORN, A. & ROBERT, T. 2001. Capillary ion electrophoresis of endogenous anions and anionic adulterants in human urine. *Forensic Science,* 46**,** 615-626.
GEINZER, M. 2017. *Inactivation of the urease enzyme by heat and alkaline pH treatment: Retaining urea-nitrogen in urine for fertilizer use.* Masters Masters, Swedish University of Agricultural Sciences.
GOULDING, K. W. T. 2016. Soil acidification and the importance of liming agricultural soils with particular reference to the United Kingdom. *Soil Use and Management,* 32**,** 390-399.
HARM, W. 1980. *Biological Effects of Ultraviolet Radiation,* Cambridge, Cambridge University Press.
HELLSTRÖM, D., JOHANSSON, E. & GRENNBERG, K. 1999. Storage of human urine: acidification as a method to inhibit decomposition of urea. *Ecological Engineering,* 12**,** 253–269.
HÖGLUND, C. 2001. *Evaluation of microbial health risks associated with the reuse of source-separated human urine.* Doctoral, Royal Institute of Technology.
HÖGLUND, C., VINNERÅS, B., STENSTRÖM, T. A. & JÖNSSON, H. 2000. Variation of chemical and microbial parameters in collection and storage tanks for source separated human urine. *Journal of Environmental Science and Health, Part A,* 35**,** 1463-1475.
HOORNWEG, D. & BHADA–TATA, P. 2012. What a waste: a global review of solid waste management. Washington, D.C: The World Bank.
HOTTA, S. & FUNAMIZU, N. 2008. Inhibition factor of ammonification in stored urine with fecal contamination. *Water Science & Technology,* 58**,** 1187-1192.
HUANG, T.-C. & CHEN, D.-H. 1991. Kinetic study of urease-catalysed urea hydrolysis. *Journal of Chemical Technology & Biotechnology,* 52**,** 433-444.
ISO10705-1 1995. Water Quality detection and enumeration of bacteriophages. Part 1: enumeration of F-specific RNA bacteriophages. International Organization for Standardization. Geneva, Switzerland.
JESPERSEN, N. D. 1975. Thermochemical study of the hydrolysis of urea by urease. *Journal of the American Chemical Society,* 97**,** 1662-1667.
JIMÉNEZ, B. 2006. Helminth ova control in wastewater and sludge for agricultural reuse. *In:* GRABOW, W. (ed.) *Water Reuse New Paradigm towards Integrated Water Resources Management in Encyclopedia of Biological, Physiological and Health Sciences.* 1a ed.: EOLSS Publishers Co. Ltd UNESCO.
JÖNSSON, H., BAKY, A., JEPPSSON, U., HELLSTRÖM, D. & KÄRRMAN, E. 2005. Composition of urine, faeces, greywater and biowaste: for utilisation in the URWARE model. *The Mistra Programme: Urban Water.* Gothenburg, Sweden: Chalmers University of Technology.
JÖNSSON, H., STENSTRÖM, T.-A., SVENSSON, J. & SUNDIN, A. 1997. Source separated urine-nutrient and heavy metal content, water saving and faecal contamination. *Water science and technology,* 35**,** 145-152.

JÖNSSON, H., STINTZING, A. R., VINNERAS, B. & SALOMON, E. 2004. Guidelines on the Use of Urine and Faeces in Crop Production. *EcoSanRes Publication Series.* Stockholm: Stockholm Environment Institute.

KABDAŞLI, I., TÜNAY, O., İŞLEK, Ç., ERDINÇ, E., HÜSKALAR, S. & TATLI, M. B. 2006. Nitrogen recovery by urea hydrolysis and struvite precipitation from anthropogenic urine. *Water Science and Technology,* 53**,** 305-312.

KING, F. H. 1911. *Farmers of Forty Centuries; Or, Permanent Agriculture in China, Korea, and Japan*, Project Gutenberg eBook.

KIRCHMANN, H. & PETTERSSON, S. 1994. Human urine - Chemical composition and fertilizer use efficiency. *Fertilizer research,* 40**,** 149-154.

KRAJEWSKA, B. 2009. Ureases I. Functional, catalytic and kinetic properties: A review. *Journal of Molecular Catalysis B: Enzymatic,* 59**,** 9-21.

LASA, B., FRECHILLA, S., LAMSFUS, C. & APARICIO-TEJO, P. M. 2000. The sensitivity to ammonium nutrition is related to nitrogen accumulation. *Scientia Horticulturae,* 91**,** 143-152.

LEDEZMA, P., KUNTKE, P., BUISMAN, C. J. N., KELLER, J. & FREGUIA, S. 2015. Source-separated urine opens golden opportunities for microbial electrochemical technologies. *Trends in Biotechnology,* 33**,** 214-220.

LIND, B. B., BAN, Z. & BYDEN, S. 2000. Nutrient recovery from human urine by struvite crystallization with ammonia adsorption on zeolite and wollastonite. *Bioresource Technology,* 73**,** 169-174.

LIU, Q., LIU, C., ZHAO, L., MA, W., LIU, H. & MA, J. 2016. Integrated Forward Osmosis-Membrane Distillation Process for Human Urine Treatment. *Water Research.*

MATUBAYASI, N., NAMIHIRA, J. & YOSHIDA, M. 2003. Surface properties of aqueous amino acid solutions. *Journal of Colloid and Interface Science,* 267**,** 144-150.

MAURER, M., PRONK, W. & LARSEN, T. A. 2006. Treatment processes for source-separated urine. *Water Research,* 40**,** 3151-3166.

MENZIES, D. J. & O'CALLAGHAN, J. R. 1971. The effect of temperature on the drying rate of grass. *Journal of Agricultural Engineering Research,* 16**,** 213-222.

MILLS, C. O., ELLAS, E., MARTIN, G. H. B., WOO, M. T. C. & WINDER, A. F. 1988. Surface Tension Properties of Human Urine: Relationship with Bile Salt Concentration. *Journal of Clinical Chemistry and Clinical Biochemistry,* 26**,** 187-194.

NG, P. P., TASIRIN, S. M. & LAW, C. L. 2006. Thin layer method analysis of spouted bed dried Malaysian paddy characteristic drying curves. *Journal of Food Process Engineering,* 29**,** 414 - 428.

NORDIN, A. 2010. *Ammonia Sanitazation of Human Excreta.* Doctoral, Swedish University of Agricultural Sciences.

NYBERG, K. A., VINNERÅS, B., LEWERIN, S. S., KJELLBERG, E. & ALBIHN, A. 2011. Treatment with Ca(OH)2 for inactivation of Salmonella Typhimurium and Enterococcus faecalis in soil contaminated with infected horse manure. *Journal of Applied Microbiology,* 110**,** 1515-1523.

PAHORE, M. M., ITO, R. & FUNAMIZU, N. 2010. Rational design of an on-site volume reduction system for source-separated urine. *Environmental Technology,* 31**,** 399-408.

PAHORE, M. M., USHIJIMA, K., ITO, R. & FUNAMIZU, N. 2011. Fate of nitrogen during volume reduction of human urine using an on-site volume reduction system. *Environmental Technology,* 33**,** 229-235.

PARKER, D. B., RHOADES, M. B., COLE, N. A. & SAMBANA, V. P. 2012. EFFECT OF UREASE INHIBITOR APPLICATION RATE AND RAINFALL ON

AMMONIA EMISSIONS FROM BEEF MANURE. *Transactions of the Asabe,* 55**,** 211-218.
PENHALLEGON, R. 2003. Nitrogen-Phophorus-Potassium Values of Organic Fertilizers. *In:* AILOR, K. (ed.). Oregon: Oregon State University Extension Service.
PETER, J., GREEN, C., HOELSCHER, M., MWABA, P., ZUMLA, A. & DHEDA, K. 2010. Urine for the diagnosis of tuberculosis: current approaches, clinical applicability, and new developments. *Current opinion in pulmonary medicine,* 16**,** 262-270.
POWER, J. F. & PRASAD, R. 1997. *Soil Fertility Management for Sustainable Agriculture,* Boca Raton, FL, CRC Press.
PROSSER, J. I. 2011. Nitrogen Transformations. *In:* HUANG, P. M., LI, Y. & SUMMER, M. E. (eds.) *Handbook of Soil Sciences: Properties and Processes.* 2 ed. Boca Raton, FL: CRC Press.
PUTNAM, D. F. 1971. Composition and concentrative properties of human urine. *National Aeronautics and Space Administration Contractor Report.* Huntington Beach, California: McDonnell Douglas Astronautics Company - Western Division.
RAMÍREZ, C. A. & WORRELL, E. 2006. Feeding fossil fuels to the soil: An analysis of energy embedded and technological learning in the fertilizer industry. *Resources, Conservation and Recycling,* 46**,** 75-93.
RANDALL, D. G., KRÄHENBÜHL, M., KÖPPING, I., LARSEN, T. A. & UDERT, K. M. 2016. A novel approach for stabilizing fresh urine by calcium hydroxide addition. *Water Research,* 95**,** 361-369.
ROCKSTRÖM, J., STEFFEN, W., NOONE, K., PERSSON, A., CHAPIN, F. S., LAMBIN, E. F., LENTON, T. M., SCHEFFER, M., FOLKE, C., SCHELLNHUBER, H. J., NYKVIST, B., DE WIT, C. A., HUGHES, T., VAN DER LEEUW, S., RODHE, H., SORLIN, S., SNYDER, P. K., COSTANZA, R., SVEDIN, U., FALKENMARK, M., KARLBERG, L., CORELL, R. W., FABRY, V. J., HANSEN, J., WALKER, B., LIVERMAN, D., RICHARDSON, K., CRUTZEN, P. & FOLEY, J. A. 2009a. Planetary Boundaries: Exploring the Safe Operating Space for Humanity. *Ecology and Society,* 14**,** 32.
ROCKSTRÖM, J., STEFFEN, W., NOONE, K., PERSSON, Å., CHAPIN, F. S., LAMBIN, E. F., LENTON, T. M., SCHEFFER, M., FOLKE, C., SCHELLNHUBER, H. J., NYKVIST, B., DE WIT, C. A., HUGHES, T., VAN DER LEEUW, S., RODHE, H., SORLIN, S., SNYDER, P. K., COSTANZA, R., SVEDIN, U., FALKENMARK, M., KARLBERG, L., CORELL, R. W., FABRY, V. J., HANSEN, J., WALKER, B., LIVERMAN, D., RICHARDSON, K., CRUTZEN, P. & FOLEY, J. A. 2009b. A safe operating space for humanity. *Nature,* 461**,** 472-475.
SCHÖNNING, C., LEEMING, R. & STENSTRÖM, T. A. 2002. Faecal contamination of source-separated human urine based on the content of faecal sterols. *Water Research,* 36**,** 1965-1972.
SHIMING, L. 2002. The utilization of human excreta in Chinese agriculture and the challenge faced. South China Agricultural University, EcoSanRes.
SIZER, I. W. 1940. The activation energy of urea hydrolysis catalyzed by soy bean urease. *Journal of Biological Chemistry,* 132**,** 209-218.
SMIL, V. 2002. Nitrogen and Food Production: Proteins for Human Diets. *Ambio,* 31**,** 126-131.
STEENARI, B.-M. 1998. *Chemical Properties of FBC Ashes.* Doctoral, Chalmers University of Technology.
STEFFEN, W., RICHARDSON, K., ROCKSTRÖM, J., CORNELL, S. E., FETZER, I., BENNETT, E. M., BIGGS, R., CARPENTER, S. R., DE VRIES, W., DE WIT, C. A., FOLKE, C., GERTEN, D., HEINKE, J., MACE, G. M., PERSSON, L. M.,

RAMANATHAN, V., REYERS, B. & SÖRLIN, S. 2015. Planetary boundaries: Guiding human development on a changing planet. *Science,* 347.

TARPEH, W. A., UDERT, K. M. & NELSON, K. L. 2017. Comparing ion exchange adsorbents for nitrogen recovery from source-separated urine. *Environmental Science and Technology,* 51**,** 2373-2381.

TCHOBANOGLOUS, G., BURTON, F. L. & STENSEL, H. D. 2003. *Wastewater Engineering: Treatment and Reuse,* Boston, McGraw-Hill.

TILLEY, E., ULRICH, L., LÜTHI, C., REYMOND, P. & ZURBRÜGG, C. 2014. *Compendium of sanitation systems and technologies. 2nd Revised Edition,* Dübendorf, Switzerland, Swiss Federal Institute of Aquatic Science and Technology (Eawag).

UDERT, K. M., BUCKLEY, C. A., WÄCHTER, M., MCARDELL, C. S., KOHN, T., STRANDE, L., ZÖLLIG, H., FUMASOLI, A., OBERSON, A. & ETTER, B. 2015. Technologies for the treatment of source-separated urine in the eThekwini Municipality. *Water SA,* 41**,** 212-221.

UDERT, K. M., LARSEN, T. A., BIEBOW, M. & GUJER, W. 2003. Urea hydrolysis and precipitation dynamics in a urine-collecting system. *Water Research,* 37**,** 2571-2582.

USEPA 1994. A Plain English guise to the EPA Part 503 Biosolids Rule. Office of Waste Management, Washington, D.C.: U.S. Environmental Protection Agency.

USEPA 2003. Appendix I Test Method for Detecting, Enumerating, and Determining the Viability of Ascaris Ova in Sludge. *Environmental Regulations and Technology: Control of Pathogens and Vector Attraction in Sewage Sludge.* Cincinnati, OH: United States Environmental Protection Agency.

VINNERÅS, B. & JÖNSSON, H. 2002. The performance and potential of faecal separation and urine diversion to recycle plant nutrients in household wastewater. *Bioresource Technology,* 84**,** 275-282.

VINNERÅS, B., PALMQUIST, H., BALMER, P. & JÖNSSON, H. 2006. The characteristics of household wastewater and biodegradable solid waste - A proposal for new Swedish design values. *Urban Water,* 3**,** 3-11.

WARNER, R. C. 1942. The Kinetics of the Hydrolysis of Urea and Arginine. *J. Biol. Chem.,* 142**,** 705-723.

WHO. 2006. *Volume 4: The use of excreta and greywater in agriculture* [Online]. Geneva: World Health Organization. [Accessed 07062016].

WILLEY, J. M., SHERWOOD, L. M. & WOOLVERTON, C. J. 2009. *Prescott's Principles of Microbiology,* New York, McGraw-Hill.

WINKER, M., VINNERÅS, B., MUSKOLUS, A., ARNOLD, U. & CLEMENS, J. 2009. Fertiliser products from new sanitation systems: Their potential values and risks. *Bioresource Technology,* 100**,** 4090-4096.

WORLD BANK 2017. Commodity Markets Outlook - Investment Weakness in Commodity Expoerters. *A World Bank Quarterly Report.* Washington: The World Bank.

WOZNY, M. A., BRYANT, M. P., HOLDEMAN, L. V. & MOORE, W. E. 1977. Urease assay and urease-producing species of anaerobes in the bovine rumen and human feces. *Appl. Environ. Microbiol. ,* 33**,** 1097-1104.

ZERNER, B. 1991. Recent advances in the chemistry of an old enzyme, urease. *Bioorganic Chemistry,* 19**,** 116-131.

ZHANG, Y., LI, Z., ZHAO, Y., CHEN, S. & MAHMOOD, I. B. 2013. Stabilization of source-separated human urine by chemical oxidation. *Water Science & Technology,* 67**,** 1901-1907.

Acknowledgements

To Björn, thank you for taking a chance on an email and Skype chat and giving me the opportunity join the Kretsloppsteknik research group. To the past and the present Kretsloppsteknik crew, thank you for the endless hours of team work – you are what made this whole Licentiate process fun. Whether it was exhausting days in the lab or late nights writing, you were cheerful, motivating and supportive. You are incredible people with big hearts. I am so glad that this isn't the end of our work together. Björn and Håkan, you are great leaders.

Till mina vänner i Sverige (i och utanför Kretsloppstekniksgruppen), tack för att ni gjort Sverige till hemma.

À ma famille (les Senecal/Smith et Blyth/Russel) et mes amis en Canada – merci pour toujour d'avoir patience et soutien à notre décision. Je sais qu'il n'est pas facile de nous avoir ici.

To Sam, thank you for the endless encouragement and logistical support (food!) and being an awesome partner.