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Title: Membrane stripping enables effective electrochemical ammonia recovery from urine while retaining microorganisms and micropollutants

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ABSTRACT

Ammonia recovery from urine avoids the need for nitrogen removal through nitrification/denitrification and re-synthesis of ammonia (NH_3) *via* the Haber-Bosch process. Previously, we coupled an alkalifying electrochemical cell to a stripping column, and achieved competitive nitrogen removal and energy efficiencies using only electricity as input, compared to other technologies such as conventional column stripping with air. Direct liquid-liquid extraction with a hydrophobic gas membrane could be an alternative to increase nitrogen recovery from urine into the absorbent while minimizing energy requirements, as well as ensuring microbial and micropollutant retention. Here we compared a column with a membrane stripping reactor, each coupled to an electrochemical cell, fed with source-separated urine and operated at 20 A m^{-2}. Both systems achieved similar nitrogen removal rates, 0.34 ± 0.21 and 0.35 ± 0.08 mol N L^{-1} d^{-1}, and removal efficiencies, 45.1 ± 18.4 and 49.0 ± 9.3%, for the column and membrane reactor, respectively. The membrane reactor improved nitrogen recovery to 0.27 ± 0.09 mol N L^{-1} d^{-1} (38.7 ± 13.5%) while lowering the operational (electrochemical and pumping) energy to 6.5 kWh_e kg N^{-1} recovered, compared to the column reactor, which reached 0.15 ± 0.06 mol N L^{-1} d^{-1} (17.2 ± 8.1%) at 13.8 kWh_e kg N^{-1}.

Increased cell concentrations of an autofluorescent *E. coli* MG1655+prpsM spiked in the urine influent were observed in the absorbent of the column stripping reactor after 24 h, but not for the membrane stripping reactor. None of six selected micropollutants spiked in the urine were found in the absorbent of both technologies.

Overall, the membrane stripping reactor is preferred as it improved nitrogen recovery with less energy input and generated an *E. coli*- and micropollutant-free product for

potential safe reuse. Nitrogen removal rate and efficiency can be further optimized by increasing the NH_3 vapor pressure gradient and/or membrane surface area.

KEYWORDS

membrane; micropollutant; nutrient recovery; pathogen; stripping; urine

1. INTRODUCTION

Nitrogen recovery from high-strength wastewater such as source-separated urine, short-cuts the conventional removal *via* nitrification/denitrification and fixation *via* the Haber-Bosch process. Multiple recovery technologies exist, ranging from adsorption of NH_4^+ with zeolites or activated carbon (Ganrot et al. 2007, Tarpeh et al. 2017), to precipitation as struvite for instance (Gagnon 2016, Ueno and Fujii 2001, Zamora et al. 2017). Struvite precipitation is available on pilot scale (Desmidt et al. 2015), but the lack of legislation (De Vrieze et al. 2016), social acceptance, and economic self-sustainability (Etter et al. 2011) limits its application when recovered from human waste sources.

Most recovery technologies are based on stripping *via* volatilization of ammonia (NH_3) from the liquid waste stream through an increase in pH and/or temperature (Arredondo et al. 2017, Böhler et al. 2015, Christiaens et al. 2017, Vanotti et al. 2017, Xu et al. 2017)(SI Table A.1). In general, the higher the NH_3 concentration, pH, and temperature, the more efficient the recovery technology will be. Column stripping is an established process, even on full scale, requiring caustic, heating, and forced air movement. The process has been developed for digester supernatant (Böhler et al. 2015) and urine (Antonini et al. 2011). A more recent stripping technology is membrane distillation, requiring caustic and heat to create a vapor pressure gradient across a hydrophobic membrane with gas or vacuum filled pores, separating the NH_3 liquid from the sweep gas, the applied vacuum (El-Bourawi et al. 2007), or the acid (Ahn et al. 2011, Böhler et al. 2015, Lauterbock et al. 2012, Zarebska et al. 2014). Membrane stripping, also called transmembrane chemisorption, or membrane contactor, works similarly but only with caustic and no heat. An acid on the other side of the membrane drives the NH_3 extraction by lowering the NH_3 concentration (Amaral et al. 2016, Bernal et al. 2016, Dube et al. 2016, Ulbricht et al. 2013, Vanotti et al. 2017). Hollow fiber configurations are commonly used because a large membrane surface area improves ammonia extraction rates and efficiencies (Darestani et al. 2017). A combination of (microbial) electrochemical or fuel cells with column or membrane stripping units has the advantage to produce caustic *in situ*, eliminating the need for chemicals, which are a concern, not only for their costs, but also in terms of a safe and reliable supply (Arredondo et al. 2017, Christiaens et al. 2017).

In all these technologies, NH_3 is often recovered in strong acidic absorbents (e.g. H_2SO_4 or HNO_3)(Arredondo et al. 2017, Bernal et al. 2016, Böhler et al. 2015, Christiaens et al. 2017, Vanotti et al. 2017, Xu et al. 2017). This increases the pH gradient and thus vapor pressure gradient between the alkaline NH_3 solution and the absorbent. The products $(NH_4)_2SO_4$ and NH_4NO_3 can be applied as a fertilizer, since the recovery process concentrates the nitrogen resulting in equal concentrations as synthetic fertilizer. Recovery as NH_4HCO_3 precipitate is possible in a 3-compartment microbial electrochemical cell in which the HCO_3^- is also extracted from urine (Ledezma et al. 2017). If NH_3 is not recovered in acid but in a microbial growth medium, this allows the recovered NH_3 to be used as a nitrogen source for microbial conversions such as protein production. The protein can be used as a feed or even food additive (Christiaens et al. 2017, Matassa 2016).

In any application, the quality of the recovered product needs to be safeguarded. Human urine contains microbes, viruses (Bischel et al. 2015, Decrey and Kohn

2017), and antibiotic resistance genes (Bischel et al. 2015, Pruden 2014). These mainly originate from fecal cross-contamination (Udert et al. 2015), and potentially from the bladder (Anderson et al. 2004, Sianou et al. 2016, Wolfe and Brubaker 2015, 2016). The most abundant genera, covering 83.5% of classifiable sequences found in male urine, had cell sizes with the smallest dimension ranging from 0.65-0.8 µm (*Lactobacillus* (Valik et al. 2008), *Veillonella* (Kraatz and Taras 2008), *Streptococcus*, and *Enterococcus* (Kokkinosa et al. 1998)) down to 0.1 µm (*Ureaplasma* (Shepard et al. 1974) and *Mycoplasma* (Waites and Talkington 2004))(Dong et al. 2011).
Micropollutants found in urine were mainly (metabolites of) pharmaceuticals (Bischel et al. 2015, Jaatinen et al. 2016, Kovalova et al. 2012), but also alternative plasticizers (Alves et al. 2017). Some of these micropollutants and metabolites (*e.g*. hydrochlorothiazide, sulfamethoxazole) are mainly hydrophilic at pH 9, that of hydrolysed urine, while others (*e.g*. irbesartan, clarithromycin, carbamazepine, diclofenac) are rather hydrophobic, based on the octanol-water partition ratio (log D_{ow})(https://chemicalize.com).

In our previous study, a strip and absorption column were coupled to the alkalifying cathode compartment of an electrochemical cell for NH_3 removal and recovery (Christiaens et al. 2017). Whereas the nitrogen removal percentage for real urine was 87 ± 6% at 20 A m^{-2} with 3M H_2SO_4 as absorbent, recovery was only 25 ± 12%, most likely due to nitrogen loss in the condensates throughout the lab set-up. In the present study, membrane stripping was chosen as an alternative technology for the strip and absorption column, since direct urine-absorbent contact eliminates the gas phase compartment and prevents the potential adsorption and loss of NH_3 in condensates, as already shown by Arredondo et al. (2017). Reactor set-ups with column and membrane stripping were compared in terms of nitrogen removal, recovery, and energy input. This study presents the first continuous test for membrane stripping of NH_3.

In addition to nitrogen transfer, the quality of the recovered product with respect to microorganisms and micropollutants originating from urine, was quantified. Three hypotheses were studied. First, the introduction of a membrane as a physical barrier between urine and the absorbent, retains microorganisms based on their size. In column stripping, microorganisms could move with the gas flow. Second, hydrophilic micropollutants are more easily transferred to the absorbent *via* water droplets moved with the gas flow, while hydrophobic compounds are not. Third, a hydrophobic membrane prevents transfer of any micropollutant due to the gas filled pores.

2. MATERIALS AND METHODS

2.1 Urine

Hydrolysed urine originated from a central storage and hydrolysis tank that collects men's undiluted urine *via* NoMix toilets and urinals at Eawag (Dübendorf, Switzerland). The average hydraulic retention time (HRT) in the tank was about 14 days at room temperature. Female urine collected (33% diluted with flush water) made up a limited part of the men's urine used in the present study, as a second storage tank for female urine had an overflow in the men's storage tank. One batch of urine was used for per test (Table 1).

Table 1 Average (± standard deviation, SD) composition of hydrolysed urine. (n ≥ 26)

pH	(-)	9.3 ± 0.1
electrical conductivity	(mS cm^{-1})	28 ± 1
Na^+	(mg L^{-1})	2041 ± 536
Total Ammonium Nitrogen	(mg N L^{-1})	4622 ± 719
K^+	(mg L^{-1})	1551 ± 205
Cl^-	(mg L^{-1})	2604 ± 297
NO_2^-	(mg N L^{-1})	<LOQ[a]
NO_3^-	(mg N L^{-1})	<LOQ[b]
PO_4^{3-}	(mg L^{-1})	472 ± 184
SO_4^{2-}	(mg L^{-1})	471 ± 56

[a]LOQ NO_2^- = 0.15 mg N L^{-1}

[b]LOQ NO_3^- = 0.18 mg N L^{-1}

2.2 Experimental set-up

Two technologies were compared for nitrogen removal and recovery, and bacterial or micropollutant contamination of the recovered nitrogen product: column and membrane stripping (Fig. 1). Column stripping was established *via* the same set-up as previously described (Christiaens et al. 2017) except for the gas counters which were replaced by a gastight acid trap, no gas bags on the feed vessel, different liquid pumps (Ismatec, Cole-Parmer GmbH, Germany), and a bubble column with sintered glass frit (40-250 µm pore size) as absorption column (6x110 cm^2, filled with 19 cm absorbent). A second set-up was built with a membrane stripping unit instead of the stripping and absorption columns. Two flat-sheet PTFE membranes (active surface area of 10.5x12 cm^2; 0.45, 0.2, or 0.1 µm pore size)(SI Table B.1) were mounted on both sides of a plexiglass holder, creating a middle compartment for the urine (10.5x12x0.65 cm^3). Bolting this unit with teflon rubbers and metal frames between two acrylic glass plates created two absorbent chambers (10.5x12x0.25 cm^3).

As with the column stripping, the catholyte of the electrochemical cell was recirculated over the middle compartment of the membrane unit to allow nitrogen removal and recovery before being fed in the anode compartment for additional nitrogen extraction. Analog manometers (WIKA, Switzerland) on the urine input and output of the membrane unit could indicate pressure loss and thus wetting of the membrane, which was not observed. Prior to entering the membrane unit, degassing of the catholyte was allowed. The exhaust gas was bubbled through 2M H_2SO_4 to determine potential nitrogen losses. Measurements indicated these were limited. The absorbent was fed in one of the two absorbent chambers of the membrane unit and was recirculated over the other chamber.

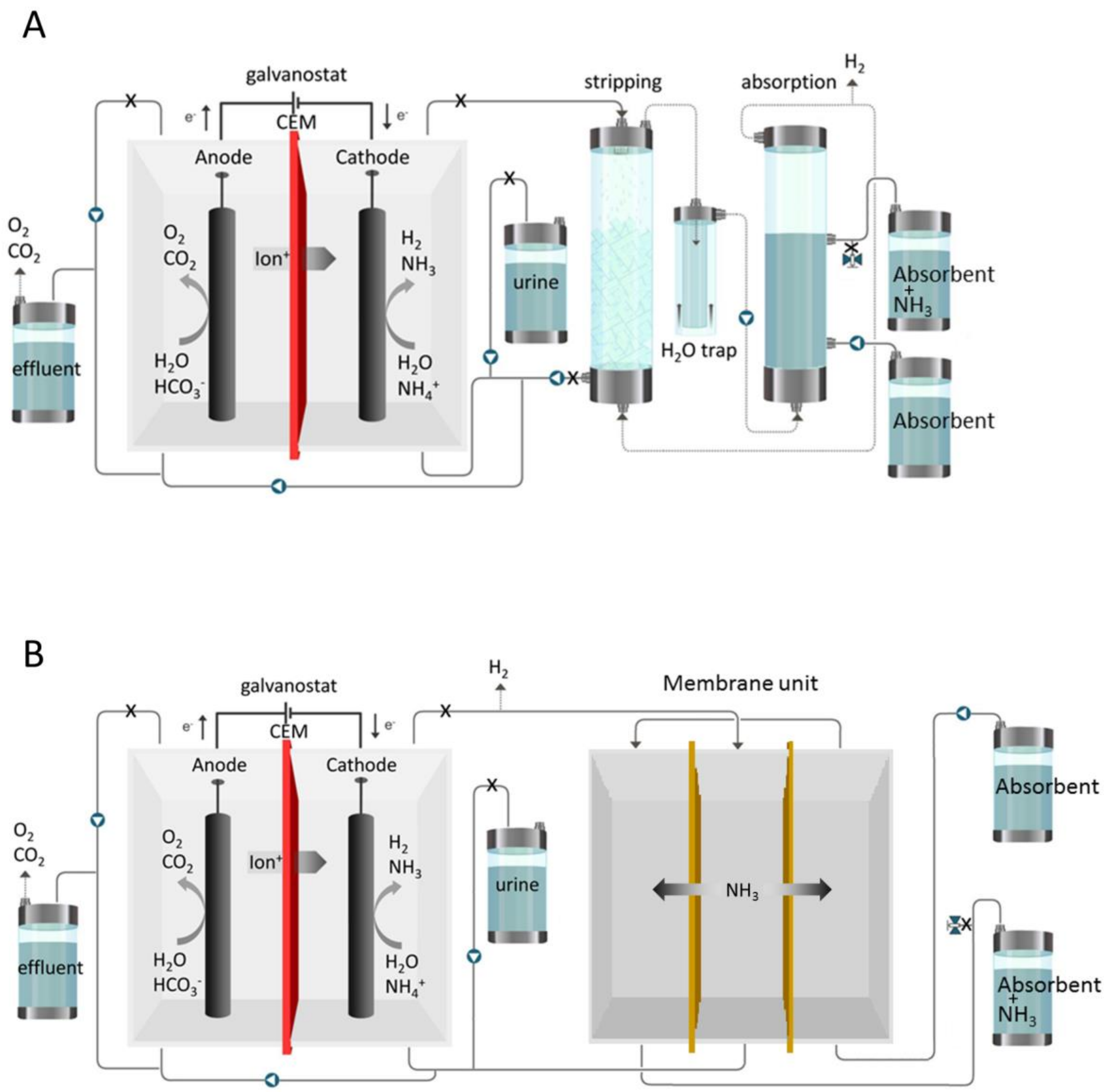

Fig. 1 Column (A) and membrane (B) stripping reactors. Sampling ports are indicated by X.

2.3 Reactor operation and product quality tests

Some applications of recovered nitrogen, such as microbial protein (MP) production, require a pH-neutral absorbent to benefit from the nitrogen alkalinity and to avoid additional chemicals for pH control (Christiaens et al. 2017). Ammonia absorption with a pH-neutral absorbent can be improved by increasing the buffer capacity and by using more absorbent *via* HRT reduction. The required phosphate buffer concentration of a neutral absorbent was calculated based on the desired maximum nitrogen recovery (SI section C. Absorbent composition) and set at 0.21 M (11.15 g $NaH_2PO_4.H_2O$ L^{-1}, 23.05 g $Na_2HPO_4.2H_2O$ L^{-1}). To avoid ion diffusion over the hydrophobic membrane, a sodium buffer was chosen to which 12.82 g $KHCO_3$ L^{-1} was added to set both Na^+ and K^+ concentrations similar to urine. The absorbent HRT (HRT_{abs}) was determined by 24 h total ammonia nitrogen (TAN) breakthrough curves with continuous urine flow and an applied current density (j) of 20 A m^{-2} (Table 2 and 4). The absorbent volume was set by the membrane unit at 0.12 L and also applied in the column stripping reactor. Liquid and gas recirculation over the membrane and column units, respectively, ensured mixing (Table 4). Samples for pH and cation analysis were collected over a 24 h time period.

Using the optimal HRT_{abs} for both technologies, their nitrogen removal and recovery was compared in continuous tests with applied current densities of 0.1 (control) and 20 A m^{-2}. Three samples were taken at steady state (*i.e.*, after 4 HRTs) and analysed for pH, EC, cations, and anions.

For bacterial and micropollutant detection in the absorbent, the reactors were operated for 24 h with continuous urine flow, as has been done for the TAN breakthrough curves. Potential bacterial transfer from urine to the absorbent product was tested by spiking the urine with an autofluorescent *E. coli* MG1655+prpsM containing a GFP plasmid that is constitutively expressed (Eawag, Dübendorf, Switzerland). In preparation for the reactor tests, the strain was grown for 24 h at 37°C and 220 rpm in LB Lennox medium (10 g L^{-1} tryptone, 5 g L^{-1} yeast extract, 5 g L^{-1} NaCl) with addition of 50 µg mL^{-1} kanamycin to retain the autofluorescent plasmid. Pellets obtained after centrifugation (10 min, 2000 rpm) were resuspended in urine.

Kanamycin was added to both urine and absorbent before filling the reactors. Samples were collected over 24 h tests for bacterial cell counts (autofluorescent FITC and live/dead staining SGPI, see 2.4 Chemical and microbial analyses), pH, and cations.

To study micropollutant transfer in both technologies, six wastewater micropollutants (Table 3) were selected (personal communication of Isabell Köpping, Eawag) ranging from hydrophilic to hydrophobic at pH 9. Concentrations spiked in urine were taken from the calculated reference urine (personal communication of Isabell Köpping, Eawag). Samples were taken over 24 h for micropollutant quantification, pH, and cations.

All tests were conducted at room temperature (20.8 ± 0.6°C) in duplicate with continuous urine feeding. Thorough cleaning with 1% HOCl and water occurred in between all (replicate) tests. Electrode potentials versus a Ag/AgCl reference electrode (± 0.247 V vs standard hydrogen electrode, 3 M NaCl, Bio-Logic, France) and cell voltages were logged with a multimeter.

Table 2 Overview of experiments, conducted in duplicate. Urine was always fed continuously.

Test	j ($A\ m^{-2}$)	Urine	Absorbent	Membrane pore size (µm)	Test duration (h)
TAN breakthrough curves	20	plain	Batch	0.1	24
Continuous TAN tests	0.1, 20	plain	Continuous	0.45	48
Pathogen tests	20	*E. coli* MG1655prpsM	Batch; kanamycin dosed	0.1	24
Micropollutant tests	20	6 micropollutants	Batch	0.2	24

Table 3 Selected micropollutants present in wastewater ranging from strongly hydrophobic to hydrophilic at pH 9. The log D_{ow} represents the octanol-water

distribution ratio of ionized and neutral forms of a compound and indicates hydrophobicity.

Micropollutant	Medical use	Log D_{ow} at pH 9[a] (-)	Aqueous solubility at pH 9[a] ($g\ L^{-1}$)	Concentration tested[b] ($\mu g\ L^{-1}$)
Irbesartan	antihypertensive	4	0.02	6.05
Clarithromycin	antibacterial	3.1	3.51	8.26
Carbamazepine	antiepileptic	2.8	0.04	4.49
Diclofenac	analgesic	0.7	296	97.2
Sulfamethoxazole	antibacterial	-0.1	253	23.5
Hydrochlorothiazide	diuretic	-0.8	9.32	102.2

[a]Source: https://chemicalize.com/

[b]Based on reference urine as determined by Isabell Köpping – personal communication)

Table 4 Hydraulic retention times (HRT, h), volumes (V, L), and average (± SD) rates ($L\ d^{-1}$) for inflow (Q) and recirculation ($Q_{recirculation}$) for all compartments in both column and membrane stripping reactors. (n = 3)

Reactor	Compartment	HRT (h)	V (L)	Q ($L\ d^{-1}$)	$Q_{recirculation}$ ($L\ d^{-1}$)
Column	Cathode & stripping column	7.7[a]	0.41	1.27 ± 0.01	326 ± 17
	Anode	3.9	0.21	1.24 ± 0.05	274 ± 25
	Absorbent	2.0[b,d]	0.55[d]	6.7 ± 0.2[d]	7 ± 0[e]
Membrane	Cathode & middle compartment membrane unit	8.3[a]	0.44	1.24 ± 0.03	288 ± 0
	Anode	3.4	0.18	1.26 ± 0.03	301 ± 1
	Absorbent	1.1[c,d]	0.12	2.8 ± 0.1[d]	285 ± 3

[a]Similar as Christiaens et al. (2017)

[b]Quantified in the nitrogen breakthrough curves for the neutral pH absorbent

[c]Intended to be the same as the HRT of the absorbent in the column stripping set-up

[d]Only in tests in which the absorbent was continuously refreshed. In batch, the absorbent volume in the column reactor was 0.12 L with an HRT of 24 h.

[e]Gas recirculation

2.4 Chemical and microbial analyses

Liquid samples were filtered (0.22 µm) before analyzing pH, electrical conductivity, and ion concentrations. Cations (Na^+, TAN, K^+) were quantified with a 761 Compact IC with a Metrosep C4/4.0 guard and a C4-150/4.0 main column (Metrohm, Switzerland)(LOQ of 1-20, 0.78-30, and 1-20 mg L^{-1} for Na^+, TAN, K^+, respectively). Anions (Cl^-, NO_2^-, NO_3^-, PO_4^{3-}, SO_4^{2-}) were quantified by a 881 compact IC Pro with a Metrosep A Supp 5/4.0 guard and a Metrosep A Supp 7 250/4.0 main column (Metrohm, Switzerland)(LOQ of 0.8-40, 0.15-7.6, 0.18-9, 0.13-6.5, 0.8-40 mg L^{-1} for Cl^-, NO_2^--N, NO_3^--N, PO_4^{3-}-P, SO_4^{2-}, respectively). Both devices were equipped with conductivity detectors (Metrohm, Switzerland).

Intact, membrane-ruptured, and autofluorescent bacterial cell populations were quantified by flow cytometry (Beckman Coulter Cytoflex, IN, US) after dilution with 0.22 µm filtered evian water (limit of detection, LOD; 10^3 events mL^{-1}). To assess the viability of the bacteria, samples were stained with a mixture of SYBR Green I (SG)(10 000 times diluted from stock, Invitrogen, US), binding all DNA, and Propidium Iodide (PI)(3 µM final concentration, Invitrogen), only staining permeable cells. After incubation at 37°C for 13 min., samples were excited by a 488 nm laser and quantified by two fluorescent detectors (525/40 and 585/42 for SG and PI, respectively). Unstained samples allowed quantifying the autofluorescent cell population by excitation at 488 nm and detection by a 525/40 fluorescent detector. Heat-killed and 0.22 µm filtered controls allowed the identification and quantification of non-viable populations and noise, respectively, *via* manual gating.

Target micropollutants were extracted from the urine matrix, and separated by an UHPLC system (Dionex, Amsterdam, The Netherlands). Chromatographic separation was achieved using reversed phase chromatography with gradient elution. The detection of target compounds was carried out using a Q-Exactive™ Benchtop HRMS (Thermo Fisher Scientific, San-Francisco, USA) fitted with a Heated Electrospray Ionization (HESI-II) source. More detailed information can be found in SI (section D. Micropollutant analysis).

All calculations performed are described in SI (section E. Calculations).

3. RESULTS & DISCUSSION

3.1 ABSORBENT HRT OPTIMIZATION FOR NH_3 RECOVERY

Ammonia recovery technologies often include absorption in (strong) acid for the production of $(NH_4)_2SO_4$ or NH_4NO_3, applicable as fertilizer. The conditional dissociation constant (pKa') of NH_3 for hydrolysed urine at 20.8°C is 9.5, as calculated after Stumm and Morgan (1996). An acid absorbent guarantees strong dissociation and thus absorption capacity. A pH-neutral solution still allows dissociation but results in a lower absorption capacity. This can be improved by reducing the HRT in the absorption unit, *i.e.*, using a higher absorbent volume compared to the urine volume, and by applying a strong phosphate (P) buffer. In this way, a pH-neutral absorbent could be an alternative to produce a slightly alkaline nitrogen solution that can be used as a nitrogen source and pH correction agent, *e.g.* in microbial protein production (Christiaens et al. 2017). This is a more safe alternative for the chemicals required as pH control in microbial production processes.

The absorption breakthrough profiles for the column and membrane stripping reactors enabled selecting the absorbent HRT at which the buffer capacity (here: 0.21 mol H^+ L^{-1}) was not exceeded (Fig. 2). As a safety margin, a buffer capacity of 0.14 mol H^+ L^{-1} was considered allowing a concentration of 0.14 mol N L^{-1} or 2 g TAN L^{-1} to build up in the absorbent. The corresponding HRT is thus 2 h and was applied in the continuous tests further on.

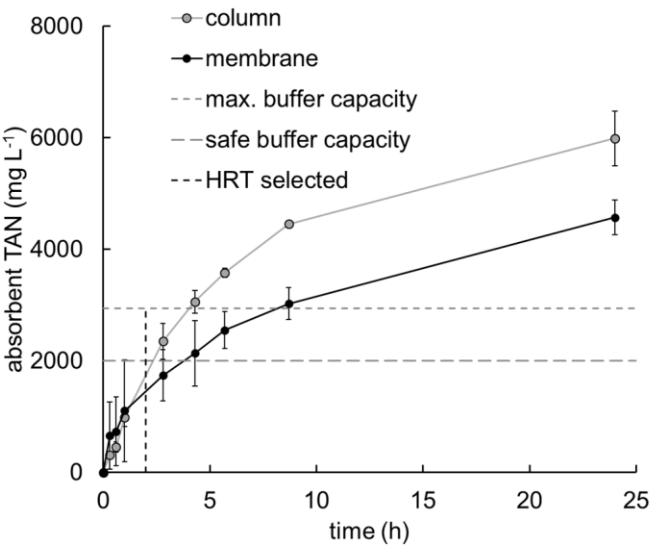

Fig. 2 Average (± SD) 24 h total ammonia nitrogen (TAN, mg N L^{-1}) concentration profiles in a phosphate buffer absorbent for the continuously operated column and membrane stripping reactors at 20 A m^{-2}. The maximum (0.21 mol P L^{-1}) and safe (0.14 mol P L^{-1}) buffer capacity and the selected HRT_{abs} are indicated by dashed lines. (n = 2)

3.2 COLUMN AND MEMBRANE STRIPPING EQUALLY EFFECTIVE FOR NITROGEN REMOVAL

To quantify nitrogen removal, recovery, and the required energy input, both the column and membrane stripping reactors were continuously operated at 0.1 (control) and 20 A m^{-2} with real urine as ammonia source and the phosphate buffer as absorbent. Both systems performed similarly in terms of nitrogen removal rates, efficiencies, and electrochemical energy efficiency under the low and high applied current density (Table 5).

Nitrogen removal rates at 20 A m^{-2} in the column stripping reactor (0.34 ± 0.21 mol N L^{-1} d^{-1}) were, however, lower compared to values reported earlier (0.58 ± 0.07 mol N L^{-1} d^{-1} in Christiaens et al. (2017)), resulting in 50% lower efficiencies and a three times higher energy demand (Table 5). Reactor set-ups and operation were the same and the hydrolysed urine had a similar composition, except for 1 g N L^{-1} less nitrogen relative to the previous study. The lower nitrogen removal rate was accompanied by a lower catholyte pH (8.7 ± 0.1 compared to 9.4 ± 0.1 for Christiaens et al. (2017)), which is usually only observed in control experiments with NH_3 stripping and limited OH^- production (SI Table F.1). As the applied current density and cathode potentials were similar in both studies (SI Table F.1), not electrocatalytic OH^- production but its consumption might be different. A hypothesis could be *in situ* Fe^{3+} precipitation as $Fe(OH)_3$ (lowest solubility at pH 8) since the cathode (*i.e.*, stainless steel) might have been partly oxidized as it was exposed to bleach during a cleaning step before starting the experiments and some precipitation was observed during the experiments. To date, no decision could be taken about this observation which does not modify our comparison between the technologies.

The membrane reactor had equally low nitrogen removal rates as the column reactor in this study, although the catholyte pH was as expected (9.5 ± 0.1). The hydrophobic membrane was limiting the nitrogen flux (3.9 ± 1.5 g N m^{-2} h^{-1}) as the removal rate almost equaled the recovery rate. However, this flux was found to be similar to other membrane stripping units (Arredondo et al. 2017, Dube et al. 2016), except for Tarpeh et al. (2018), who reported 42 g N m^{-2} h^{-1} for undiluted urine stripping.

Table 5 Average (± SD) nitrogen removal and recovery rates (mol N d^{-1} and mol N L^{-1} d^{-1}), efficiencies (%), and electrochemical energy requirements (kWh_e kg^{-1} N) in steady state for both column and membrane stripping reactors in this study (col and mem, respectively) with phosphate buffer as absorbent, and the column stripping reactor (ColSCP) operated with acid absorbent as described in Christiaens et al. (2017). (n = 3)

j	A m^{-2}		0.1		20	
			removal	recovery	removal	recovery
Rate	mol N d^{-1}	ColSCP	-0.06 ± 0.07	0.07 ± 0.06	0.32 ± 0.04	0.10 ± 0.05
		Col	0.05 ± 0.05	0.03 ± 0.01	0.21 ± 0.13	0.09 ± 0.04

		Mem	0.08 ± 0.06	0.06 ± 0.03	0.22 ± 0.05	0.17 ± 0.06
	mol N L^{-1} d^{-1}	ColSCP	-0.11 ± 0.13	0.13 ± 0.11	0.58 ± 0.07	0.18 ± 0.09
		Col	0.08 ± 0.08	0.05 ± 0.02	0.34 ± 0.21	0.15 ± 0.06
		Mem	0.13 ± 0.09	0.09 ± 0.05	0.35 ± 0.08	0.27 ± 0.09
Membrane flux[a]	g N m^{-2} h^{-1}	Mem	n.a.	1.5 ± 0.7	n.a.	3.9 ± 1.5
Efficiency	%	ColSCP	4.7 ± 1.4	25.3 ± 23.1	87.1 ± 6	25.0 ± 12.1
		Col	11.4 ± 12.8	7.5 ± 3.4	45.1 ± 18.4	17.2 ± 8.1
		Mem	19.7 ± 16.8	14.7 ± 9.8	49.0 ± 9.3	38.7 ± 13.5
Energy	kWh_e kg^{-1} N	ColSCP	0.05	0.04	1.9	5.8
		Col	0.01 ± 0.06	0.1 ± 0.03	5.9 ± 3.1	12.2 ± 4.8
		Mem	0.04 ± 0.01	0.1 ± 0.03	4.6 ± 1.1	6.3 ± 2.3

n.a. means not applicable

[a]Parameter only applicable to membrane set-up

3.3 MEMBRANE STRIPPING IMPROVES NITROGEN RECOVERY, REDUCING ENERGY COST

Nitrogen recovery at 20 A m^{-2} was almost two fold higher for membrane stripping (0.27 ± 0.09 mol N L^{-1} d^{-1}) compared to the column stripping reactor (0.15 ± 0.06 mol N L^{-1} d^{-1}). As a result, the nitrogen recovery efficiency doubled with only half the electrochemical energy requirement of the column stripping reactor (Table 5). The direct liquid-liquid contact likely prevented evaporative losses that might have occurred in the column stripping reactor. The unintended lower absorbent HRT in the membrane reactor could have contributed to this improved nitrogen recovery (Table 4). The extra energy for this lower absorbent HRT did hardly make a difference in the overall operational energy requirements on lab scale, since the absorbent volume and thus flow rate for the column reactor was higher (Table 4). The total (electrochemical and pumping) operational energy required for the membrane reactor was calculated at 6.5 kWh_e kg N^{-1} recovered while for the column reactor this was 13.8 kWh_e kg N^{-1} recovered (SI Eq. E.10-13; Table G.1). These values include power consumption of the electrochemical cell, feed and recirculation pumps, and the gas recirculation pump for the column reactor. Differences in HRT and thus flow rate made up only minor differences in energy requirements between both reactor set-ups. Even the gas recirculation pump was not a major contribution in this lab scale set-up. The liquid recirculation over the stripping column made the difference as a height of at least 1 m needed to be overcome. Additionally, the membrane unit is considered to be safer because the H_2 gas, *in situ* produced at the cathode that enhances NH_3 stripping, is not recirculated in a separate gas loop.

Overall, the membrane reactor consumed less energy, was safer to operate, and improved nitrogen recovery compared to the column reactor, but nitrogen removal should be optimized. Increasing the NH_3 vapor pressure gradient could already be sufficient since the average absorbent pH in steady state at 20 A m^{-2} (9 ± 1 for the membrane and 7.9 ± 0.2 for the column reactor) was close to the feed pH (9.3 ± 0.1 and 9.2 ± 0.1, respectively). This could be implemented by further decreasing the absorbent HRT (Ahn et al. 2011). In addition, heat could be applied, shifting to membrane distillation. However, a temperature difference of 30-40°C is required to drastically improve the nitrogen removal and recovery efficiency, increasing the operational energy costs (Ahn et al. 2011, Derese 2018). Enlarging the membrane surface area could have an impact although the surface area was calculated based on the nitrogen flux expected in membrane distillation (Derese 2018).

Coupling a membrane stripping unit to an electrochemical cell comes with advantages if conceptually compared to other nitrogen recovery technologies, such as conventional column stripping, membrane distillation, or membrane contactors. First, the *in situ* production of H_2 gas and caustic, with only sustainably produced electricity, is considered safer, eliminates the need for chemicals, and thus their cost and transport. Second, the absorbent can be a microbial medium instead of a strong acid, which can be used to make a high-value product, such as microbial protein (Matassa 2016).

3.4 TAN ELECTROMIGRATION INSTEAD OF DIFFUSION

The coulombic efficiency at 20 A m^{-2}, defined as the amount of cations extracted from the anolyte over the cation exchange membrane (CEM) compared to the amount of applied electrons, indicated that the nitrogen flux over the CEM, or the transmembrane flux, was mainly driven by electromigration in both the membrane and column stripping reactors (Fig. 3). The TAN transmembrane flux towards the cathode compartment was three times higher compared to the one reported in Christiaens et al. (2017)(SI Table F.1), where diffusion was pointed as the main driver for the nitrogen flux.

The anolyte ion concentration, but also electrical charge and diffusion coefficients, determine which ions will account for the charge balance *via* electromigration (Christiaens et al. 2017, Cord-Ruwisch et al. 2011, Desloover et al. 2012). The lower nitrogen removal *via* stripping in this study resulted in a TAN accumulation in the catholyte and thus also in the anolyte. The TAN anolyte concentrations were three times higher (3126 ± 403 and 3070 ± 841 mg N L^{-1} for column and membrane stripping, respectively) compared to Christiaens et al. (2017)(1155 ± 692 mg N L^{-1}). As the anolyte pH was 7.7 ± 0.2 and 8.0 ± 0.6 for column and membrane stripping, respectively, anolyte TAN was mainly present as NH_4^+-N (98% and 96%, respectively). Conversely, the anolyte concentrations of other main ions (Na^+ and K^+) were similar in both studies and two times lower compared to the tripled TAN concentrations. Consequently, electromigration of TAN was favored. These results confirm earlier work where an EC fed with real hydrolysed urine in the anode compartment was coupled to a stripping column (Luther et al. 2015) and coupled to a membrane contactor (Arredondo et al. 2017).

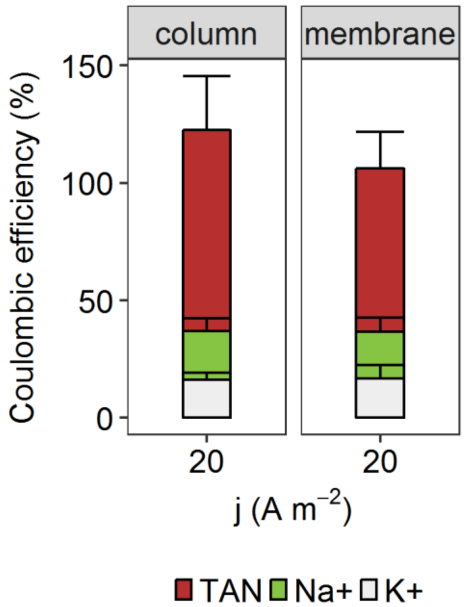

Fig. 3 Average (± SD) coulombic efficiency (%) at 20 A m^{-2} for monovalent ions in steady state for both column (left) and membrane (right) stripping reactors. The coulombic efficiency is defined as the amount of cations extracted from the anolyte over the cation exchange membrane compared to the amount of applied electrons. The sum of all ions makes up for a 100% efficiency indicating electromigration. (n = 3)

3.5 A HYDROPHOBIC MEMBRANE PREVENTS TRANSFER OF CONTAMINATING BACTERIA

If the recovered nitrogen will be used to produce proteins for feed or food applications, contamination by microorganisms present in urine needs to be avoided. If microorganisms are still present, a post-treatment step might be required (*e.g.* filtration or sterilization by ozone, UV, or chlorination)(Jones et al. 2018, Macauley et al. 2006), which adds energy and chemical costs.

During 24 h batch NH_3 stripping tests, the feed and absorbent of both column and membrane stripping reactors were monitored for autofluorescent *E. coli* MG1655+prpsM cells that had been spiked into the feed bottle as a tracer organism, but also for total intact and ruptured cells. The initial concentration of *E. coli* MG1655+prpsM in the feed bottles of the column and membrane reactors was $5.8 \cdot 10^{8} \pm 1.9 \cdot 10^{8}$ and $6.1 \cdot 10^{8} \pm 9.9 \cdot 10^{7}$ events mL^{-1}, respectively, while the concentration in the absorbents was below the LOD (10^{3} events mL^{-1})(Fig. 4). After 24 h there was a slight increase in spiked *E. coli*, intact, and ruptured cells of the influent as the intake tubing reached the settled biomass.

The spiked *E. coli* only increased in the absorbent of the column reactor, from $1.2 \cdot 10^{3} \pm 7.5 \cdot 10^{2}$ to $1.3 \cdot 10^{4} \pm 1.6 \cdot 10^{4}$ events mL^{-1} after 24 h. The absorbent of the membrane reactor increased for intact and ruptured cell counts after 24 h from $2.1 \cdot 10^{5} \pm 2.2 \cdot 10^{5}$ to $1.4 \cdot 10^{6} \pm 1.7 \cdot 10^{6}$ events mL^{-1}, and from $1.1 \cdot 10^{6} \pm 5.4 \cdot 10^{5}$ to $7.9 \cdot 10^{6} \pm 9.4 \cdot 10^{6}$ events mL^{-1}, respectively. Growth was rather unlikely as the absorbent accumulated free ammonia (FA) to 4759 ± 550 and 3895 ± 408 mg N L^{-1} after 24 h at a pH of 9.9 ± 0.1 and 10.2 ± 0.1 for the column and membrane reactors, respectively (Fig. H.1). Vinneras et al. (2008) reported ammonia concentrations of 2100 mg N L^{-1} at pH 8.9 to rapidly inactivate enteric pathogens. Allievi et al. (1994) found that mainly FA inactivated bacterial cells if 10°C is reached, enhanced by an alkaline pH and/or increased salinity, while other studies showed a pH increase to be sufficient (Diez-Gonzalez et al. 2000, Ogunyoku et al. 2016). In microbial cells, FA probably disintegrates the cell membrane (Jenkins et al. 1999) by membrane and protein denaturation (Bujoczek 2001) and alkalinisation of the cytoplasm (Diez-Gonzalez et al. 2000).

More plausible is the transfer of the tracer organism *E. coli* to the absorbent *via* urine aerosols (Benami et al. 2016, Heinonen-Tanski et al. 2009). Transfer of the smallest microorganisms reported in urine (0.1 µm)(Dong et al. 2011, Shepard et al. 1974, Waites and Talkington 2004, Wang et al. 2007) could have happened through the hydrophobic membrane which had an average measured pore size of 0.274 ± 0.003 µm, whereas the manufacturer reported 0.1 µm (SI Table B.1). Only *via* wetting of the gas-filled pores microorganisms could have transferred. However, these small cells apparently could not transfer *via* the gas phase in the column stripping reactor, although *E. coli*, a large microorganism with minimum diameter of 1 µm (Gagnon 2016), could. Innovative hydrophobic gas permeable membranes with track-etched pores could provide a tailor-made and extremely narrow pore size to prevent future transfer of microorganisms (Apel 2001).

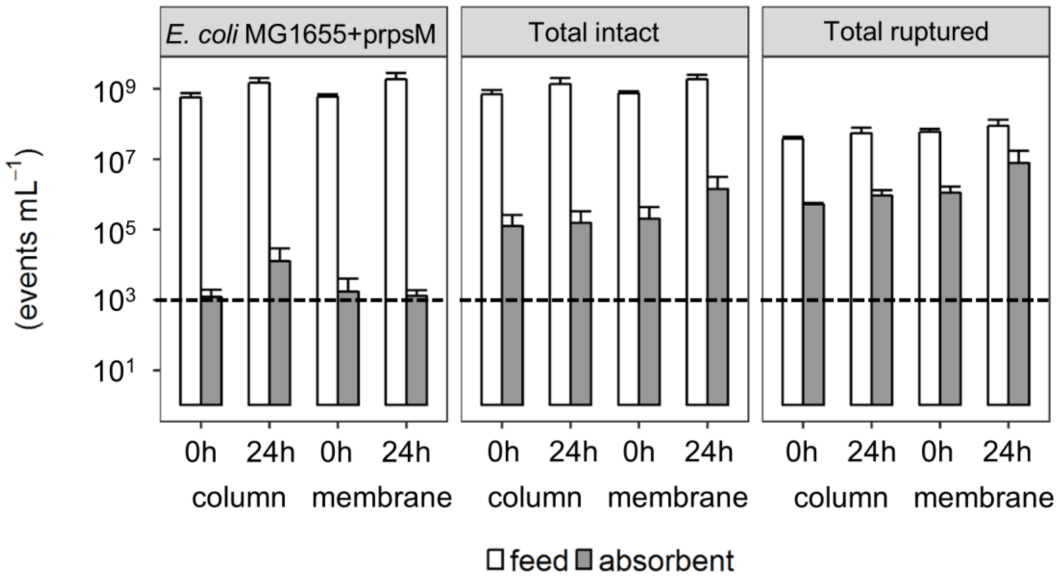

Fig. 4 Average (± SD) cell counts (events mL^{-1}) at 0 and 24 hours for autofluorescent *E. coli* MG1655+prpsM, total intact, and total ruptured microorganisms, in the feed and absorbent compartments of the column and membrane reactors. The LOD for flow cytometry is at 10^3 cells mL^{-1}, as indicated by the dashed line. (n = 2)

3.6 NO MEASURED MICROPOLLUTANTS TRANSFERRED TO THE RECOVERED PRODUCT

The absence of urine-derived micropollutants in the absorbent with recovered nitrogen is a prerequisite for reuse applications. Six micropollutants, ranging from hydrophobic to hydrophilic at pH 9 (Table 3), were spiked in the urine fed to both column and membrane stripping reactors and monitored in both the feed and absorbent during 24 h batch NH_3 stripping tests. Measured carbamazepine concentrations in urine at the start of the tests exceeded the spiked concentration (4.49 µg L^{-1}) as it was already present at 37.54 ± 7.83 µg L^{-1} (replicate 1) and 35.38 ± 0.44 µg L^{-1} (replicate 2).

Despite the hypothesis that hydrophilic compounds could transfer more easily in the column reactor and hydrophobic compounds might transfer better through the hydrophobic membrane, no difference could be observed between the column and membrane stripping reactor (Fig. 5). Moreover, while micropollutant concentrations in the feed urine reflected spiked concentrations (except for carbamazepine), no micropollutants were detected in the absorbents (below LOD 0.25 and 0.50 µg L^{-1}). These results are in line with recent work by Tarpeh et al. (2018) who reported the absence of urine-derived trace organics (<0.1 µg L^{-1}) in the acid trap after a hydrophobic NH_3 extraction membrane. However, Böhler et al. (2015) reported the transfer of micropollutants *via* column stripping to the absorption column. Carbamazepine, for instance, reached 0.1 µg L^{-1} in the $(NH_4)_2SO_4$ recovered product. With respect to the LODs reported here, we can conclude that both column and membrane stripping techniques can be safely used for nitrogen recovery in view of detectable micropollutant concentrations. However, additional concentration quantification with devices where the LOD reaches to 0.1 µg L^{-1} would be recommended.

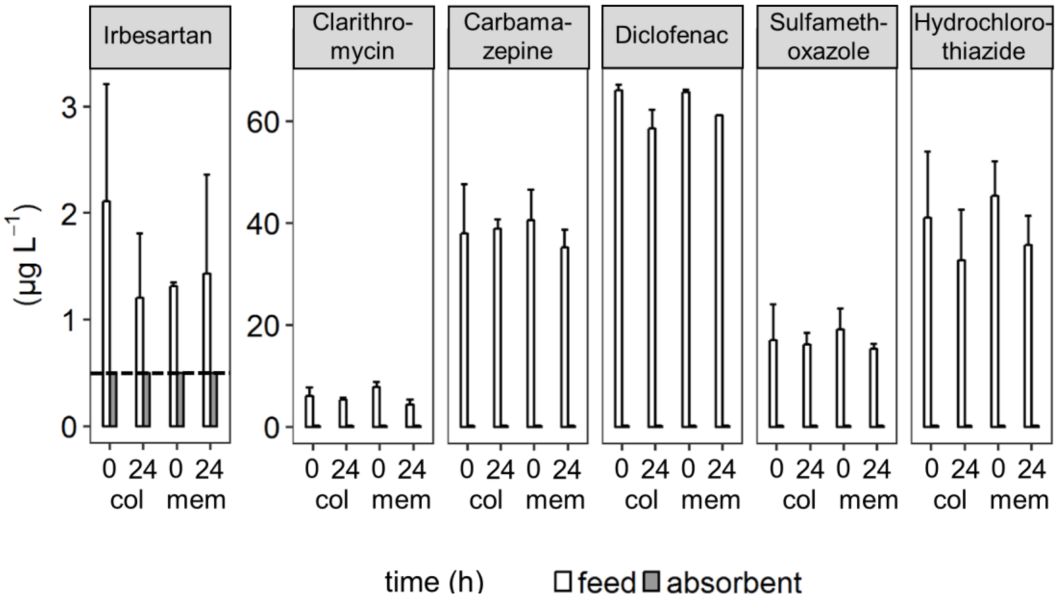

Fig. 5 Average (± SD) micropollutant concentrations (µg L^{-1}) at 0 and 24 hours in the feed and absorbent compartments of the column (col) and membrane (mem) reactors. All concentrations for the absorbent were below the LOD (0.5 µg L^{-1} for irbesartan, indicated by the dashed line, and 0.25 µg L^{-1} for the other micropollutants). (n = 2)

4. CONCLUSIONS

Nitrogen removal and recovery from source-separated urine and the safety of the recovered product were evaluated for two technologies: column and membrane stripping, both coupled to an electrochemical cell.

- Both technologies performed similarly for nitrogen removal. Nitrogen recovery was clearly improved in a membrane stripping reactor by reducing nitrogen losses, that potentially occurred *via* condense water in the gas phase of a stripping column.
- Membrane stripping reduced operational energy requirements (electrochemical and pumping) with 50% compared to the column stripping reactor.
- Lower overall nitrogen removal rates for both stripping technologies compared to earlier work resulted in nitrogen accumulation in the anolyte compared to Na^+ and K^+, which caused TAN electromigration towards the catholyte.
- Membrane stripping prevented the transfer of the autofluorescent *E. coli* MG1655+prpsM spiked in the urine towards the absorbent, whereas increased cell concentrations were observed in the absorbent of the column stripping reactor.
- Urine-derived micropollutants were below the LOD in the absorbent of both stripping technologies.

Overall, membrane stripping should be optimized for nitrogen removal by increasing the NH_3 vapor pressure gradient and/or membrane surface area. However, it is preferred over column stripping since it improved nitrogen recovery, consumed less energy, and prevented transfer of spiked microbial cells and micropollutants into the recovered nitrogen product.

The authors declare no competing financial interest.

The Supporting Information is available free of charge on the Elsevier website at DOI: …

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