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Urea hydrolysis and recovery of nitrogen and phosphorous as MAP from stale human urine

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Abstract

Laboratory-scale tests for magnesium ammonium phosphate (MAP) precipitation following urea hydrolysis of human urine were conducted using orthogonal experiment design. The effects of initial pH, temperature and the volumetric ratios of stale urine to fresh urine, on urea hydrolysis in urine were studied to determine the final hydrolysis time to recover most nitrogen from separated human urine by MAP. With a volumetric ratio of stale to fresh urine >10% and at temperature ⩾20°C, urea hydrolysis could be completed in two days. Alkaline pH inhibited urea hydrolysis progress. The final pH values were all around 9.0 following urine hydrolysis, while the suspension pH might act as an indicator to detect the start and extent of urea hydrolysis. Over 95% of ammonium nitrogen and over 85% of phosphorus from hydrolyzed urine as MAP precipitate were obtained using $MgCl_2 \cdot 6H_2O$ and $Na_2HPO_4 \cdot 12H_2O$ as precipitation agents at pH 8.5, molar ratio of Mg^{2+}:NH_4^+-N:PO_4^{3-}-P at (1.2–1.3):1:1, mixing speed of 120 r/min, and precipitation time and reaction time of 3 h and 15 min, respectively. The precipitate has a structure resembling pure MAP crystal.

Key words: urea hydrolysis; human urine; magnesium ammonium phosphate (MAP); ammonium-nitrogen (NH_4^+-N); phosphorus

Introduction

Nutrient removal from wastewater has gained increasing attention in wastewater treatment plants owing to the strict discharge standards to receiving waters and demand of recycling nutrients for the replenishing depleting resources such as phosphorous. The concentrations of nitrogen and phosphorous in domestic wastewater are generally low and the energy demand of nutrient recovery processes is typically high; hence, the currently applied treatment technologies aim at removing N or P without much recovery.

Human urine comprises higher concentrations of sodium chloride, urea, phosphate and potassium, and trace levels of calcium, sulfate and magnesium (Larsen and Gujer, 1996). The pH value of fresh urine ranges 5.6–6.8 (Fittschen and Hahn, 1998). Human urine contributes less than 1% volume of municipal wastewater quantity, but contributes 80% of N, 50% of P, and 90% of K in municipal wastewater (Larsen *et al*., 2001). Moreover, since the specific energy consumption rates are high for nutrient removal (45×10^6 J/kgN and 49×10^6 J/kgP) (Maurer *et al*., 2003), and source-separation of urine could significantly improve effluent quality and reduce energy, as well as investment costs of the receiving wastewater treatment plants (WWTPs) (Wilsenach and Van Loosdrecht, 2003, 2004), the collected human urine itself presents a concentrated source of nutrients for recovery at low energy demand. In addition, the source-separated human urine prevents its incorporated heavy metals and certain micro-pollutants, such as pharmaceutics and hormones to "pollute" the main stream of municipal wastewater (Jönsson *et al*., 1997; Larsen and Udert, 1999; Pronk *et al*., 2006). Thus urine source-separation was regarded beneficial to wastewater treatment practice and nutrient recovery, although numerous technical drawbacks existed to date (Lind *et al*., 2000; Pahl-Wosti *et al*., 2003; Udert *et al*., 2003a; Ban and Dave, 2004). The use of "NoMix" toilets is an example for implementing urine source-separation concept in Sweden (Hanæus *et al*., 1997).

The source-separated urine needs treatments or final disposal options, such as recovery of nitrogen and phosphorus through struvite crystallization and adsorption (Lind *et al*., 2000; Ban and Dave, 2004), autotrophic denitrification by ANAMMOX bacteria (Udert *et al*., 2003b), removal of micro-pollutants by nanofiltration (Pronk *et al*., 2006), direct utilization in urban agriculture (Kirchmann and Pettersson, 1995), volume reduction and concentration by freezing (Lind *et al*., 2001), storage by acidification (Hellström *et al*., 1999), and ammonia stripping by air (Basakcilardan-Kabakci *et al*., 2007). The magnesium ammonium phosphate (MAP, mineralogically as struvite) crystallization and precipitation process can recover

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ammonium nitrogen and phosphorous simultaneously to produce MAP as slow-release fertilizer (Li and Zhao, 2003) through the following chemical reaction:

$$Mg^{2+} + NH_4^+ + PO_4^{3-} + 6H_2O \longrightarrow MgNH_4PO_4{\cdot}6H_2O \downarrow \quad (pK_s = 12.6, 25°C) \tag{1}$$

Urea should be completely hydrolyzed to NH_4^+ before the precipitation reaction in Reaction (1) can be accomplished. Urease from eucaryotic and procaryotic organisms can hydrolyze urea as follows (Mobley and Hausinger, 1989):

$$NH_2(CO)NH_2 + 2H_2O \longrightarrow NH_4^+ + HCO_3^- + NH_3 \tag{2}$$

obviously in Reaction (2), pH, NH_4^+-N and bicarbonate concentrations increase with urea hydrolysis, leading to changes in PO_4^{3-}-P concentration following hydrolysis of urea (Kirchmann and Pettersson, 1995; Fittschen and Hahn, 1998; Udert *et al*., 2003a). Udert *et al*. (2003a) reported that urea could be completely hydrolyzed in the collection tank in one day if urease was added with sufficient mixing and the hydrolysis temperature was maintained at 25°C. However, existing related works in the published report did not comprehensively reveal the relationship between hydrolysis time for urine under different initial pH, temperatures, and volumetric ratios of urine (Udert *et al*., 2003a; Kabdaşli *et al*., 2006) and the optimal operational conditions for the subsequent MAP process (Lind *et al*., 2000; Li and Zhao, 2003; Ban and Dave, 2004). The objective of this study was to determine the final hydrolysis times of the separated urine at different initial conditions and to provide the optimal operational conditions for the subsequent MAP process with the aid of orthogonal experiment.

1 Materials and methods

1.1 Urine samples

The human urines were collected using cleaned plastic barrels from 5–8 healthy males aged between 25 and 34 (the fresh urine). Two fresh urine samples (S#1, S#2) were tested in hydrolysis experiments. The initial pH, NH_4^+-N and PO_4^{3-}-P concentrations for S#1 were 6.71, 755, and 171 mg/L, and for S#2 were 6.78, 624, and 161 mg/L, respectively. The completely hydrolyzed urine was named as the stale urine (SU). The SU with respective pH, NH_4^+-N and PO_4^{3-}-P concentrations of 9.35, 7,220, and 206 mg/L was tested in the MAP precipitation experiments.

1.2 Urea hydrolysis experiments

The 500 ml urine sample (S#1) was placed equally in five beakers. One beaker filled with fresh urine with no added chemicals was used as the control. The pH values of filled urine in the remaining four beakers were adjusted to 8–11 using 10 mol/L NaOH. Then the beakers were placed steadily to simulate the conditions in urine collecting tank at temperature of 20–22°C. The supernatants were collected to measure the ammonium nitrogen concentrations and pH values at 12 h intervals.

The urine following 15 d test had been completely hydrolyzed and was the SU samples herein. Another batch of samples was prepared by mixing fresh (S#2) and the SU samples at different volumetric ratios (Table 1). The mixed samples were placed in 500 ml glass covered bottles, standing still at 18–20°C with regular supernatant sample collection. Part of the mixed samples at volumetric ratio of 10% was tested at 2.5, 10, 15, 20, 25, and 30°C, respectively.

Table 1 Volumetric ratio of stale urine (SU) to fresh urine (S#2) in urea hydrolysis experiment

Item	SU				
	0	5%	10%	15%	20%
Stale urine volume (ml)	0	25	50	75	100
S#2 volume (ml)	500	475	450	425	400
Total volume (ml)	500	500	500	500	500
Initial pH	6.78	8.49	8.84	8.93	9.01
NH_4^+-N concentration (mg/L)	624	1,025	1,373	1,697	2,027

1.3 MAP precipitation experiments

The MAP precipitation tests were carried out in 1,000 ml beakers with analytical grade $MgCl_2{\cdot}6H_2O$ and $Na_2HPO_4{\cdot}12H_2O$ as precipitation agents. 10 mol/L NaOH solution was used to adjust pH. The pH, NH_4^+-N and PO_4^{3-}-P of supernatants collected following MAP precipitation tests was conducted using L_{16} ($4^4 \times 2^3$) orthogonal table (Table 2). The precipitate containing MAP products were filtered through 0.45 μm filter film, then were dried at below 60°C before analysis (Ando *et al*., 1968).

1.4 Analytical methods

The suspension pH was measured using PHS-3C type pH meter (Shanghai Hongyi Co. Ltd., China). The NH_4^+-N and PO_4^{3-}-P concentrations were analyzed according to the Standard Methods (APHA, 1995). A definite amount of 0.1500 g drying precipitate harvested from tests was weighed accurately and dissolved completely in 500 ml 1+19 ($V + V$) hydrochloric acid (i.e., its concentration was about 0.6 mol/L), respectively. Total P and metals including K, Na, Ca, Ni, Mn, and Mg of the harvested precipitate solutions were analyzed by ICP (OES Optima 5300DV, America PE, USA), and total N by TNM-1 (TOC-VCPN TN unit, Shimadzu, Japan). The minerals in the precipitates were identified by X-ray Diffraction (XRD, D/max-Rb, Japan).

2 Results and discussion

2.1 Urea hydrolysis at different initial pH (S#1)

Alkaline condition prefers struvite crystallization. If urea hydrolysis could be achieved at alkaline conditions, no further pH adjustment might be needed in the subsequent MAP precipitation process. A steady pH range of 8.9–9.3 could be reached following 84 h of urea hydrolysis

Table 2 Factor and its level in orthogonal design for magnesium ammonium phosphate (MAP) precipitation

Level	Factor					
	pH	Mg:N (mol:mol)	P:N (mol:mol)	θ (h)	v (r/min)	t (min)
1	8.0	1.0 : 1	1.0 : 1	1	60	15
2	8.5	1.1 : 1	1.05 : 1	2	120	30
3	9.5	1.2 : 1	1.1 : 1	3		
4	10.0	1.3 : 1	1.15 : 1	4		

θ: precipitation time; v: mixing speed; t: reaction time.

in all tested cases (Fig.1a). The NH_4^+-N concentration first increased but slowly decreased with hydrolysis in these tests (Fig.1b). For fresh urine (control), the pH became steady after 60 h while the highest NH_4^+-N concentration appeared at 72 h of test. The samples at higher initial pH needed longer hydrolysis time to reach steady state at higher NH_4^+-N concentration compared with the control. For example, at an initial pH of 8 or 10, the highest NH_4^+-N concentration appeared at a hydrolysis time of 96 or 108 h, respectively. The reason of higher pH affecting the urea hydrolysis obviously was that the urease activity was very low at higher pH, because the urea hydrolyzing bacteria died more quickly at pH 8.9 and 10.5 than at pH 6.0 (Höglund *et al*., 1998). Restated, the alkalescence hydrolysis effects on separated human urine were insignificant.

Both ammonium and bicarbonate ions are produced during urea hydrolysis (Reaction (2)). Two equilibriums in stale urine exist as follows:

$$NH_4^+ + OH^- \longleftrightarrow NH_3(aq) + H_2O \quad (pK = 9.24, 25°C) \tag{3}$$

$$CO_3^{2-} + H^+ \longleftrightarrow HCO_3^- + H^+ \longleftrightarrow H_2CO_3 \quad (pK_1 = 10.33, pK_2 = 6.35, 25°C) \tag{4}$$

further, phosphate ions in stale urine could achieve chemical equilibrium as follows:

$$PO_4^{3-} + H^+ \longleftrightarrow HPO_4^{2-} + H^+ \longleftrightarrow H_2PO_4^- \quad (pK_1 = 12.35, pK_2 = 7.21, 25°C) \tag{5}$$

thus, the stale urine has strong buffer capacity to the added alkali or acid. However, the fresh urine has weaker buffer action than stale urine owing to its lower concentrations of ammonium, bicarbonate, and phosphate ions (Fig.1a). Hellström *et al*. (1999) also noted a final pH of 9.0 with an initial pH of urine adjusted to less than 2.0. The final pH of source-separated urine would be all around 9.0 regardless of initial suspension pH owing to the strong buffer effects on the stale urine.

2.2 Urea hydrolysis with different volumetric ratio of stale to fresh urine

Source-separated urine was generally collected in a tank and urea was decomposed during storage. When fresh urine was mixed with stale urine, urea hydrolysis would occur with the function of the available urease in a short time (Mobley and Hausinger, 1989). The pH, NH_4^+-N and PO_4^{3-}-P changes following urea hydrolysis at different volumetric ratios of stale to fresh urines as illustrated in Fig.2.

Urea hydrolysis was enhanced in mixed urines (Fig.2) compared with those with fresh urines. For instance, with fresh urine (0%, volumetric ratio), the pH and NH_4^+-N concentration became steady at 72 and 84 h, respectively. At a volumetric ratio of 20%, the corresponding time became 36 h (Figs.2a and 2b). It means that the presence of residual stale urine in the collecting tank significantly decreases the hydrolysis time of urea. The hydrolysis time of urea by adding stale urine was longer than that by adding urease which only needed 1.5 h (Kabdaşli *et al*., 2006), but the former was more economical and convenient in practice.

During the hydrolysis experiment, the PO_4^{3-}-P concentration rapidly decreased at 12 h and was steady with the exception of the control experiment (0%, SU). The changes

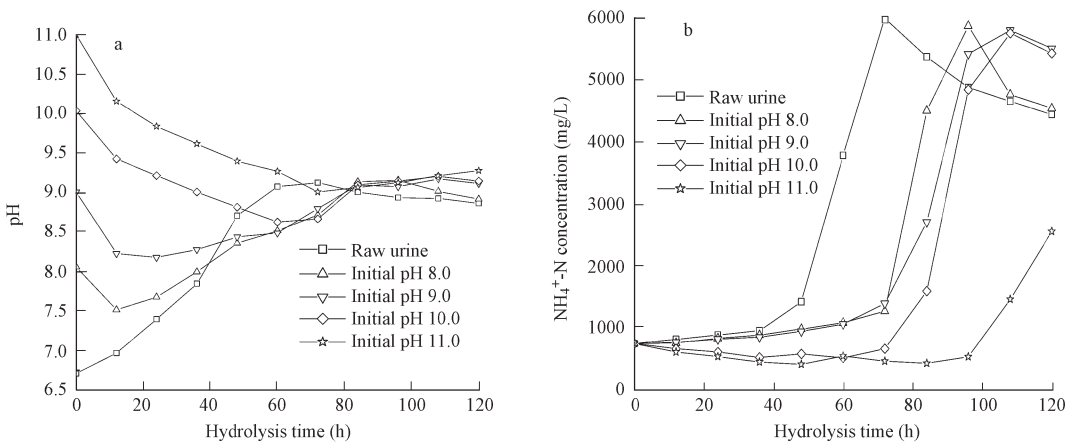

Fig. 1 Urine pH (a) and NH_4^+-N concentration (b) change with urea hydrolysis time at different initial pH.

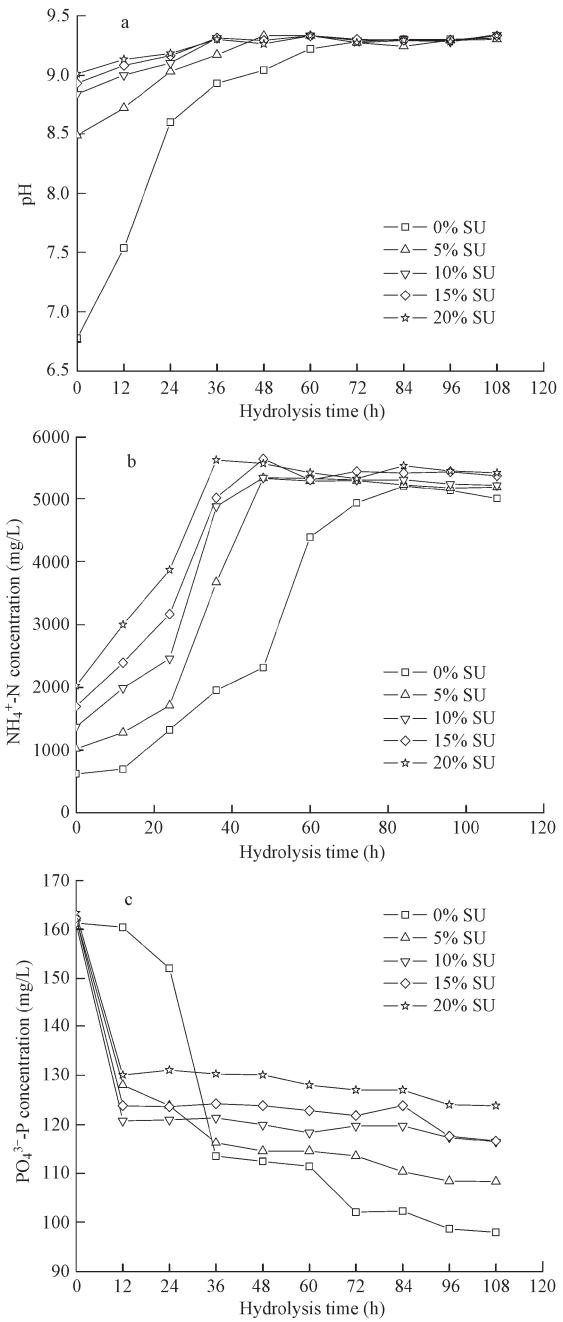

Fig. 2 Urine pH (a), NH_4^+-N concentration (b), and PO_4^{3-}-P concentration (c) change with hydrolysis time at different volumetric ratio of stale urine (SU).

in PO_4^{3-}-P concentration following hydrolysis was smaller than those in NH_4^+-N concentration. Udert *et al.* (2003c) noted that MAP and hydroxyapatite (HAP) are the sole minerals in undiluted urine. The HAP reaction can be stated as follows:

$$3PO_4^{3-} + 5Ca^{2+} + OH^- \longleftrightarrow Ca_5(OH)(PO_4)_3 \quad (p\mathit{K}sp = 57.5, 25°C) \tag{6}$$

restated, both MAP and HAP reactions would mainly take place in alkaline condition, particularly Reaction (1) occurs at pH > 8 for satisfactory MAP precipitation (Li and Zhao, 2003; Adnan *et al.*, 2003). In urine, both Ca^{2+} and Mg^{2+} are deficient compared to NH_4^+-N and PO_4^{3-} according to Reactions (1) and (6) (Udert *et al.*, 2003c; Katarzyna and Grietje, 2006). Therefore, when the pH was increased to 8.0 or above, the PO_4^{3-}-P concentration in urine decreased sharply by MAP and HAP precipitation, and then became steady due to the deficiency of Ca^{2+} and Mg^{2+} (Fig.2c).

2.3 Urea hydrolysis at different temperatures

High temperature prefers urea hydrolysis (Fig.3). For instance, at 30°C the highest NH_4^+-N concentration could be reached at 36 h, meanwhile, the NH_4^+-N concentration was only 40% or less of the highest value at temperature < 15°C (Fig.3b). At temperature > 20°C, the corresponding

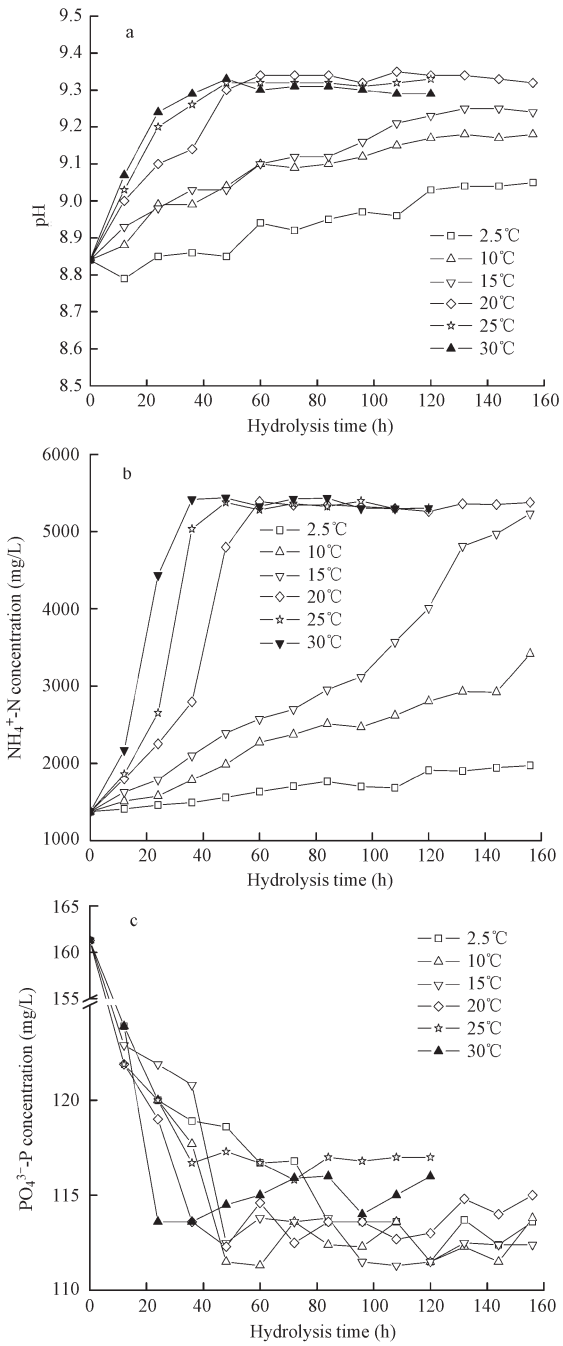

Fig. 3 Urine pH (a), NH_4^+-N concentration (b), and PO_4^{3-}-P concentration (c) change with hydrolysis time at different temperature.

urea hydrolysis time would not be decreased further. In other words, the urea could be hydrolyzed within 60 h when the volumetric ratio of stale urine to fresh urine exceeded 10% at temperature > 20°C. The PO_4^{3-}-P concentrations rapidly decreased at 12 h of hydrolysis when the temperature was above 15°C, while at temperature < 15°C, the change rates in PO_4^{3-}-P concentration declined slowly, and reached plateau values at > 48 h (Fig.3c).

Initial suspension pH affected the highest NH_4^+-N concentration achievable in hydrolysis. However, the ammonia loss owing to evaporation should be considered in interpreting these data since the tests were conducted in open beakers (Fig.1b). Sealing should be applied in the source-separated urine collection tank to minimize ammonia loss (Hanæus *et al*., 1997).

In the experiments, the hydrolysis time and the average pH and PO_4^{3-}-P concentration of fresh urine were 6.7–6.8 and 161–171 mg/L, respectively, slightly different from those reported in the published report (Udert *et al*., 2003c). These differences may be yielded by the fact that the present study collected fresh urine using barrels without pre-disinfection, for simulating practical urinal flush, thus certain urease pollution may occur.

In the present tests, suspension pH closely corresponded to the noted NH_4^+-N concentration. Hence, the suspension pH, which was easily measured during experiments, may be used as an indicator to the extent of urea hydrolysis if the long-lasting period with slow hydrolysis was disregarded (comparing Fig.1a with Fig.1b, Fig.2a with Fig.2b, and Fig.3a with Fig.3b).

2.4 MAP precipitation tests

The orthogonal table L_{16} ($4^4 \times 2^3$) was designed and 16 groups of experiments were conducted with the results shown in Table 3. The molar ratio of Mg:N was the most significant, while the mixing speed was the least factor affecting the NH_4^+-N and PO_4^{3-}-P recovery rates. The best results amongst the sixteenth experiments yielded residual NH_4^+-N and PO_4^{3-}-P concentrations of 87.5 and 8.34 mg/L, respectively. Taking the economic aspect into consideration as well, the optimum reaction pH, Mg:N (mol:mol), P:N (mol:mol), θ, v, and t were 8.5, (1.2–1.3):1, 1:1, 3 h, 120 r/min, and 15 min, respectively.

To validate the results of the orthogonal experiments, two further tests with respective Mg:N:P (mol:mol:mol) of 1.2:1:1 and 1.3:1:1 were conducted with all other conditions fixed at those determined at optimum. The residual NH_4^+-N and PO_4^{3-}-P concentrations were 251 and 25 mg/L (1.2:1:1), and 318 and 9.0 mg/L (1.3:1:1), respectively. Hence, the MAP precipitation could recover most NH_4^+-N and PO_4^{3-}-P from stale human urine, and the NH_4^+-N and PO_4^{3-}-P recovery efficiency were above 95% and 85%, respectively. Moreover, the two precipitates with the presence of trace calcium, potassium and sodium collected revealed similar chemical structure to pure MAP (Table 4). The XRD analysis also confirmed that the precipitates were mostly pure $MgNH_4PO_4 \cdot 6H_2O$ (Fig.4).

Since the NH_4^+-N concentration is higher than PO_4^{3-}-P concentration in stale urine, the higher NH_4^+-N removal rate could only be obtained at more chemical doses of $MgCl_2 \cdot 6H_2O$ and $Na_2HPO_4 \cdot 12H_2O$ with NaOH. Such an

Table 3 L_{16} ($4^4 \times 2^3$) orthogonal design for MAP precipitation

Sample	Factor						Residual	
	pH	Mg:N (mol:mol)	P:N (mol:mol)	θ (h)	v (r/min)	t (min)	NH_4^+-N (mg/L)	PO_4^3-P (mg/L)
1	8.0	1.1:1	1.1:1	2	120	15	386	969.42
2	9.5	1.3:1	1:1	2	60	30	628	15.7
3	8.5	1.3:1	1.1:1	3	120	30	115	13.2
4	10.0	1.1:1	1:1	3	60	15	438.5	11.2
5	8.0	1.2:1	1:1	4	120	30	546.5	6.78
6	9.5	1:1	1.1:1	4	60	15	614	1,779.2
7	8.5	1:1	1:1	1	120	15	815	1,483.56
8	10.0	1.2:1	1.1:1	1	60	30	227	16.97
9	8.0	1:1	1.15:1	3	60	30	791	2,434.13
10	9.5	1.2:1	1.05:1	3	120	15	245	7.2
11	8.5	1.2:1	1.15:1	2	60	15	68.5	147.32
12	10.0	1:1	1.05:1	2	120	30	708.5	1,560.28
13	8.0	1.3:1	1.05:1	1	60	15	489	18.76
14	9.5	1.1:1	1.15:1	1	120	30	261	1,686.19
15	8.5	1.1:1	1.05:1	4	60	30	323.5	560.4
16	10.0	1.3:1	1.15:1	4	120	15	87.5	8.34
Mean of residual NH_4^+-N								
KN1	553.1	732.1	607	448	447.4	392.9		
KN2	330.5	352.3	441.5	447.8	395.6	450.1		
KN3	437	271.8	335.5	397.4				
KN4	365.4	329.9	302	392.9				
RN	222.6	460.3	305	55.1	51.8	57.2		
Mean of residual PO_4^{3-}-P								
KP1	857.3	1814.3	379.3	801.4	623	553.1		
KP2	551.1	806.8	536.7	673.2	716.9	786.7		
KP3	872.1	44.6	694.7	616.4				
KP4	399.2	14	1,069	588.7				
RP	472.9	1,800.3	689.7	212.7	93.9	233.6		

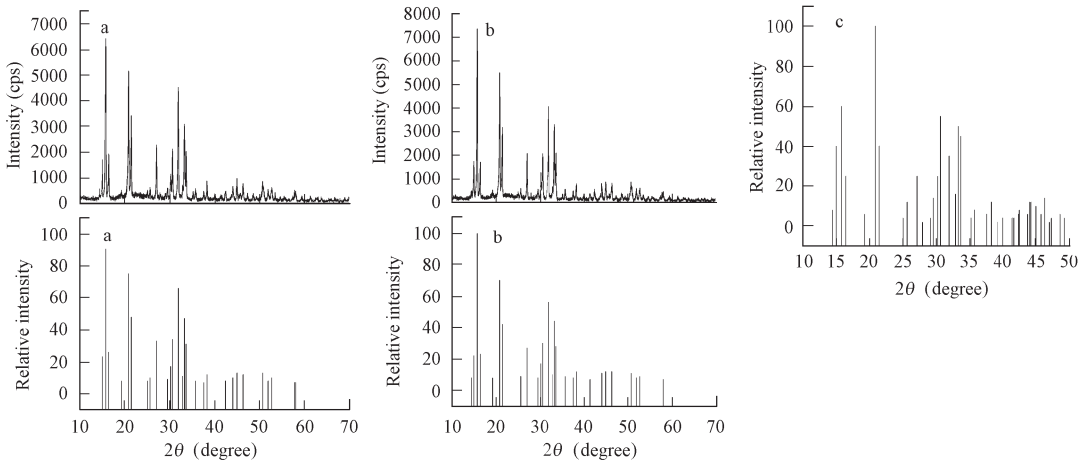

Fig. 4 X-ray diffractographs of the precipitates and pure MAP. (a) precipitate at 1.2:1:1 molar ratio of Mg:N:P; (b) precipitate at 1.3:1:1 molar ratio of Mg:N:P; (c) standard MAP (ICDC Standard #15-0762, 1996).

Table 4 Chemical content of pure MAP and precipitates obtained under different mole ratios of Mg:N:P (%)

	Sample	P	N	Mg	Ca	K	Ni	Na	Mn
	Pure MAP	12.62	5.70	9.91	–	–	–	–	–
Precipitate	1.2:1:1 (mol:mol:mol)	13.46	4.48	8.84	0.12	0.35	< 0.001	1.08	< 0.001
	1.3:1:1 (mol:mol:mol)	13.51	4.44	8.72	0.12	0.31	< 0.001	1.41	< 0.001

occurrence increases operational cost of MAP process. Most phosphorous and part of the NH_4^+-N could be recovered from urine using MAP process without adding with $Na_2HPO_4 \cdot 12H_2O$ and NaOH, which may be a compromise between high nutrient recovery and low operation cost aspects.

3 Conclusions

This study investigated the urea hydrolysis of source-separated human urine and following MAP precipitation process to recover nitrogen and phosphorous as a resource. High pH and low temperature of suspension inhibited urea hydrolysis, while the presence of stale urine assisted this process. Specifically, the urea hydrolysis could be delayed in more than 1 d at initial pH > 8. At temperature > 20°C and a volumetric ratio of stale to fresh urine >10%, the hydrolysis time would be limited in 2 d; while at < 15°C the corresponding hydrolysis time would exceed 6.5 d (156 h).

The pH of urine increased and stabilized at around 9.0 following hydrolysis, which may be used as an indicator to the extent of urea hydrolysis. The PO_4^{3-}-P concentration was rapidly decreased at pH > 8, but would reach steady state at deficient supply of Ca^{2+} and Mg^{2+} ions.

The recovery efficiency of NH_4^+-N and PO_4^{3-}-P using MAP process from stale urine could reach above 95% and 85%, respectively, at the optimal operational conditions identified as follows: molar ratio of Mg^{2+}:NH_4^+-N:PO_4^{3-}-P of (1.2–1.3):1:1, pH 8.5, precipitation time of 3.0 h, mixing speed of 120 r/min, and reaction time of 15 min. The chemical structures of the MAP precipitates were similar to those of the pure MAP crystals, which could be used as a slow-release fertilizer.

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