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# Waste activated sludge hydrolysis and acidification: A comparison between sodium hydroxide and steel slag addition

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## ABSTRACT

Alkaline treatment with steel slag and NaOH addition were investigated under different pH conditions for the fermentation of waste activated sludge. Better performance was achieved in steel slag addition scenarios for both sludge hydrolysis and acidification. More solubilization of organic matters and much production of higher VFA (volatile fatty acid) in a shorter time can be achieved at pH 10 when adjusted by steel slag. Higher enzyme activities were also observed in steel slag addition scenarios under the same pH conditions. Phosphorus concentration in the supernatant increased with fermentation time and pH in NaOH addition scenarios, while in contrast most phosphorus was released and captured by steel slag simultaneously in steel slag addition scenarios. These results suggest that steel slag can be used as a substitute for NaOH in sludge alkaline treatment.

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## Introduction

The number of wastewater treatment plants (WWTPs) worldwide has increased dramatically in recent decades due to rapid urbanization and population growth, resulting in a significant increase of waste activated sludge (WAS) production. Typically, WAS contains a large amount of organics, heavy metals and pathogens, which pose great threats to the environment, and its disposal cost can account for 20%–60% of the capital and operating cost of a WWTP (Fang et al., 2014; Su et al., 2013). Therefore, environmentally friendly and cost-effective means for sludge disposal are greatly needed.

Anaerobic digestion is one of the most widely used methods for activated sludge stabilization. Worldwide attention has been drawn to this technique due to its capability of simultaneously

achieving sludge mass reduction and generating valuable products, including methane, which is regarded as an important energy source, and volatile fatty acid (VFA), which could be used as carbon sources to enhance biological nutrient removal in WWTPs (Chen et al., 2007; Su et al., 2013). However, the application of anaerobic digestion has often been featured by long retention times (20–50 days), large digester volumes and low overall degradation efficiency (20%–50%) (Kim et al., 2010). Among the three well-known steps involved in anaerobic digestion, the hydrolysis step has been identified as the rate-limiting step (Higgins and Novak, 1997). Also, the major reasons for low digestibility are microbial cell walls containing glycan strands cross-linked by peptide chains and the existence of a large amount of extracellular polymeric substances (EPS) (Devlin et al., 2011). In order to improve the hydrolysis efficiency

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and digestibility of sludge, various pretreatment methods, also referred to as sludge disintegration techniques, have been investigated, including chemical treatment using alkali, acid or ozone (Devlin et al., 2011; Li et al., 2012; Zhang et al., 2009), mechanical treatment with ultrasound, high pressure homogenization or ball milling (Apul and Sanin, 2010; Wett et al., 2010; Zhang et al., 2012), thermal treatment (Xue et al., 2015), treatment with additional enzymes (Yang et al., 2010) and some of their combinations (Kim et al., 2010; Shehu et al., 2012). These pretreatment methods can normally destroy sludge flocs and cell walls, release intracellular and extracellular organic matters, and consequently accelerate sludge hydrolysis.

Alkaline treatment is a widely examined sludge disintegration method which is believed to be very effective in terms of solubilizing cellular substances and EPS (Fang et al., 2014; Li et al., 2008). Compared to other methods, alkaline treatment holds the advantages of simpler equipment, easy operation and lower energy demand. In addition, alkaline conditions are particularly favorable to the production of short-chain fatty acids or VFA, which are suitable carbon sources for biological nutrient removal processes and phosphorus recovery with MAP (magnesium ammonium phosphate hexahydrate) or HAP (calcium phosphates) (Tong and Chen, 2007). NaOH is the most commonly used chemical for alkaline treatment and has been found to be more effective in terms of sludge disintegration than lime ( $\text{Ca}(\text{OH})_2$ ) (Li et al., 2008). However, the consumption of NaOH involves higher chemical cost and thus will increase sludge treatment expense. New alternative chemicals with low cost must be further explored.

Steel slag is a porous non-metallic by-product produced in the steelmaking industry, the amount of which can be equivalent to 10%–20% of crude steel output. Every year there is around 100 million tons of steel slag generated in China, while only 50%–60% of it can be reutilized in proper ways, leaving a large amount of slag piled at the steel plants, which poses a great threat to groundwater quality. Although the chemical composition of steel slag varies with different slag production processes, normally it consists of  $\text{CaO}$ ,  $\text{SiO}_2$ ,  $\text{FeO}$ ,  $\text{MgO}$ ,  $\text{Al}_2\text{O}_3$ ,  $\text{MnO}$ ,  $\text{P}_2\text{O}_5$ , etc. (Shi, 2004). Steel slag has been used for treating acid mine drainage, since its alkaline compounds such as lime and magnesia can leach out, creating high levels of alkalinity (Simmons et al., 2002). Meanwhile, steel slag was found to be a very effective material for phosphorus removal in wastewater as a result of phosphorus adsorption onto metal oxides/oxyhydroxides throughout the porous slag matrix and on the slag surface, and precipitation in forms of metal phosphates such as Fe-phosphates and Ca-phosphates (Pratt et al., 2007). In the literature, however, the enhancement of sludge hydrolysis and acidification by steel slag has never been studied, although the capability of creating alkalinity and phosphorus capture make it a potentially useful chemical for sludge alkaline treatment.

Therefore, the purpose of this study was to investigate the feasibility of steel slag addition as a substitute of NaOH for the purpose of sludge hydrolysis and acidification enhancement and phosphorus recovery. As pH is a key parameter in alkaline or acid treatment, the effect of steel slag addition on WAS hydrolysis and acidification for different pH conditions was studied in comparison with NaOH addition scenarios.

## 1. Materials and methods

### 1.1. Sludge and steel slag

The waste activated sludge used in this study was collected from the secondary sedimentation tank of a municipal wastewater treatment plant in Shanghai, China. The plant is operated with an enhanced biological phosphorus removal process. The sludge was concentrated by settling at 4°C for 12 hr and was measured afterward (Table 1).

The steel slag used in this work was obtained from Baoshan Iron & Steel Co. Ltd. in Shanghai, China. The steel slag was crushed and sifted through a 50-mesh sieve before being added into the WAS. Elemental composition analysis with EDS (energy dispersive spectroscopy) showed the elemental composition (expressed as atomic percentage) of the steel slag to be O 33.7%, Fe 22.4%, C 15.6%, Ca 10.9%, Mg 6.5%, Mn 3.2%, Au 2.4%, Si 2.3%, etc.

### 1.2. Experimental procedure

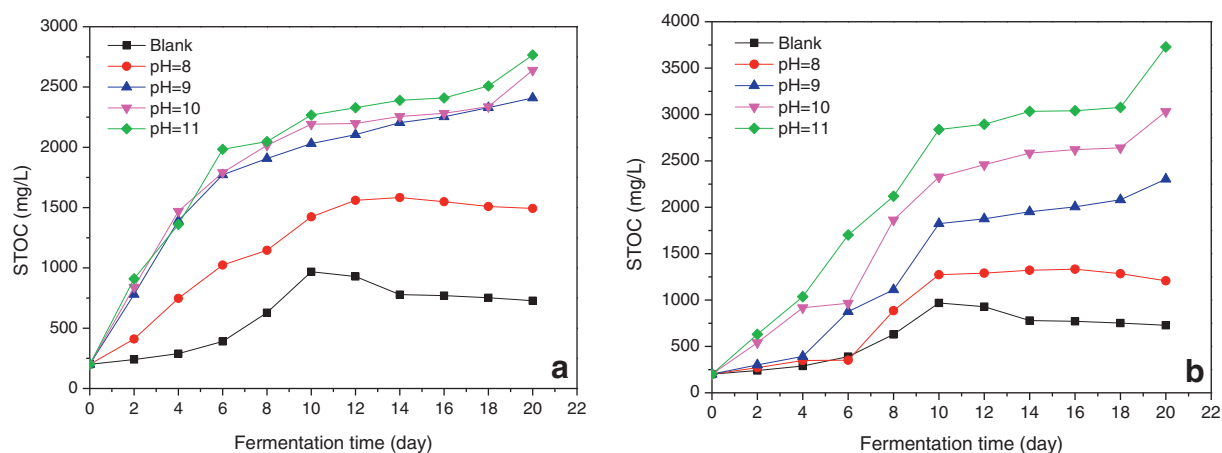
Nine 1-L glass vessels with working volume of 0.9 L were utilized as the anaerobic fermentation reactors. A volume of 0.9 L of WAS were fed into each reactor. The pH in reactors 1–4 was controlled at 8.0, 9.0, 10.0 and 11.0 respectively, with steel slag and the same pH was established in reactors 5–8 with sodium hydroxide. Reactor 9, in which the pH was not adjusted, was set as the blank test. After 5 min nitrogen blowing, all reactors were placed in a shaking incubator maintained at the temperature of  $25 \pm 1^\circ\text{C}$  with a shaking speed of 100 r/min. Samples of the mixed liquor were taken at intervals. Supernatant samples were obtained after 3 min settling of the mixed liquor.

### 1.3. Analytical methods

Water content, TS (total solids), VS (volatile solids), and COD (chemical oxygen demand) were determined according to the standard methods (APHA et al., 2012). Sludge pH was measured with a pH meter (EUTECH Cyberscan510, Singapore).  $\text{PO}_4^{3-}\text{-P}$  and total phosphorus (TP) of the supernatant were also analyzed according to the standard methods (APHA et al., 2012). TP in steel slag was determined by the Capacity Method of Ammonium Phosphomolybdate (Song, 2007), while TP in the sludge was measured by the Vanadium Molybdate Yellow Colorimetric Method after Microwave Digestion (Wen et al., 2003). Soluble carbohydrate was determined using the phenol-sulfuric acid method (Miller, 1959). Soluble protein was determined by the Lowry-Folin method with BSA (Bovine Serum Albumin) as standard (Lowry et al., 1951).

**Table 1 – Characteristics of sludge.**

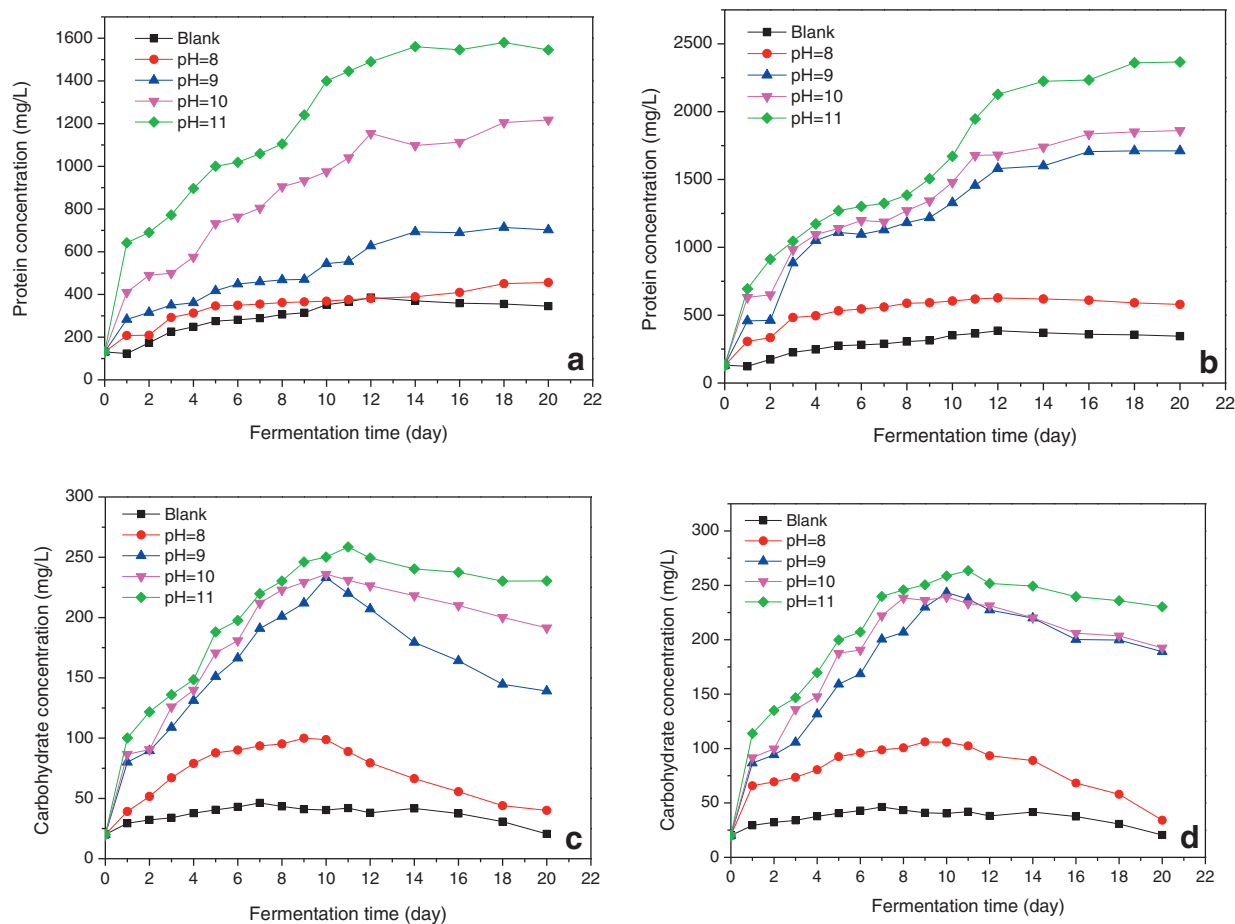
Parameter	Value
pH	6.8–7.0
Water content (%)	97–98
Total solids (TS, g/L)	13–21
Volatile solids (VS, g/L)	6–14
Total chemical oxygen demand (TCOD, mg/L)	14,000–20,000
Soluble chemical oxygen demand (SCOD, mg/L)	40–100



**Fig. 1 – Variations of organic matter solubilization at different pH values adjusted by NaOH (a) and steel slag (b). STOC: soluble total organic carbon.**

Total organic carbon (TOC) was determined using a TOC-VCPN Analyzer (Shimadzu, Tokyo). An Agilent 6890N GC with flame ionization detector equipped with a 30 m  $\times$  0.32 mm  $\times$  0.25  $\mu$ m CPWAX52CB column was utilized to analyze the composition of VFAs. The analytical method

used in the analysis was developed according to previous publication (Chen et al., 2007). The microstructures and elemental composition of the steel slag were examined by scanning electron microscopy equipped with energy dispersive X-ray spectroscopy (SEM-EDX, S-4800, Hitachi Co., Japan).



**Fig. 2 – Variations of protein and carbohydrate concentration in supernatant at different pH values adjusted by NaOH and steel slag. (a) and (b) refer to the variation of protein concentration for NAC (NaOH-adjusted conditions) and SSAC (steel slag-adjusted conditions); (c) and (d) refer to the variation of carbohydrate concentration for NAC and SSAC, respectively.**

Enzyme activities of four hydrolytic enzymes including protease,  $\alpha$ -glucosidase, alkaline and acid phosphatase were measured as per the methods introduced by Nybroe et al. (1992). Activities of five enzymes involved in acid fermentation were examined. The measurement of acetate kinase (AK) and butyrate kinase (BK) activity were carried out as per the method of Allen et al. (1964); while analyses of phosphate transacetylase (PTA) and phosphate transbutyrylase (PTB) were carried out as per Andersch et al. (1983); oxaloacetate transcarboxylase (OATC) activity was assessed as per Wood et al. (1969).

## 2. Results and discussion

### 2.1. Sludge disintegration

Alkaline disintegration of WAS accelerates hydrolysis by disruption of flocs and sludge cells, which leads to the release and solubilization of extracellular and intracellular organic matters (Doğan and Sanin, 2009; Fang et al., 2014; Li et al.,

2008). Thus, the effect of steel slag and NaOH addition on sludge disintegration can be expressed by the change of STOC (soluble total organic carbon), which is determined by measuring the TOC of the supernatant. Fig. 1 shows the effect of steel slag and NaOH addition on STOC during WAS fermentation within 20 days at different pH values. It was found that the concentration of STOC under both NaOH- and steel slag-adjusted conditions (the following abbreviations were used: NAC, for NaOH-adjusted conditions; SSAC for steel slag-adjusted conditions) increased gradually with the fermentation time. In the selected range of pH, more solubilization of organics was observed at higher pH, consistent with previous studies (Chen et al., 2007; Doğan and Sanin, 2009) in which SCOD was used as an indicator. It is worth noting that at higher pH values (pH 10 and 11) SSAC had a significantly higher value of STOC than that of NAC after 10 days fermentation. The STOC was 393 and 963 mg/L higher in the fermentation reactors of SSAC than that of NAC at fermentation time 20 days for pH 10 and pH 11, respectively, indicating that steel slag may be better than NaOH in the enhancement of sludge disintegration.

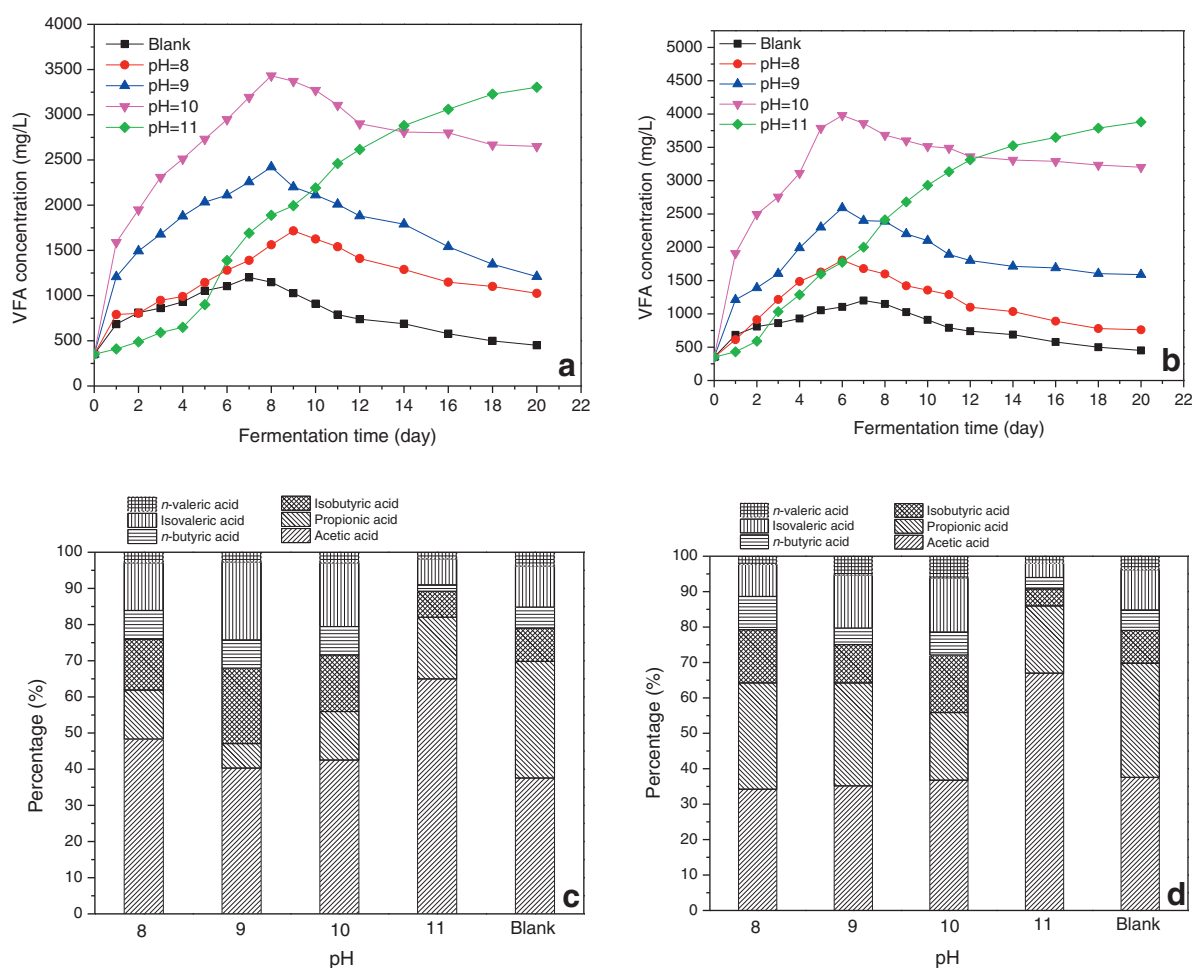
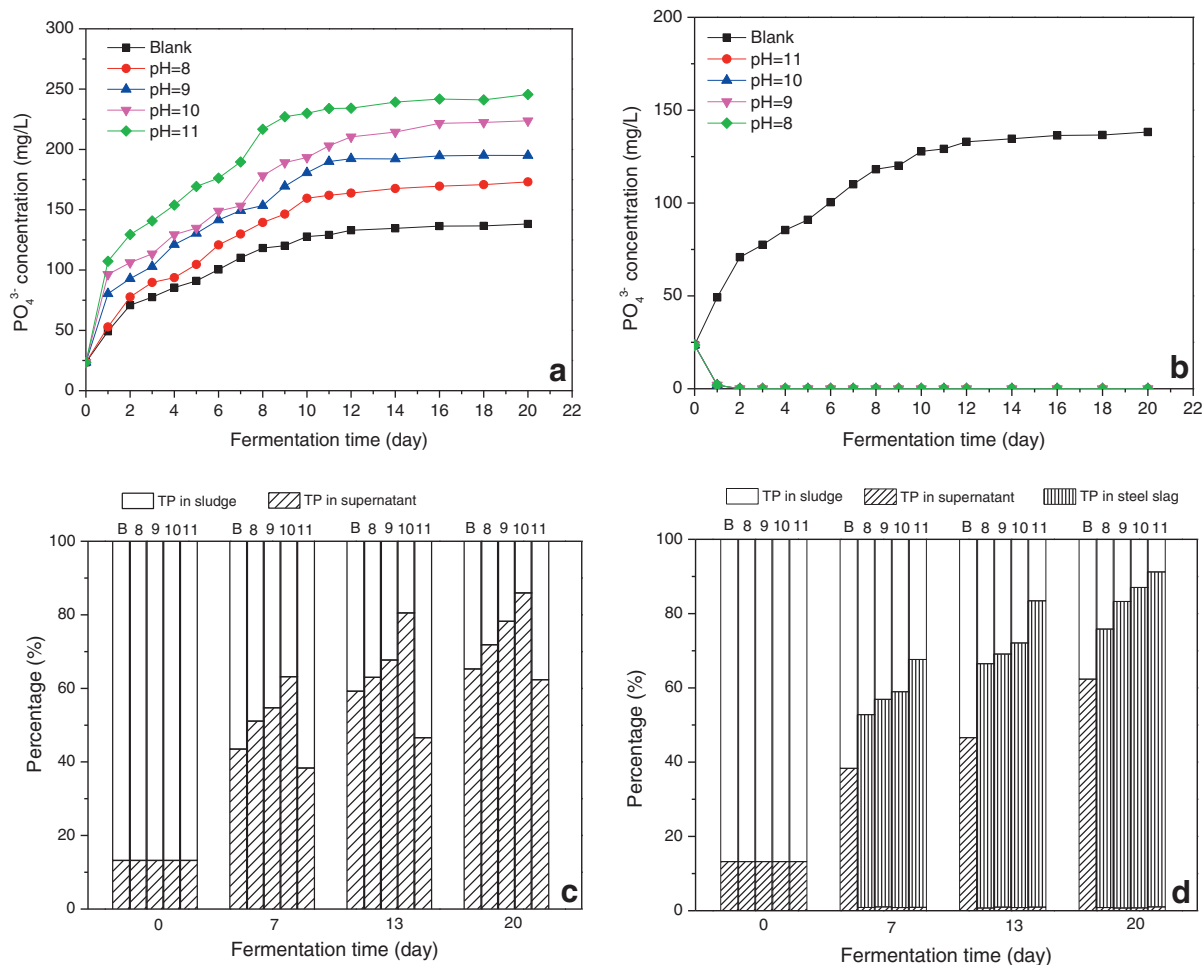


Fig. 3 – Variations of total VFA (volatile fatty acid) concentration and VFA composition in supernatant at different pH values adjusted by NaOH and steel slag. (a) and (b) show the variation of total VFA concentration for NAC and SSAC; (c) and (d) show VFA composition at highest total VFA level for NAC and SSAC, respectively.

## 2.2. Protein and carbohydrate solubilization

It has been reported that protein and carbohydrate are the predominant constituents of EPS in WAS (Chen et al., 2007; Yan et al., 2010). In addition, alkaline treatment is believed to be very effective in solubilizing EPS (Chen et al., 2007; Fang et al., 2014). Thus, the solubilization of organic matters was expressed in terms of soluble protein and carbohydrate. As shown in Fig. 2, the concentration of soluble protein and carbohydrate for both NAC and SSAC reactors increased with pH, which is in accordance with the change of STOC. In addition, the concentration at alkaline pH (pH = 9, 10, 11) was much higher than that at near-neutral pH (blank and pH = 8). This is probably because alkaline pH resulted in the dissociation of acidic groups in EPS and repulsion between the negatively charged EPS (Wingender et al., 1999). In agreement with the literature (Chen et al., 2007; Su et al., 2013), the concentrations of soluble protein and carbohydrate fluctuated slightly with the increase of fermentation time in both NAC and SSAC reactors due to the competition between solubilization and degradation. Especially for carbohydrate, the concentration showed an obvious decline after 10 days, whereas the soluble protein concentration tended to remain constant after 12 days. The rate of carbohydrate

concentration decline was lower at pH 11 than other scenarios in both NAC and SSAC reactors, which indicated that the degradation rate of carbohydrate might be reduced under strongly basic conditions. The carbohydrate concentration in SSAC was slightly higher than that of NAC. However, a remarkable difference was observed between the protein concentrations of SSAC and NAC, with the highest concentrations being 2300 mg/L and 1500 mg/L, respectively, further demonstrating that steel slag has an advantage over NaOH in terms of sludge solubilization. In SSAC, basic conditions were provided as a result of the continuous dissolution of the major metal oxides in the steel slag including CaO and MgO, which is the most important cause of steel slags' contribution to sludge disintegration and organic matter solubilization. The better performance of steel slag addition compared with NaOH addition could be attributed to two possible reasons. First, steel slags have large acid neutralizing capacities, and the dissolution of alkaline components took place gradually. Thus, more stable pH conditions could be kept in SSAC compared to the manual adjustment required with NaOH in NAC. Second, concomitant mechanical disruption of microbial cells may have taken place due to collisions between the sludge and steel slag particulates with the shaking of the reactors.



**Fig. 4 – Variation of  $\text{PO}_4^{3-}\text{-P}$  concentration in supernatant and the variation of TP (total phosphorus) distribution at different pH values adjusted by NaOH (a, c) and steel slag (b, d). In these columns, B refers to the blank test, and 8, 9, 10, and 11 refer to the scenarios under these pH conditions.**



**Table 2 – Elemental composition of steel slag before and after fermentation.**

Elements	Before fermentation		After fermentation	
	Weight percentage (%)	Atomic percentage (%)	Weight percentage (%)	Atomic percentage (%)
C	5.6	15.6	5.8	13.7
O	16.1	33.7	17.1	34.9
Na	0.6	0.9	0.3	0.6
Mg	4.7	6.5	4.1	6.7
Al	0.6	0.7	0.1	0.2
Ca	13.2	10.9	13.1	12.4
Fe	38.3	22.4	34.9	19.23
P	0.8	0.85	8.9	7.4
Si	1.9	2.3	0.7	1.1
Cr	0.4	0.3	0.2	0.1
Mn	5.3	3.2	4.5	2.1
Au	12.4	2.4	10.3	1.4

### 2.3. VFA production and composition

Total VFA production from WAS was defined as the sum of the concentrations of acetic, propionic, *n*-butyric, isobutyric, *n*-valeric and isovaleric acids (Su et al., 2013). The total VFA production at different pH values adjusted by NaOH and steel slag is shown in Fig. 3a and b. In both NAC and SSAC fermentation reactors, the VFA concentration increased with fermentation time gradually, and then decreased after reaching the maximum level around 6–9 days at all pH values except for pH 11. At pH 11, the VFA concentration kept rising during the fermentation time of 20 days, which was in accordance with the result of Chen et al. (2007), where the initial lower VFA production was explained as being due to the ineffectiveness of acidogenic bacteria as a result of the toxic effects of the strongly basic conditions, and the subsequent significant increase showed the adaptation of bacteria to the environment. The greatest VFA production was observed at pH 10, which was 3431 mg/L and 3981 mg/L for NAC and SSAC reactors, respectively. Not only did SSAC have higher VFA production, but also it took a shorter time (6 days versus 8 days) to reach the maximum level. As is well known, three stages are involved in sludge anaerobic fermentation: hydrolysis, acidification and methane generation. The better performance of steel slag addition in VFA production could be caused by either the presence of more soluble substrate for acidification due to a higher hydrolysis rate or inhibition of the activities of methanogens (Yuan et al., 2006). It was demonstrated that more soluble protein and carbohydrate were available in SSAC fermentation reactors, while methanogen activities were not examined in this study, and should be further investigated in future work.

Fig. 3c and d shows the percentage of individual VFAs out of total VFAs at different pH values when total VFAs was at the highest level. All six common short-chain fatty acids in WAS (acetic, propionic, *n*-butyric, isobutyric, *n*-valeric and isovaleric acids) were detected. In agreement with the literature (Chen et al., 2007; Su et al., 2013), acetic acid was the predominant acid, and its proportion tended to be greater at higher pH, probably owing to the inhibiting effect of stronger alkaline conditions on methanogens and the subsequent lower

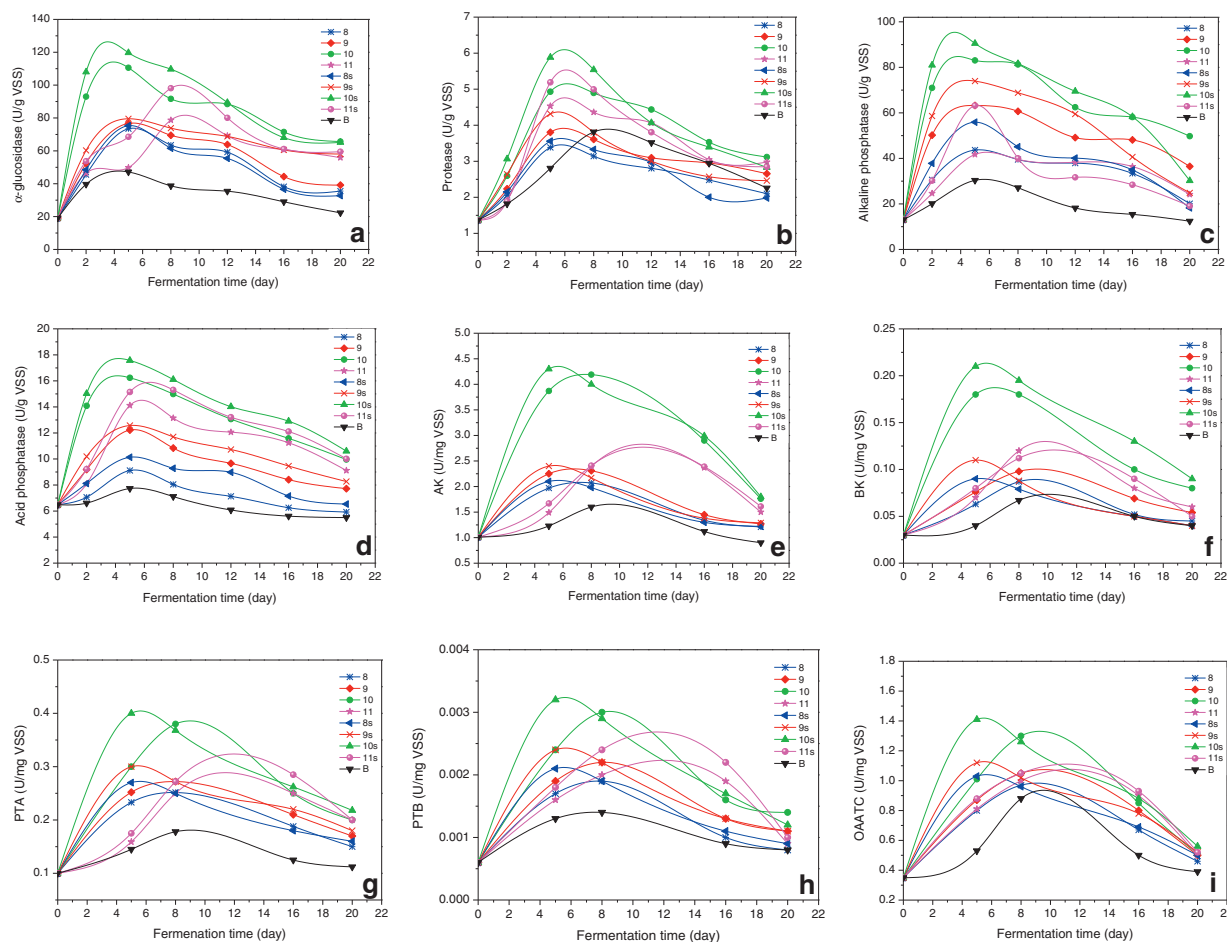
consumption of acetic acid. The order of individual VFAs was acetic > propionic > isovaleric, isobutyric > *n*-butyric, *n*-valeric. No observable difference was discovered between NAC and SSAC in terms of VFA composition.

### 2.4. $\text{PO}_4^{3-}$ release

Fig. 4a–b shows the variation of orthophosphate concentration at different pH values in NAC and SSAC fermentation reactors. In NAC reactors, it can be seen that the orthophosphate concentration increased gradually with fermentation time as well as pH. The maximum concentration observed at pH 11 after 20 days was 250 mg/L, which could be explained by stronger alkaline conditions being more effective in cell lysis with more nucleic acids released and hydrolyzed. In addition, further disruption of sludge cells at pH 11 and above may lead to disruption of cell membranes composed of phospholipid bilayers, and thus more phosphorus will be released (Bi et al., 2014). However, in SSAC reactors, almost no orthophosphate was detected after 1 day fermentation. Two possible reasons for this phenomenon are: the addition of steel slag significantly inhibited sludge phosphorus release; or the orthophosphate released into the supernatant was captured by the steel slag particles. In order to further explore these alternatives, the distribution of total phosphorus was examined (Fig. 4c–d). The proportion of TP in sludge declined along with fermentation time in both NAC and SSAC reactors, while the majority of TP migrated into steel slag rather than the supernatant in SSAC. The precipitation of  $\text{Ca-PO}_4\text{-P}$  complexes as a result of  $\text{Ca}^{2+}$  dissolution has been recognized in the literature as the dominant mechanism of phosphorus removal with steel slag under alkaline pH conditions (Bowden et al., 2009). SEM-EDX analysis was conducted to investigate the morphology and elemental composition of steel slag before and after the fermentation. Although no obvious morphology difference was found between the SEM images, an apparent phosphorus weight percentage change from 0.8% to 8.9% in the elemental composition of steel slag surface was observed (Table 2), further demonstrating that a large amount of phosphorus was transferred to the surface of steel slag. Meanwhile, the atomic and weight percentage of most of the other elements were slightly reduced as a result of the increased phosphorus. However, a very slight increase in oxygen percentage was observed, which could result from the opposing effects of  $\text{PO}_4\text{-P}$  precipitation and oxide dissolution. After fermentation for 20 days at pH 11, 91.6% TP was released and transferred to steel slag, whereas 85.0% TP was observed in the supernatant of NAC. Therefore, by steel slag addition the release of phosphorus in sludge was enhanced and simultaneously removed effectively from the supernatant. By capturing phosphorus immediately after the release of orthophosphate, the solubilization effect of phosphorus could have been enhanced in SSAC as well.

### 2.5. Enzyme activities

The major constituents of WAS such as protein and carbohydrate are all biological macromolecules. Their degradation is heavily dependent on microorganisms and enzymes. Thus, the activities of key enzymes for hydrolysis and VFA



**Fig. 5 – Variation of hydrolytic and acid-forming enzyme activities at different pH values. B refers to the blank test, and 8, 9, 10, and 11 refer to the scenarios under these pH conditions adjusted by NaOH, while 8s, 9s, 10s, and 11s refer to the scenarios under these pH conditions adjusted by steel slag. AK, BK, PTA, PTB are abbreviations of acetate kinase, butyrate kinase, phosphate transacetylase, phosphate transbutyrylase and oxaloacetate transcarboxylase, respectively.**

production were examined to further understand the effect of NaOH and steel slag addition on WAS fermentation. Four hydrolytic enzymes (protease,  $\alpha$ -glucosidase, alkaline phosphatase and acid phosphatase) and five acid-forming enzymes (AK, BK, PTA, PTB and OAATC) were selected for this study. Usually, protease breaks the peptide bonds in protein molecules, while  $\alpha$ -glucosidase breaks the linkage in maltose to release glucose. Phosphatase hydrolyzes phosphate esters and releases the phosphate group, and acid/alkaline phosphatase differ in terms of the optimum pH for maximum activity as well as the reaction mechanisms on different substrates (Goel et al., 1998). Fig. 5a–d shows the variation of hydrolytic enzyme activities at different pH values in NAC and SSAC. It was clear that alkaline treatment had a positive effect on hydrolytic enzyme activity, probably because of the higher amount of substrate available under alkaline conditions. The highest enzyme activities of protease,  $\alpha$ -glucosidase, alkaline phosphatase and acid phosphatase observed were in the fifth day of pH 10 conditions with the values of 4.93, 110.6, 83.0, 16.2 U/g VSS for NAC, and 5.88, 119.8, 90.5, 17.6 U/g VSS for SSAC. Although pH 10 may not be the optimal pH for these enzymes, this result indicates that in this process the

enhancement of enzyme activity by the additional amount of substrates can outweigh the negative effect of non-optimal pH conditions. Hydrolytic enzymes in SSAC showed higher activities than that of NAC, which may result from its better performance in sludge solubilization and could contribute to more phosphorus release and VFA production. It is noted that at pH 11 a longer time was required to reach the highest enzyme activities than at any other pH, and the activities were lower than those at pH 10 despite the higher organic matter solubilization at pH 11, which suggested that pH higher than 10 had a stronger inhibitory effect on enzyme activities, in accordance with the lower effectiveness of pH 11 at VFA production in the initial days of fermentation. In all scenarios, the enzyme activities decreased gradually after reaching the maximum values, possibly because the accumulation of products inhibited the enzymatic reaction.

In the literature, Feng et al. (2009) proposed a metabolic pathway for VFA production from protein and carbohydrate in which AK, BK, PTA, PTB and OAATC were identified as the key enzymes to transform the hydrolytic products of protein and carbohydrate into VFA. Fig. 5e–i shows the variation of acid-forming enzyme activities at different pH values in NAC

and SSAC. The highest activities of AK, BK, PTA, PTB and OAATC were observed at pH 10, 6 days and 8 days for NAC and SSAC, respectively, which was in good agreement with the maximum total VFA production mentioned above. Also, the activities of key enzymes in SSAC were slightly higher (5.88, 4.30, 0.21, 0.0032, 1.41 U/g VSS) than those in NAC (4.93, 4.19, 0.18, 0.0030, 1.30 U/g VSS).

### 3. Conclusions

Alkaline treatment with steel slag addition showed better performance than NaOH in the solubilization of organic matters and VFA production. Higher enzyme activities were also observed in SSAC than NAC under the same pH conditions. The orthophosphate concentration in the supernatant increased with fermentation time and pH in NAC, while in contrast most orthophosphate released in SSAC were transferred to steel slag. These results demonstrated that steel slag can be used as a substitute for NaOH in sludge alkaline treatment. Moreover, the steel slag, which successfully captured a large amount of phosphorus, could be further used as an efficient agricultural fertilizer.

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