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Variations in distribution and composition of extracellular polymeric substances (EPS) of biological sludge under potassium ferrate conditioning: Effects of pH and ferrate dosage

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**A B S T R A C T**

The effect of pH on chemical conditioning with potassium ferrate (K\textsubscript{2}FeO\textsubscript{4}) for improving sludge filtration dewatering performance has been studied. The variation of extracellular polymeric substances (EPS) in conditioning process was investigated in detail to unravel the reaction mechanism. The results indicated that sludge dewaterability was improved by decreasing solution pH in terms of filtration rate and cake solids content. At acid conditions, protonation of EPS resulted in reduction of negative charge and densification of sludge floc. Sludge conditioning efficiency was improved with decrease in pH. Ferrate can solubilize EPS through oxidation process and also remove a portion of soluble EPS (SEPS) by charge neutralization and interfacial adsorption of hydrolyzed ferric ions, consequently compressing EPS and decreasing total extractable EPS content. In addition, when pH and potassium ferrate were 3 and 0.1 g/gTSS, the sludge filtration dewatering rate and extent reached the maximum. Deterioration of sludge dewaterability was observed as the ferrate was overdosed (>0.2 g/gTSS). This could be attributed to the release of a large amount of bound EPS (BEPS) as a result of ferrate oxidation, consequently increasing the sludge filtration resistance.

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1. Introduction

The management of wastewater sludge, now often referred to as biosolids, accounts for a major portion of the cost of the wastewater treatment process and represents significant technical challenges. High-performance dewatering has been proven to be an efficient method to reduce sludge volume, cutting transportation and disposal cost [1]. Generally, the moisture in activated sludge can be classified into free water (about 70%), interfacial water (about 20%) and bound water (about 10%) [2]. Except for the sludge characteristics, the dewatering efficiency was mainly dependent on the selection of device and chemical conditioning process.

Many studies suggested that the sludge composition was the major factor affecting sludge dewatering performance. The extracellular polymeric substances (EPS) accounted for 60-80% of sludge biomass [3]. The distribution and chemical composition of EPS had significant effect on sludge dewatering property [4,5]. Houghton found that the there existed a certain EPS mass at which the sludge dewaterability reach the maximum [6]. Again, EPS content also greatly affected the charge property and floc stability [4]. Higgins and Novak demonstrated that the sludge dewaterability was mainly affected by the ratio of protein and polysaccharide. Proteins exhibited the more significant influence on sludge dewaterability than polysaccharide, high protein/polysaccharide was detrimental to dewatering process [7]. This observation was in agreement with Murtagy and Novak’s findings [8].

Prior to dewatering sludges are always conditioned with chemicals in order to improve operating performance of sludge dewatering devices. Addition of traditional chemical conditioners (inorganic salt coagulants and organic polymers) can agglomerate fine sludge colloids to form large flocs through charge...
neutralization and bridging, which can be more easily separated from the water [9,10]. Considering the high water binding properties of EPS, the traditional chemical conditioners are ineffective to remove the bound and intercellular water in sludge flocs. Therefore, many advanced sludge conditioning processes (ASCP) have been developed to improve sludge dewatering performance and to facilitate handling and ultimate disposal. ASCP can destroy the floc structure and solubilize EPS components, consequently converting the bound water into free water, consequently reducing the cake moisture content after dewatering step [1]. These techniques include (photo) Fenton oxidation [11–13], acid/alkaline [14] and thermal treatment [1] and enzymatic treatment or the integrated processes.

Potassium ferrate (K2FeO4) is known as a new generation green chemical which can oxidize and degrade a wide variety of organic contaminants in water [15]. Additionally, ferrate(VI) ions will be reduced to Fe(III) ions or ferric hydroxide, which can serve as a coagulant in the mixing unit process. K2FeO4 has been used as sludge conditioner in some previous studies [16,17], but the underlying mechanism was not fully understood, and some results are contradictory. Zhang et al. stated that sludge dewaterability was improved with ferrate treatment at pH of 3, while it was deteriorated at pH values above 4 [16]. Ye et al. found that K2FeO4 treatment resulted in deterioration of sludge filterability while improvement of settleability and dewatering performance. Meanwhile, the loosely bound (LB)-EPS concentration was increased with increasing K2FeO4 dosage, while tightly bound (TB)-EPS content showed an opposite pattern under conditioning [17].

Most of previous studies focused on effect of K2FeO4 conditioning on sludge dewatering performance, but few of them investigated the variation in EPS characteristics (distribution and composition) with advanced analytical techniques in chemical conditioning. Thus, the objectives of this study are to: (1) understand the impact of pH on sludge conditioning efficiency with ferrate; (2) unravel the underlying mechanism of different sludge conditioning processes by analyzing the variation in distribution and composition of EPS.

Table 1

| Indicator | Moisture content (%) | pH  | VSS/TSS | CST (s) | d15 (µm) | Zeta potential (mV) | SCOD (mg/L) |
|----------|----------------------|-----|---------|---------|----------|---------------------|------------|
| Value    | 98.3                 | 7.5 | 0.75    | 320     | 62       | -15.2              | 130        |

is 200 thousand ton. The wastewater is reclaimed with combined process of MBR and ozonation.

2.2. Sludge conditioning with K2FeO4

A 200 mL of sludge sample was added in a 500 Erlenmeyer flask. Then, under vigorous stirring using a magnetic stirrer, appropriate amounts of ferrate potassium were added in the beaker at varying pH values. After 2 h reaction, the solution was adjusted to pH 7. The suspension was used to extract the SEPS and bound EPS. Each experiment was performed in triplicate.

2.3. Analytical methods

2.3.1. Determination of sludge dewaterability

Specific resistance to filtration (SRF) is widely used to evaluate sludge dewatering performance in filtration process [9,10,16,17]. It can be obtained by eq. (1):

\[ r = \frac{2PA^2b}{\mu a} \]

where \( P \) (kg m⁻²) denotes pressure, \( A \) (m²) is filtration area, \( \mu \) (kg s m⁻²) is kinetic viscosity, \( \alpha \) (kg m⁻³) denotes dry solid weight per unit volume sludge on the filtrate media, \( b \) is slope of filtration equation–\( dt/dV = bV + a \), and \( t \) (s) is time, \( V \) (m³) denotes volume of filtrate. The raw and conditioned sludge was poured into a Buchner funnel with a 0.45 µm cellulose acetate membrane to filter under a pressure of 0.6 MPa of vacuum filtration. Volume of filtrate was recorded every 10 s before surface cracking was observed.

2.3.2. EPS extraction procedure

Firstly, raw sludge sample was settled down at 5000 g for 10 min, and the supernatant was collected as SEPS. The sediment was resuspended to its initial volume with 0.05% NaCl solution. And then the suspension was transferred and heated for 30 min in water-bath at the temperature of 60 °C. The extracted solution was centrifuged at 5000 × g for 10 min and separated as bound EPS [18]. The particles present in the two EPS fractions were removed with polytetrafluoroethylene membranes with a pore size of 0.45 µm prior to organic analysis.

2.3.3. EPS analysis

2.3.3.1. Protein and polysaccharide measurement. All chemical analyses were carried out in duplicate using chemicals of analytical grade. The protein and carbohydrate in extracted EPS were

### Table 1

| Indicator | Moisture content (%) | pH  | VSS/TSS | CST (s) | d15 (µm) | Zeta potential (mV) | SCOD (mg/L) |
|----------|----------------------|-----|---------|---------|----------|---------------------|------------|
| Value    | 98.3                 | 7.5 | 0.75    | 320     | 62       | -15.2              | 130        |

Surplus sludge (Table 1) was sampled from sludge return line of membrane bioreactor (MBR) in Northern brook wastewater treatment plant of Beijing. Now the daily wastewater treatment capacity

### Table 2

| pH | SEPS | Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid | pH | BEPS | Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid |
|----|------|-------------------|------------------|------------|-------------|----|------|-------------------|------------------|------------|-------------|
| 2  | 74.922 | 50.044 | 69.376 | 79.665 | 2  | 23.422 | 54.795 | 2.796 | 7.304 |
| 3  | 74.322 | 61.084 | 39.286 | 43.785 | 3  | 62.802 | 90.175 | 3.35  | 6.317 |
| 4  | 71.712 | 56.974 | 28.276 | 30.965 | 4  | 48.872 | 76.405 | 2.214 | 5.167 |
| 5  | 73.062 | 70.684 | 28.566 | 29.815 | 5  | 156.162 | 228.705 | 5.105 | 3.987 |
| 6  | 69.062 | 74.784 | 27.176 | 32.615 | 7  | 192.462 | 263.405 | 5.426 | 6.657 |
| 9  | 134.802 | 141.134 | 47.366 | 71.675 | 9  | 203.262 | 284.405 | 14.355 | 9.697 |
| 11 | 344.102 | 116.034 | 50.556 | 73.115 | 11 | 289.062 | 371.105 | 13.775 | 4.317 |

Sample of SEPS and BEPS were diluted by 20 and 100 times respectively.
Fig. 1. Effect of pH level on SRF and sludge cake moisture content with/without ferrate addition (ferrate dose was 0.1 g/gTSS).

Polysaccharide was measured using the anthrone method with a glucose standard (Sinopharm). Protein was determined with the Lowry procedure using bovine serum albumin (BSA) (Sigma) as standard.

2.3.3.2. 3-dimension excitation-emission matrix. The sample was diluted with Milli Q water until concentration of DOC was below 10 mg/L. The peak locations, peak intensities and the ratios of different peaks in EEM spectra of the EPS samples were not substantially influenced by ionic strength. Three-dimensional excitation emission matrix (3-DEEM) spectra were measured by a Hitachi F-4500 fluorescence spectrophotometer with an excitation range from 200 to 400 nm at 10 nm sampling intervals and an emission range from 280 to 500 nm at 10 nm sampling interval. The spectra were recorded at a scan rate of 12,000 nm/min, using excitation and emission slit bandwidths of 10 nm. Each scan had 37 emission and 27 excitation wavelengths [20].

2.3.3.2.1. High performance size-expulsion chromatography. Molecule weight analysis was conducted according to procedure described by Wang et al. [21,22]. MW was measured by a Waters liquid chromatography system which was consisted of Waters
2487 Dual λ. Absorbance Detector, Waters 1525 pump system. A Shodex KW 802.5 column (Shoko, Japan) was used for separation. The mobile phase, Milli Q water buffered with 5 mM phosphate to pH 6.8, and 0.01 M NaCl, was filtered through a 0.22 μm membrane, and then degassed for 30 min by means of ultrasonication for 0.5 h before being used to the column. 600 μL sample was injected at a flow rate of 0.8 mL min⁻¹. Polystyrene sulfonate standards (Sigma–Aldrich, USA) of molecular weights 1.8–32 kDa were used for apparent molecular weight (AMW) calibration.

2.3.4. Others

Other sludge parameters, including total suspended solids (TSS), volatile suspended solids (VSS) and chemical oxygen demand (COD) were measured with standard methods (APHA–AWWA–WEF, 1998). pH was measured by a pH5-SC (Shanghai, China) pH meter, which was calibrated using pH 7.01 and pH 9.18 buffers.

3. Results and discussion

3.1. Influence of pH on sludge filtration dewatering performance with/without ferrate addition

The influence of pH on SRF and cake moisture content in the absence of ferrate was given in Fig. 1(a). SRF was decreased from 4.6E + 13 m/kg to 2.5E + 12 m/kg when solution pH was reduced from 11 to 2. The change in moisture content showed a similar pattern with SRF. The cake moisture content was decreased from 82.3% to 50% by reducing pH from 11 to 2. These results demonstrated that the sludge dewatering performance was significantly improved by lowering the pH level of sludge suspension. The EPS properties were further characterized to understand the mechanism. In general, pH didn’t only affect the EPS and sludge properties, but change the chemical speciation and reactivity of K₂FeO₄. It was observed from Fig. 1(b) that sludge conditioning efficiency was improved by decreasing pH level. After ferrate treatment, SRF and cake moisture content were reduced from 9.6E + 12 m/kg and 81.8% to 3.1E + 12 m/g and 76.8% by decreasing pH from 11 to 2.

Meanwhile, the SRF value of sludge conditioned with K₂FeO₄ was significantly less than that of the blank at different pHs. K₂FeO₄ oxidation and acidification showed a significant synergistic effect in conditioning process.

3.2. Impact of pH on EPS properties with/without ferrate treatment

3.2.1. Effect of pH on sludge EPS characteristics

It can be seen from Fig. 2(a) that total extractable EPS concentration at alkaline conditions was significantly higher than that at acid conditions. EPS was primarily composed of protein and polysaccharide. Sludge particles were always negatively charged within certain pH range due to the ionization of anionic functional groups, such as carboxyl, amino and phosphate groups and so on. The presence of negative charge on the surface of particles can produce electronic repulsion and keep stability of a colloidal system. Acidic treatment could not only destroy EPS structure but caused protonation of anionic functional groups of EPS and aggregation of sludge system, consequently resulting in conversion of bound water into free water [23]. Low pH might affect proteins in two ways. Firstly, dissolution constant of proteins with high molecular weights (MW) reached the minimum at the pH of 2–3, the isoelectric point [3,24]. At the same time, the solubilization of protein reach the minimum, thus extractable BEPS content was reduced with decreasing pH value. On the other hand, the protein-like substances of low MW were solubilized and released into sludge bulk in acidic environment, consequently leading to increase of SEPS concentration. Additionally, high total extractable EPS concentration at alkaline conditions can be attributed to the increases in proteins solubility and negative charge on the surface of sludge particles due to unprotonation of EPS, consequently decreasing the binding strength of EPS and producing flocs with loose structure. The impact of pH level on polysaccharide and protein was given in Fig. 2(b) and (c) respectively. It is obvious that protein-like substances were the dominant fraction and accounted for more than 85% of total mass of sludge EPS. The changes in protein and polysaccharide contents

| Table 3 | Influence of pH on EEM profile of soluble EPS and BEPS after ferrate treatment. |
|---------|---------------------------------|
| pH     | SEPS                        | pH  | BEPS                        |
|        | Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid |        | Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid |
| λmax/nm | 280/335 | 225/340 | 330/410 | 275/425 | λmax/nm | 280/335 | 225/340 | 330/410 | 275/425 |
| 2      | 95.262  | 102.734 | 75.856 | 108.935 | 2      | 48.872  | 76.805 | 2.214  | 2.167   |
| 3      | 79.172  | 95.714  | 27.996 | 40.595  | 3      | 92.582  | 118.005 | 5.26   | 5.077   |
| 4      | 88.282  | 94.664  | 28.866 | 43.255  | 4      | 193.562 | 222.605 | 9.392  | 4.287   |
| 5      | 89.252  | 104.534 | 29.766 | 40.855  | 5      | 247.562 | 284.905 | 5.995  | 7.567   |
| 7      | 59.272  | 91.974  | 31.576 | 40.695  | 7      | 192.462 | 263.405 | 5.426  | 6.657   |
| 9      | 161.002 | 183.634 | 57.956 | 91.665  | 9      | 399.062 | 526.505 | 9.995  | 7.067   |
| 11     | 846.202 | 684.734 | 72.196 | 158.835 | 11     | 744.662 | 797.605 | 5.525  | 4.787   |

Sample of SEPS and BEPS were diluted by 20 and 100 times respectively.

| Table 4 | Influence of ferrate dosage on EPS fluorescent intensity after ferrate treatment. |
|---------|---------------------------------|
| Ferrate dosage (g/TSS)/SEPS | Ferrate dosage (g/TSS)/BEPS |
| Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid | Tryptophan protein | Aromatic protein | Humic acid | Fulvic acid |
| λmax/nm | 280/335 | 225/340 | 330/410 | 275/425 | λmax/nm | 280/335 | 225/340 | 330/410 | 275/425 |
| 0      | 69.062  | 74.784  | 27.176 | 32.615  | 0      | 92.462  | 133.405 | 5.426  | 6.657   |
| 0.005  | 62.422  | 61.655  | 8.316  | 17.917  | 0.005  | 49.952  | 64.685  | 3.961  | 3.637   |
| 0.01   | 22.622  | 26.425  | 7.008  | 13.817  | 0.01   | 60.722  | 86.095  | 4.572  | 5.337   |
| 0.02   | 23.752  | 48.655  | 7.303  | 15.177  | 0.02   | 55.632  | 73.375  | 4.1    | 4.647   |
| 0.05   | 19.432  | 31.395  | 6.522  | 12.747  | 0.05   | 43.932  | 52.495  | 3.228  | 2.227   |
| 0.1    | 15.232  | 17.835  | 7.04   | 11.447  | 0.1    | 32.392  | 38.775  | 3.002  | 3.539   |
| 0.2    | 12.982  | 18.925  | 7.353  | 12.497  | 0.2    | 40.552  | 44.475  | 3.287  | 2.067   |

Sample of SEPS and BEPS were diluted by 20 and 100 times respectively.
in SEPS showed a similar pattern with a rise of pH. The concentrations of polysaccharide and protein of SEPS increased firstly and then reduced with increasing pH from 2 to 11 and reached the minimum at neutral condition. For BEPS, the concentration of protein increased with a rise of pH level while that of polysaccharide was relatively stable. It was noted that pH had a more significant effect on proteins which were obviously solubilized and released into sludge bulk at pH less than 5. According to many previous reports, proteins in LB-EPS and soluble EPS were the major factors affecting sludge dewatering behavior [18,26]. It was found that higher concentration of extractable EPS, especially high protein concentration was always related to poor sludge dewatering performance. In addition, the protein hydrolysis and coagulation might be the major mechanism of sludge dewaterability improvement in acid environment.
The influence of pH on EEM spectra and fluorescent intensities of SEPS and BEPS were given in Fig. 3 and Table 2 respectively. Firstly, four fluorescent peaks could be detected in SEPS fraction: Peak A ($\lambda_{ex/em} = 280/335$)—tryptophan-like protein, Peak B ($\lambda_{ex/em} = 225/340$)—aromatic proteins, Peaks C ($\lambda_{ex/em} = 330/410$)—humic substances and Peak D ($\lambda_{ex/em} = 275/425$)—fulvic acid while BEPS only contained Peak A and Peak B [25]. Sheng and Yu demonstrated that the fluorescent intensity in EEM spectra can be used to quantify the EPS concentration when DOC of water sample was less than 10 mg/L [20]. It was observed that the fluorescent intensity of peaks A, B, C and D was improved from 75.7, 76.9, 30.3 and 48.2 to 88.3, 82.4, 75.8 and 81.9 when pH was reduced from 7 to 2. The variation in fluorescent intensity of protein-like substance was in accordance with analytical results from Lowry analysis. It was reported that humic and fulvic substances were more hydrophobic, thus they were prone to form complexes with cations and firmly embedded in the interior biofloc. The complexes were solubilised in the presence of acid, consequently resulting in release of humic and fulvic acids into the supernatant. Furthermore, the intensity of all four fluorescent peaks of SEPS was increased by raising sludge pH. The fluorescent intensity of peaks A and B improved from 26.0 and 64.2 to 320.9 and 411.2 by increasing pH from 2 to 11. The protein-like substances with low molecular weight (MW) was solubilized and converted into SEPS at acid conditions, while the binding strength of proteins with high MW to microbial cells was enhanced due to isoelectric point effect. However, under alkaline conditions, both negative charge density of proteins and solubility were increased under alkaline conditions, which weakened the adhesion force of EPS on microbial cells. As a result, increase in SEPS component was detected.

Based on the relationship of elution time and standard sodium polystyrene sulfonate (PSS) with different MW, the MW of organic matter with UV signal was classified. Since the polysaccharide contains no conjugated double bond, they only can be determined in the form of glycoprotein and glycolipid [27]. As depicted in Fig. 4, five MW peaks in SEPS could be observed: 41 kDa, 2500 Da, 1900 Da, 900 Da and 780 Da. According to report of Lyko et al., EPS could be divided into three fractions: high MW organic compounds (>5000 Da)—protein and polysaccharide, medium MW compounds (1000–5000 Da)—mainly composed by humic substances and organic matters of low MW (<1000)—building blocks [27]. No obvious variation in ME distribution of SEPS was observed at pH of 5–9. Two new MW peaks (320 Da and 130 Da) were detected by decreasing pH to below 4, while organics with MW of 210 Da, 320 Da and 190 Da were observed at pH higher than 11. EEM analysis also indicated that FI related to tryptophan- and tyrosine-like substances were significantly increased under strong alkaline conditions, so the organic matters with MW of 210 Da and 320 Da were very likely to be associated with amino acids or peptides. Overall, the signal intensity of all organic compounds was enhanced with increase in pH level. Additionally, it was worthy to note that peak intensities of the organic matters of high MW were reduced at acid environments (pH 2–4), especially the MW of 41 kDa could not be detected at pH of 2. This revealed that acidic treatment effectively removed the high MW organic fractions which were widely considered to be the factors resulting in high stickiness of sludge and thus difficult to dewater [10].

3.2.2. Impact of pH on distribution and composition of EPS after \(K_2\)FeO\(_4\) conditioning

The influence of pH on SEPS and BEPS concentration after ferrate treatment was given in Fig. 5(a). With increase in pH, BEPS content always increased, while SEPS concentration was reduced firstly and then increased and with the minimum at neutral condition. pH had important effect on chemical speciation of \(K_2\)FeO\(_4\) [15]. FeO_4^{2–} and FeO_4^{2+} are predominant in neutral and alkaline pH, in which Fe(VI) was known to be relatively stable towards its spontaneous decomposition to ferric ion (Fe(III)). At pH~10 the only species i.e., FeO_4^{2–} exists in aqueous solutions, FeO_4^{2–} and H_2FeO_4 are relatively more active species than the unprotonated species FeO_4^{2–}, are predominant species below neutral pH. It can be seen from reactions (1) and (2) the redox potential of ferrate(VI) is much higher in acidic medium than that in alkaline medium. Therefore, a powerful oxidizing condition is provided at the elevated concentration of the Fe(VI) species such as H_3FeO_4^{4+} and H_2FeO_4:

\[
\text{FeO}_4^{2–} + 8\text{H}^+ + 3\text{e}^- \rightarrow \text{Fe}^{3+} + 4\text{H}_2\text{O}^0 = +2.20 \text{V} 
\]

(1)

\[
\text{FeO}_4^{2–} + 4\text{H}_2\text{O} + 3\text{e}^- \rightarrow \text{Fe(III)} + 5\text{OH}^– + \text{H}_2\text{O}^0 = +1.20 \text{V} 
\]

(2)
Compared to the sludge samples without ferrate conditioning, the total extractable EPS (both SEPS and bound EPS) was decreased after K₂FeO₄ treatment, indicating that the binding strength of EPS on microbial cells was enhanced and flocs became denser. Furthermore, protein content in SEPS fraction was decreased at pH below 7 compared to that of blank group. The proteins exhibited high affinity with ferric ions compared to polysaccharide [27], so they were more prone to transfer from exterior to interior of sludge floc and more resistant to extraction under ferric coagulation process. These observations revealed that floc strength was intensified and sludge compressibility was reduced after ferrate treatment, consequently resulting in improvement of sludge filtration dewatering efficiency (Fig. 5).

The change in EEM spectra and FI of SEPS and BEPS after ferrate conditioning under different pH was presented in Fig. S1 in SI and Table 3 respectively. In SEPS, alkaline treatment resulted in intensification of four fluorescent peaks. However, the no obvious variation in two fluorescent peaks related to proteins was observed after acidic treatment, while fluorescent peaks of humic and fulvic substances were significantly enhanced at pH less than 2. With regard to BEPS, fluorescent peaks A and B were intensified from 48.9 and 76.8 to 744.7 and 797.6 by raising pH from 2 to 11, but that of humic and fulvic substances were very stable.

It can be seen from Fig. 6 that most of the organic matters with high MW in SEPS were removed after ferrate treatment at pH less than 5. Again, the removal of organic compounds with high and mid-MW (800 Da–20 kDa) was improved by decreasing pH levels. Only organic matters with 310 Da and 130 Da could be detected when the pH was reduced to 2, and other peaks disappeared. As mentioned above, the oxidative ability of K₂FeO₄ was enhanced with decrease in pH, thus leading to the improvement of removal efficiency of high and mid MW organic matters. Since K₂FeO₄ exhibited high reactivity at acid conditions, it was converted into polynuclear iron hydroxide species through hydrolysis.

### 3.3. Influence of K₂FeO₄ dose on sludge properties

#### 3.3.1. Effect of K₂FeO₄ dose on sludge dewatering performance

The influence of K₂FeO₄ dose on SRF and cake moisture content was given in Fig. 7. Both SRF and cake moisture content were greatly reduced in comparison to original sludge after K₂FeO₄ conditioning. This observation was inconsistent with the report of Ye et al., who found that the sludge filterability in term of SRF was deteriorated after K₂FeO₄ treatment regardless of its dose, while cake moisture content was reduced [17]. As K₂FeO₄ dosage was 0.05 g/gTSS, SRF was reduced to 2.9E + 12 m/kg, while SRF and cake moisture content were increased by increasing K₂FeO₄ dosage to 0.2 g/gTSS. In addition, both SRF and cake moisture content reached the minimum at the K₂FeO₄ dosage of 0.1 g/gTSS.

#### 3.3.2. Impact of K₂FeO₄ dosage EPS properties

From Fig. 8, it can be seen that the change in content of SEPS and BEPS showed the similar pattern with increasing K₂FeO₄ dosage, which is in agreement with that of SRF. As mentioned above, SRF correlated well with extractable EPS concentration, and high extractable EPS content was always associated with loose floc structure, high compressibility and poor dewaterability of sludge system. The change trend of protein in SEPS and BEPS was different. Protein concentration in SEPS fraction was increased with increasing K₂FeO₄ dosage while that of BEPS was reduced. However, the polysaccharide concentration in both BEPS and SEPS increased firstly and then reduced with increasing ferrate dosage. K₂FeO₄ can serve a dual function as oxidizer and coagulant. At low ferrate dosages, oxidation played a dominant role in chemical conditioning. Oxidation process can result in release of EPS and the iron hydroxide produced from reduction of K₂FeO₄ was not able to

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**Fig. 5.** Effects of pH on content of different EPS fractions (a) and their protein (b) and polysaccharide (c) after ferrate treatment (ferrate dosage was 0.1 g/gTSS).
sufficiently adsorb and remove the released EPS. However, coagulation might play a more important role in conditioning process at high ferrate doses. The hydrolyzed ferric ions can compress EPS structure via electrical neutralization and bridging, consequently decreasing extractable EPS content. When $K_2FeO_4$ was overdosed, content of proteins with low MW in SEPS fraction was increased, this can be explained by the fact that ferric coagulation exhibited limited effects on organic matters with low MW [10]. Sludge filterability in term of SRF was negatively correlated with protein concentration in SEPS component [26,27], the protein with high MW was oxidized into smaller ones, consequently increasing the sludge resistance to filtration.

The variation in EEM spectra and fluorescent intensity with $K_2FeO_4$ dosage is presented in Fig. S2 in SI and Table 4 respectively. Compared to original sludge, the fluorescent intensities of humic acid and fulvic acid in SEPS were greatly weakened after $K_2FeO_4$ conditioning. Humic acid and fulvic acid were typical hydrophobic organic compounds which were more easily transfer into solid phase through coagulation process. The change of protein in SEPS fraction showed a much more complicated pattern: increase–decrease–reincrease, which was different from Lowry analysis. On the other hand, the fluorescent intensities of tryptophan- and aromatic proteins in SEPS were significantly reduced after ferrate treatment. After ferrate oxidation, protein-like substances were decomposed into small organic matters which still maintained the fluorescent features of amino acid. This might cause the difference in analytical results between EEM and Lowry method.

The influence of $K_2FeO_4$ on molecular weight distribution of sludge SEPS could be found in Fig. 9. MW peak at 210 Da appeared after $K_2FeO_4$ conditioning. This observation confirmed the results from EEM analysis that ferrate oxidation caused decomposition of
high MW organic matters into smaller ones. At ferrate dosage was less than 0.1 g/gTSS, ferrate conditioning had no obvious influence on high MW biopolymers (1900 Da–40 kDa) while organic matters with low MW (210 Da and 900 Da) was significantly increased by increasing ferrate dosage. K$_2$FeO$_4$ oxidation resulted in solubilization and release of BEPS, of which high MW fraction was transferred into solid phase via ferric coagulation while smaller ones were difficult to be absorbed. On the other hand, except MW peak of 40 kDa remained unchanged, other MW peak intensities were greatly enhanced at high ferrate dosage (0.2 g/gTSS). This observation confirmed that BEPS was solubilized into the sludge bulk and coagulation was not sufficient to remove the released EPS which was mainly composed of protein-like substances. As mentioned above, many studies have demonstrated that sludge dewaterability was greatly dependent on protein content in SEPS fraction [26,27]. Therefore, release of a large amount of proteins caused a deterioration of sludge filterability as ferrate was overdosed.

Fig. 8. Influence of ferrate dosage on content of different EPS fractions (a) and their protein (b) and polysaccharide (c) after ferrate treatment (pH 3).
4. Conclusions

This study focused on the pH effect of chemical conditioning with K$_2$FeO$_4$ on EPS distribution and composition and sludge filtration dewatering performance. Some conclusions can be drawn as followed:

- Sludge dewaterability was improved by lowering pH level. pH affected sludge dewaterability through changing EPS properties. EPS was compressed and swelled under different pH conditions. This was caused by protonation and unprotonation of EPS (especially protein like substances). Therefore, the total extractable EPS at acid conditions was more abundant than that at alkaline environments;
- Sludge dewaterability could be improved enhanced after ferrate treatment regardless of pH, but its conditioning efficiency was enhanced with decreasing pH value. In addition, ferrate can serve as a dual function of oxidation and coagulation. BEPS was solubilized and released under oxidation process, and the produced ferric ion was able to adsorb and compress the structure of EPS via electric neutralization and interfacial adsorption. Thus, the total extractable EPS concentration was reduced after ferrate treatment;
- Both sludge filtration dewatering rate and moisture reduction efficiency reached the maximum when pH and ferrate dosage were 3 and 0.1 g/gTSS respectively. Overdosed ferrate (>0.2 g/gTSS) resulted in deterioration of sludge dewatering performance. This can be explained by a large amount of EPS released via oxidation of high dose of ferrate, leading to increase in sludge filtration resistance.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bej.2015.11.004.

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