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Study on the Efficiency of On-Site Sludge Reduction Using Ti/SnO_2-Sb and Ti/RuO_2-IrO_2 Electrodes Based on a Cell Lysis-Cryptic Growth System

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Abstract: The present study investigated the parameters and the mechanism of action of electrochemical cell lysis to reduce the return sludge from secondary settlers based on the theory of cell lysis-cryptic growth. The factors influencing the functioning of two electrodes (Ti/SnO_2-Sb and Ti/RuO_2-IrO_2) were investigated to determine the optimal cell lysis parameters for each electrode, and the effects of the two electrodes on cell lysis were compared under these conditions. Finally, the Ti/SnO_2-Sb electrode was selected for the subsequent experiments. The electrolysis reaction was performed using the following parameters: the initial sludge concentration was 7000 mg/L, the working voltage was 18 V, the plate spacing was 1 cm, the initial pH was 6.8 to 7.0, and the electrolysis duration was 90 min. The degree of disintegration of the sludge and the cell lysis rate reached 25.35% and 20.15%, respectively. In summary, electrochemical cell lysis has a good prospect for sludge reduction.

Keywords: electrochemical oxidation; on-site sludge; cell lysis-cryptic growth; Ti/SnO_2-Sb; Ti/RuO_2-IrO_2

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1. Introduction

Nowadays, the large volume of excess sludge has become a problem for wastewater treatment plants [1,2]; thus, it is necessary to reduce sludge production. The treatment and disposal of sludge is difficult owing to its complex composition and high water content. In addition, the sludge also contains a certain proportion of pathogens [3], steroid hormones [4], antibiotics [5] and other chemical drug residues [6]. Traditional sludge treatment and disposal methods, such as landfills, composting, or incineration, have negative environmental impacts [7]. In addition, if not properly handled, there will be highly negative impacts on the ecological environment and on human health [8–10]. Organic matter in excess sludge mainly comprises living cells and their metabolites. Generally, this matter cannot be reused by wastewater treatment systems and is therefore discharged. One of the solutions to this problem is to lyse and kill the sludge cells to release the intracellular organic compounds. These can form substrates for the growth of viable cells of the same population, thereby reducing the production of excess sludge [11]. Differing from the typical growth of microorganisms using substrates in raw wastewater, the process in which these microorganisms use dead cells as growth substrates is called cryptic growth [12,13]. Cell lysis through destructing cell walls and membranes is essential to reducing sludge

production using cryptic growth. This is also a rate-limiting step in the process of cell lysis–cryptic growth [14]. Compared with other techniques, such as an uncoupling metabolism, endogenous metabolism, and predation on bacteria, cell lysis–cryptic growth is simple and practical. Only one cell lysis unit is required in the reflux line. While this has limited impact on effluent water quality, it does improve sludge settleability. The current cell lysis methods do not meet several requirements, including those for affordability, efficiency, and low environmental impact. In contrast, electrochemical oxidation is an environmentally friendly technique with low requirements for chemical use, produces little pollution, and has good prospects for application in sludge cell lysis [15].

Electrochemical oxidation generally refers to an anodic electrochemical process, which can be categorized into direct oxidation and indirect oxidation according to its oxidation mechanism [16–18]. Direct oxidation refers to the oxidation of organic matter directly on the electrode surface. In this process, organic macromolecules resistant to degradation will be degraded into easily degradable organic micro-molecules, or directly mineralized into inorganic matter (CO_2 and H_2O) [19,20]. This process makes use of the high potential of the anode surface itself, and electron transfer and transport occur directly between the organic matter and the electrode. Indirect oxidation refers to the process in which strong oxidizing substances produced by the electrochemical reaction are used as reactants or catalysts to decompose and indirectly transform organic matter. Products of the process contain not only strong oxidizing substances such as chlorate, hypochlorite, and O_3, but also strong oxidizing intermediates such as solvated electrons (es), $HO_2\cdot$, OH, and O_2^- [21,22]. At present, there are two main research directions regarding sludge treatment using electrochemical oxidation: first, improving the dewaterability of excess sludge, and second, promoting subsequent anaerobic/aerobic digestion or cryptic growth as a pretreatment before cell lysis. Yuan et al. [17] investigated the effect of electrochemical treatment on sludge dewaterability using a combination of electrolysis pretreatment and the addition of a surfactant. Capillary suction time (CST) and specific resistance to filtration (SRF) were used to evaluate sludge dewaterability, and they found that electrolysis pretreatment and the addition of cetyl trimethyl ammonium bromide (CTAB) enhanced sludge dewaterability. Loginov et al. [23] investigated the pretreatment of electro-dewatering for drilling sludge using an electrode made of rigid titanium mesh with a constant current density of 80 A/m^2. They found that the dry weight of filter cake reached above 67% after pretreated drilling sludge was dewatered by pressure filtration and the addition of lime. In 2010, Song et al. [24] applied Ir/RuO_2 mesh electrodes to the anaerobic digestion pretreatment of activated sludge for the first time. Under optimal conditions, including the duration of electrolysis, electrolytic power, initial pH value, and initial sludge concentration, mixed liquor volatile suspended solids (MLVSS) decreased by 7.87%, and MLVSS decreased by 39.59% in the following 17.5 days of aerobic digestion. Feki et al. [25] investigated the pretreatment of activated sludge using thermo-alkaline (TA), H_2O_2 oxidation, electrolysis, and electrochemical oxidation (EO) processes. Under optimal conditions, the maximum chemical oxygen demand (COD) solubilization reached 28 % using the EO technique. During subsequent anaerobic digestion, a significant increase in biogas yield of up to 78% was observed in the EO pretreated samples compared to raw sludge, which reflected the great advantage of the EO technique in cell lysis pretreatment.

This study investigates the effects of Ti/RuO_2-IrO_2 and Ti/SnO_2-Sb electrodes on the efficiency of sludge cell lysis using the electrochemical oxidation technique by measuring the initial concentration of sludge, working voltage, plate spacing, initial pH, and duration of electrolysis. It further identifies the preferred electrode for sludge treatment using the electrochemical oxidation technique. The study also explains the mechanism of electrochemical cell lysis based on the changes in characteristics of cell-lysed sludge and provides evidence for future studies on the electrochemical cell lysis technique.

2. Materials and Methods

2.1. Electrochemical Cell Lysis Experimental Setup

The experiment was conducted in a cylindrical glass reactor. The reactor volume was 500 mL, and the sludge volume was 400 mL throughout the experiment. The Ti/SnO_2-Sb mesh electrode (hereafter also called the Sn-Sb electrode) and the Ti/RuO_2-IrO_2 mesh electrode (hereafter also called the Ru-Ir electrode) were used as anodes, and the two cathodes were made of titanium mesh. Both the cathode plate and anode plate areas were 5 cm × 5 cm and the plate spacing was adjustable. A direct current (DC) stabilized voltage supply was used to control the voltage during the reaction. A magnetic stirrer was used during the electrolytic process to ensure the uniformity of the reaction system. In addition, a water bath was used for temperature control and the temperature of the system was controlled at 23 ± 2 °C. A schematic diagram of the setup is shown in Figure 1.

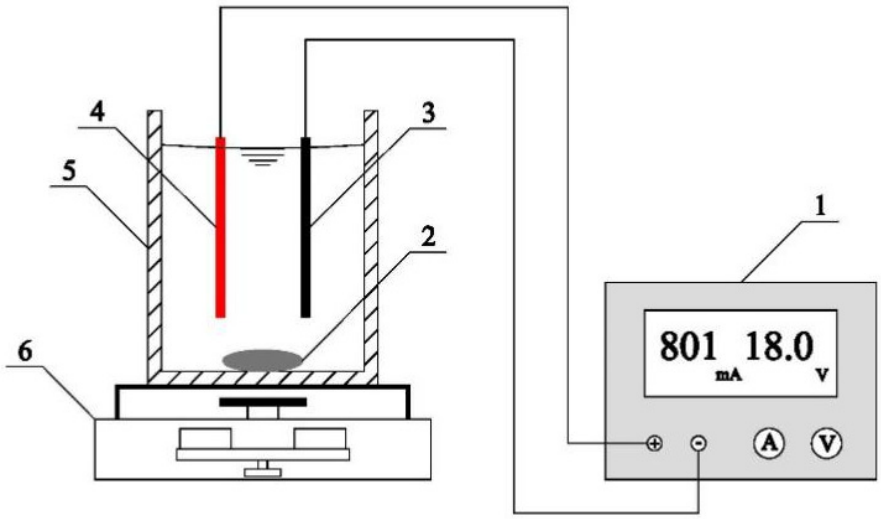

Figure 1. Schematic diagram of electrochemical cell lysis static experimental setup. 1. DC stabilized power supply; 2. Stir bar; 3. Titanium cathode; 4. Titanium coating electrode (anode); 5. Electrolytic cell; 6. Magnetic stirrer.

2.2. The Source of Sludge

The sludge used in the experiment was obtained from the return sludge of the secondary settler of Nanshan Wastewater Treatment Plant in Shenzhen. After gravity settlement for 24 h, the concentrated sludge and the supernatant were stored in a refrigerator at 4 °C for a maximum of 72 h. During the experiment, the working concentration of sludge was prepared and adjusted by the raw sludge and the supernatant. The basic characteristics of the concentrated sludge are shown in Table 1.

Table 1. Basic properties of concentrated sludge.

Indicator	Range
pH	6.2~6.8
Moisture content (%)	97.9~98.8
Mixed liquor suspended solids (MLSS) (g/L)	11.5~19.6
Mixed liquor volatile suspended solids (MLVSS) (g/L)	5.69~9.74
MLVSS/MLSS	0.46~0.50

2.3. Analysis Methods

2.3.1. Determination of Cell Lysis Rate (C)

Cell lysis rate made the detection by referring to the experimental method selected by Yuan et al. [26], and calculated the cell lysis rate of sludge according to the changes in

soluble chemical oxygen demand (*SCOD*) before and after electrochemical cell lysis. Cell lysis rate was calculated according to Equation (1):

$$C = \frac{SCOD_t - SCOD_0}{SCOD_{\mathrm{NaOH}} - SCOD_0} \times 100\% \quad (1)$$

where C is the cell lysis rate (%); $SCOD_{\mathrm{NaOH}}$ is the dissolved *COD* concentration in the supernatant measured after the original sludge was mixed with 1 mol/L NaOH for 24 h (mg/L); $SCOD_0$ is the concentration of dissolved *COD* in the original sludge supernatant (mg/L); and $SCOD_{\mathrm{t}}$ is the concentration of *COD* in supernatant at electrochemical cell t minute (mg/L).

2.3.2. Determination of Sludge Disintegration Degree (R)

Sludge disintegration degree (R) was based on MLVSS before and after electrochemical cell dissolution of sludge which was selected by Song et al. [24], calculated according to Equation (2):

$$R = \frac{MLVSS_t - MLVSS_0}{MLVSS_0} \times 100\% \quad (2)$$

where R is the sludge disintegration degree (%); $MLVSS_0$ is the volatile suspended solid concentration of the mixture of the original sludge (mg/L); and $MLVSS_t$ is the concentration of volatile suspended solids in the sludge mixture at electrochemical cell t minute (mg/L).

2.3.3. Determination of SOUR

SOUR was calculated according to Equation (3) by measuring MLVSS and oxygen consumption rate (DO).

$$SOUR = \frac{DO_0 - DO_t}{t \times MLVSS} \quad (3)$$

where *SOUR* is the specific oxygen consumption rate of sludge ($mgO_2 \cdot gMLVSS^{-1} \cdot min^{-1}$); DO_0 is the initial concentration of dissolved oxygen (mg/L); DO_t is the concentration of dissolved oxygen at time t (mg/L); t is the electrolytic time (min); and MLVSS is the concentration of volatile suspended solids in the sludge mixture (g/L).

2.3.4. Extraction and Determination of Extracellular Polymeric Substances (EPS) from Sludge

EPS was extracted from sludge by an improved thermal extraction method [27]. A 50 mL sludge sample was added into a centrifuge tube, centrifuged at 4000× g for 10 min, and the supernatant was collected as soluble EPS (SB-EPS). The remaining sludge was then resuspended to 50 mL with 0.05% NaCl solution. The supernatant was centrifuged at 8000× g for 10 min to collect loosely bound EPS (LB-EPS). The remaining samples were resuspended to the original volume, heated in a water bath at 60 °C for 30 min, and centrifuged at 12,000× g for 30 min to collect the supernatant which was tightly bound EPS (TB -EPS). In the end, three EPSs were filtered by 0.45 m filter and testing. The proteins in EPS were determined by the fast Lowry method, and the polysaccharide was determined by phenol-sulfuric acid method.

2.3.5. Other Analytical Methods

Water content (WC), MLSS, and MLVSS were analyzed using Standard Analysis Methods [28]. The sludge particle size was measured using a laser particle size analyzer (Mastersizer 2000, Malvern Panalytical, Malvern, UK). The determination of sludge capillary water absorption time (CST) was performed by the CST tester [29] (304M, Triton Electronics Ltd, Dunmow, UK). The microscopic morphology of sludge was observed by scanning electron microscope (SU8010, Hitachi Ltd., Tokyo, Japan). The structure changes of sludge functional groups before and after electrochemical treatment were analyzed and measured by Fourier transform infrared spectrometer (FTIR, Nicolet iS50, Thermo Fisher

Scientific, Waltham, MA, USA). The electrical conductivity (EC) was measured with a Digital Conductivity meter (HQ14d, HACH, Loveland, CO, USA).

2.3.6. Data Analysis

In this study, all measurements were done in triplicate. Experimental results in figures and tables were presented as mean value ± standard deviation. The statistical significances of the results were analyzed by SPSS software (version 19.0, IBM, Armonk, NY, USA).

3. Results

3.1. Investigation of Factors Influencing Cell Lysis Using Two Electrodes

3.1.1. Effect of Working Voltage on the Efficiency of Cell Lysis

Preliminary experiments showed that when the voltage was less than 6 V, electrochemical oxidation had a slight effect on sludge disintegration. Therefore, the effect of different voltage levels between 6 V and 30 V on the degree of sludge disintegration was investigated in the experiment using the following parameters: the pH was 6.8 to 7.0, the initial sludge concentration was 7000 mg/L, the plate spacing was 1 cm, and the duration of electrolysis was 30 min. The variation trend is shown in Figure 2a.

When the voltage was less than 10 V, for either degree of sludge disintegration or cell lysis rate as the evaluation parameters, the lysis rate increased slowly, indicating that such low voltage has a particularly limited effect on sludge disintegration. With a continuous increase in voltage, the degree of sludge disintegration in the Ru-Ir electrode reactor also increased rapidly. Once the voltage was 16 V, there was limited improvement in the degree of sludge disintegration, and with continuous increase in voltage, the side reaction from the electrolysis of water became more severe. When the voltage was higher than 20 V, the gas released from the electrode resulted in large quantities of sludge float-up, leading to the stratification of the sludge and water. Therefore, 16 V was set as the optimal voltage for subsequent electrolysis in the Ru-Ir electrode reactor, which not only ensured the production of sufficient oxidizing free radicals to maintain a good effect of sludge disintegration but also avoided the stratification of the sludge and water caused by the strong side reaction.

When the voltage increased from 6 V to 30 V, the cell lysis efficiency of the Sn-Sb electrode initially increased and then decreased. When the voltage was between 12 V and 18 V, the cell lysis rate increased most rapidly, although the rate increased significantly more slowly at higher voltages. Therefore, 18 V was set as the working voltage of the Sn-Sb electrode in the subsequent experiments. Noticeably, at low voltage ($U \leq 6$ V), the content of SCOD in the supernatant showed a negative increase. This was because at low voltage there was insufficient electrochemical oxidation to disrupt the floc structure and cell membrane of the sludge, but only COD in the supernatant was oxidized, and its content was reduced.

3.1.2. Effect of Initial Sludge Concentration on Cell Lysis Efficiency

The effect of initial sludge concentration on cell lysis efficiency is mainly reflected in two aspects: when the sludge concentration is relatively low, the fluidity of the mixture is relatively good. The electrochemical oxidation rate is mainly dominated by the concentration; that is, the total number of sludge particles that are in contact with the unit plate area. While the sludge concentration was relatively high, the viscosity of the mixture increased, and the fluidity decreased. The mass transfer then becomes the rate-limiting step that controls the electrochemical oxidation rate. In this experiment, MLSS was selected to represent the sludge concentration.

The total COD of sludge at different concentrations also differed. We also investigated the changes in SCOD and its growth rate, as well as the change in MLVSS reduction rate to better reflect the change in electrolytic efficiency [30,31]. The variation trend under the experimental conditions is shown in Figure 2b, with the pH ranging from 6.8 to 7.0, the working voltage of the Ru-Ir electrode was 16 V, the working voltage of the Sn-Sb electrode

was 18 V, the plate spacing was 1 cm, the duration of electrolysis was 30 min, and the five initial sludge concentrations were set as 3000 mg/L, 5000 mg/L, 7000 mg/L, 9000 mg/L, and 11,000 mg/L, respectively.

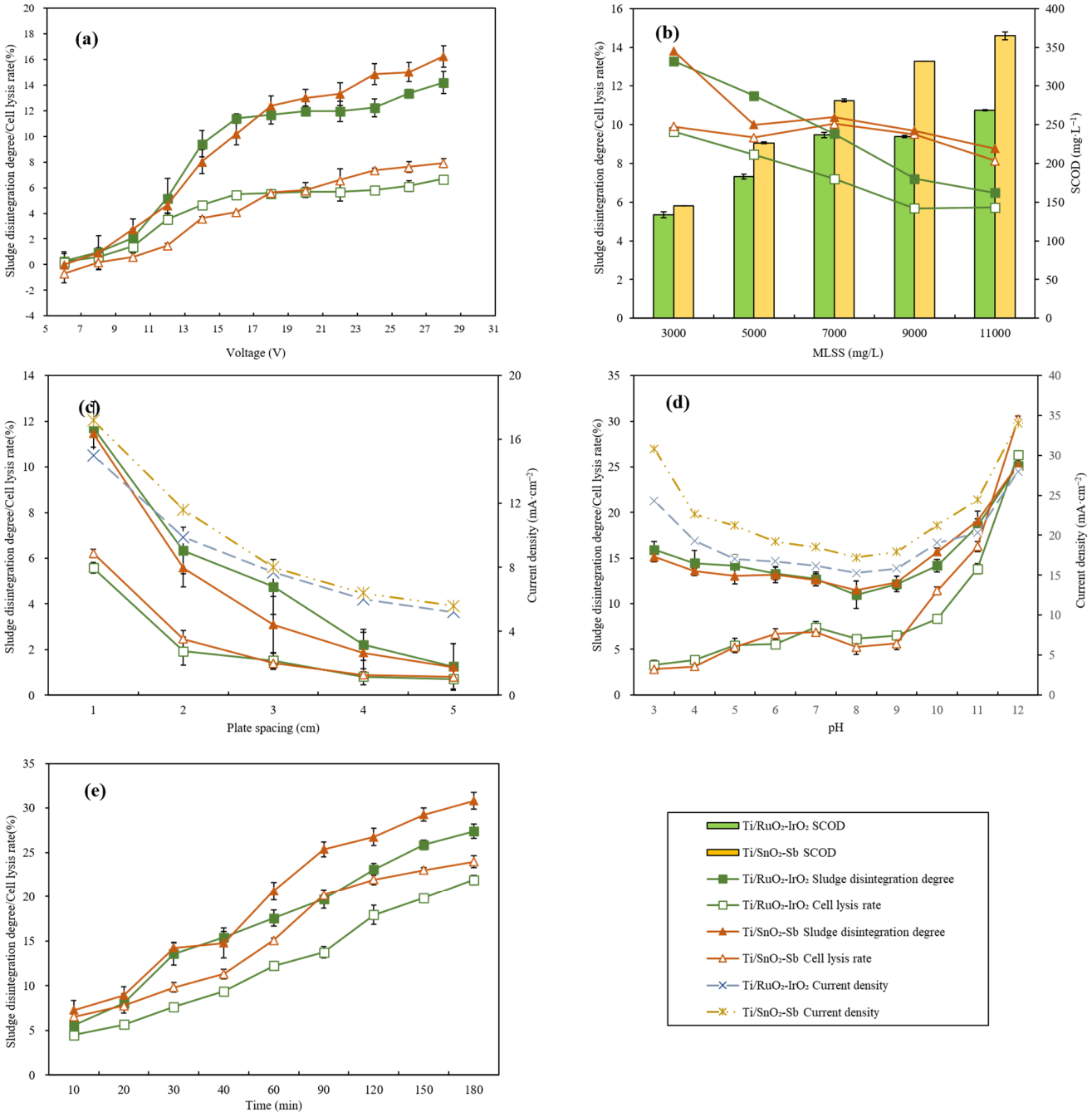

Figure 2. The effect of cell lysis using Ru-Ir and Sn-Sb electrodes varying: (**a**) Voltage, (**b**) MLSS, (**c**) Plate spacing, (**d**) pH, and (**e**) Time.

As sludge concentration increased, SCOD in the supernatant also increased after electrolysis for both electrodes. When the sludge concentration increased from 3000 mg/L to 11,000 mg/L, SCOD in the supernatant of the Ru-Ir electrode reactor increased from 133.88 mg/L to 268.69 mg/L and the corresponding cell lysis rate decreased from 9.64% to 5.72%. The cell lysis rate and degree of disintegration of the sludge decreased by 40.7% and 51.2%, respectively. The degree of sludge disintegration in the Sn-Sb electrode reactor decreased from 13.81% to 8.76%, but the magnitude of this decrease was slightly smaller

than that of the Ru-Ir electrode. The variation trend of cell lysis contrasted to that of SOCD because the calculated value of cell lysis rate was negatively correlated with TCOD. Further, with sludge concentration increasing, the TCOD of the mixture also increased. Thus, the cell lysis rate still decreased despite the increase in SOCD in the supernatant. In consideration of the practical application of the project, it was unsuitable for concentrating and diluting sludge. To obtain the data closest to the in situ environment, the concentration of raw sludge (7000 mg/L) was used for the subsequent experiments.

3.1.3. Effect of Plate Spacing on Cell Lysis Efficiency

Plate spacing that is too wide leads to a decrease in cell lysis efficiency due to the decrease in migration rate, while plate spacing that is too narrow tends to cause short circuits. Therefore, this experiment investigated the changes in cell lysis efficiency with plate spacing varying between 1 cm and 5 cm. The specific experimental parameters were as follows: the pH value was 6.8 to 7.0, the initial sludge concentration was 7000 mg/L, the working voltage of the Ru-Ir electrode was 16 V, the working voltage of the Sn-Sb electrode was 18 V, and the duration of electrolysis was 30 min. The experimental results are shown in Figure 2c.

The degree of sludge disintegration decreased with increasing plate spacing, and the magnitude of this reduction decreased continuously. When the spacing increased from 1 cm to 5 cm, the corresponding degree of sludge disintegration in the Ru-Ir electrode reactor decreased from 11.71% to 1.27%. The degree of sludge disintegration in the Sn-Sb electrode reactor decreased from 11.46% to 1.24%, and the apparent current density decreased from 17.2 mA/cm^2 to 5.6 mA/cm^2. The downward trend in the current density at different plate spacings paralleled the degree of disintegration which was consistent with previous results [32]. The reason why the degree of disintegration decreased was that, as the plate spacing increased, the resistance between the plates increased, the current decreased, and the electrochemical oxidation became weak, leading to a reduced lysis rate.

3.1.4. Effect of Initial pH on Cell Lysis Efficiency

On the one hand, pH affects the catalytic oxidation performance of electrode materials. On the other hand, it also affects the mass transfer rate. The cell lysis rate and degree of sludge disintegration from pH 3 to 12 were measured to investigate the changes in cell lysis efficiency at different pH values. The specific experimental parameters were as follows: the initial sludge concentration was 7000 mg/L, the working voltage of the Ru-Ir electrode was 16 V, the working voltage of the Sn-Sb electrode was 18 V, the plate spacing was 1 cm, and the duration of electrolysis was 30 min. The experimental results are shown in Figure 2d.

The variation trends in degree of sludge disintegration and cell lysis rate differed. When the pH of the original sludge was around 7, the degree of sludge disintegration in both electrode reactors was improved regardless of whether the pH was acidic or alkaline [33]. However, as for the cell lysis rate, it was increased only when the alkalinity improved, but it was reduced under acidic conditions. The reason for increased sludge disintegration despite a decreased rate of cell lysis was that, at low pH, the mineralization ability of electrochemical oxidation was stronger, and most of the dissolved organic matter completely degraded to inorganic matter.

Noticeably, when the pH was adjusted to 8, both the degree of sludge disintegration and the cell lysis rate decreased, although by different magnitudes. There were two reasons: (1) the addition of NaOH enhanced the conductivity of the solution and increased the current density; (2) due to the increase in pH, the oxygen evolution overpotential of the electrode gradually decreased, which led to intensified side reactions of electrochemical oxidation and prevented improvement in lysis efficiency. The addition of OH^- also disrupted the floc structure and cell wall, resulting in damage to the natural form of the proteins, lipid saponification, and RNA hydrolysis [34,35].

Because of the interaction of the above-mentioned factors, the lysis rate first decreased, and then increased as the alkalinity increased. It can be seen from Figure 2d that the

optimal pH was 12. Given that the real experiment would be carried out with continuous flow, and the pH value would be difficult to adjust, raw sludge (pH 6.8 to 7.0) was set for subsequent experiments.

3.1.5. Effect of Duration of Electrolysis on Cell Lysis Efficiency

The duration of electrolysis is also an important factor affecting cell lysis. On the one hand, the production of strong oxidizing free radicals increases with duration of electrolysis, and the lysis rate should be increased; on the other hand, when electrolysis is proceeding, some reductive substances can be released while cell lysis is underway, and these substances may, in turn, participate in the electrode reaction, which reduces the cell lysis efficiency using the electrode. In this experiment, the changes in the cell lysis effect using the two electrodes were investigated within 0–180 min under conditions where the initial sludge concentration was 7000 mg/L, the working voltage of the Ru-Ir electrode was 16 V, the working voltage of the Sn-Sb electrode was 18 V, the plate spacing was 1 cm, and the pH value was 6.8–7.0. The experimental results are shown in Figure 2e.

During the first 30 min, both electrodes experienced a rapid increase in the degree of sludge disintegration. After 30 min, the cell lysis efficiency decreased slightly with increasing duration of electrolysis, but the change was not significant, and the degree of sludge disintegration and the rate of cell lysis continued to rise. At 30 min, the degree of sludge disintegration and cell lysis rate using the Ru-Ir electrode reached 13.54% and 7.62%, respectively; at 180 min, the degree of sludge disintegration and cell lysis rate reached 27.38% and 21.90%, respectively.

At 30 min, the degree of sludge disintegration using the Sn-Sb electrode reached 14.21%; during the period 30–90 min, the degree of sludge disintegration decreased slightly; at 90 min, the degree of sludge disintegration reached 25.35%; after more than 90 min had elapsed, the degree of sludge disintegration slowed down significantly; once duration had doubled to 180 min, the degree of sludge disintegration increased by 21.50%. The variation trend was more significant when the cell lysis rate was used as the evaluation parameter. During the initial 90 min, the cell lysis rate increased almost linearly, reaching 20.15% at 90 min, but the cell lysis rate only increased by 12.16% at 180 min. In summary, it was more economical and feasible to select 90 min of electrolysis as one of the parameters for subsequent experiments.

It can be seen from Figure 2e that when the degree of sludge disintegration was used as the evaluation parameter, the two electrodes exhibited a similar cell lysis effect during the first 40 min; after 40 min, the Sn-Sb electrode was dominant. When cell lysis rate was used as an evaluation parameter, the Sn-Sb electrode was dominant all the time. Therefore, it had a better effect on sludge cell lysis, and it was selected as the anode in the subsequent experiments. The corresponding electrolysis parameters were as follows: the initial sludge concentration was 7000 mg/L, the working voltage was 18 V, the plate spacing was 1 cm, pH was 6.8 to 7.0, and the duration of electrolysis was 90 min. The EC before and after electrochemical cell lysis treatment were 1930 μs/cm and 2170 μs/cm, respectively.

3.2. Effect of Electrolytic Cell Lysis on Sludge Physical Properties Using the Sn-Sb Electrode as the Anode

3.2.1. Concentration

Figure S1 shows that during electrolysis from 0 to 90 min, MLSS decreased from 6710 mg/L to 5694 mg/L, MLVSS decreased from 3191 mg/L to 2382 mg/L, and IS decreased from 3580 mg/L to 3312 mg/L, with corresponding reduction rates of 15.14%, 25.35%, and 7.48%, respectively. The results showed that electrochemical oxidation effectively solubilized solid parts of the sludge, and the solubilized part mainly comprised organic matter. The solubilization rate of sludge solids gradually decreased with increasing duration of electrolysis, and the solubilization rate was fastest during the initial ten minutes.

The solubilization rate of IS was similar to that of MLSS and MLVSS, but the variation magnitude was relatively small. This was owing to (1) electrochemical oxidation that

disrupted the sludge cells, and some inorganic matter that was adsorbed inside and outside the cells, such as heavy metals and inorganic salts which were released into the supernatant; and (2) the strong oxidizing free radicals produced by the electrode that can oxidize some inorganic matter in the process of cell lysis; this oxidized inorganic matter can be dissolved in the supernatant.

3.2.2. Particle Size

In the experiment, the particle size distribution of sludge particles before and after electrolysis (for 90 min before and after the treatment) was analyzed using a laser particle analyzer, and the experimental results are shown in Figure 3.

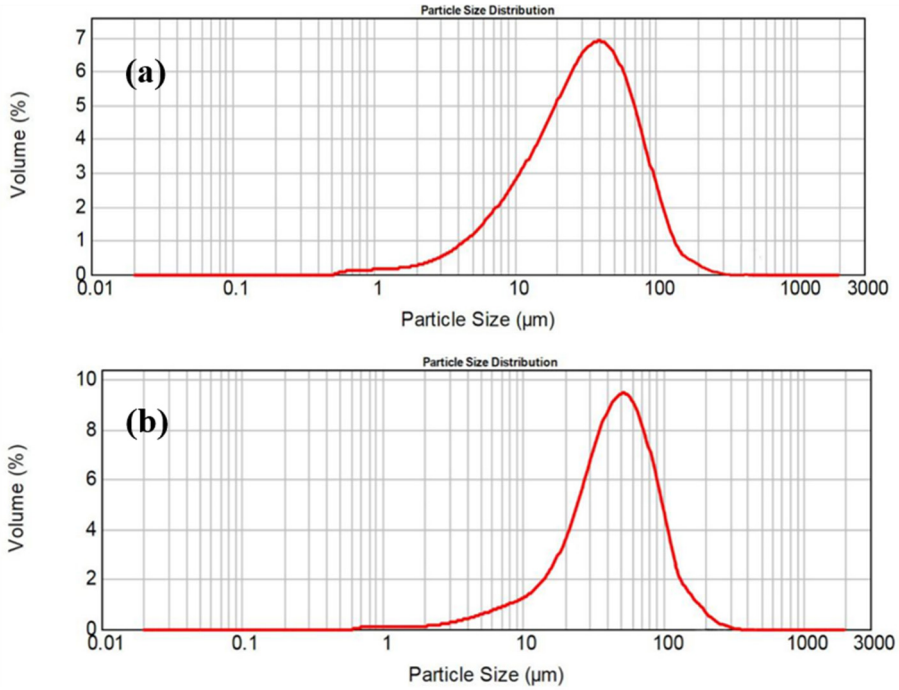

Figure 3. (**a**) Particle size distribution of sludge before electrochemical treatment, (**b**) Particle size distribution of sludge after electrochemical treatment.

After electrochemical cell lysis treatment, the particle size distribution of sludge particles was more dispersed, and the percentage of median particle size decreased significantly. Meanwhile, the median particle size d50 of sludge decreased from 44.428 μm (before electrolysis) to 36.871 μm (after electrolysis), decreasing by 17.01%. The experimental results verified our hypothesis that electrochemical oxidation did contribute to the disruption of sludge flocs, the disruption of micelles, and the reduction of the median particle size of sludge.

3.2.3. pH

The pH changes during 0–90 min was investigated in the experiment, and the results are shown in Figure S2. The pH of the original sludge was 6.95 ± 0.08, and the pH of the sludge mixture decreased with the duration of electrolysis. The pH variation rate was slow during the initial 20 min and then decreased rapidly from 20–60 min. It was speculated that electrochemical oxidation first disrupted the sludge flocs and then exerted an effect on the sludge cells with electrochemical oxidation proceeding. After 60 min, the decrease in pH slowed down again, probably because the oxidation of sludge reached relative saturation.

The decrease in pH might be related to acidic substances released by cell disruption. Song et al. [36] demonstrated that the concentration of volatile organic acids, such as acetic acid, propionic acid, and lactic acid in the supernatant of sludge, was increased during cell

lysis. In addition to direct release, the source of these organic acids with low molecular weight may be oxidized by macromolecular organic matter.

3.2.4. Settleability

The above experiments illustrated that electrochemical treatment decreased sludge particle size. Given that there was a specific relationship between sludge settleability and particle size, in this study the sludge settling ratio (SV_{30}) was used as the evaluation parameter to investigate the changes in settleability in the electrolysis process. The experimental results are shown in Figure S3. The sludge settling ratio showed an overall downward trend with duration of electrolysis. When the duration was 0–90 min, the corresponding SV_{30} values were 44.6% and 24.6%, respectively, decreasing by 44.84%, indicating that the electrochemical cell lysis process could effectively improve sludge settleability. The results were consistent with the study published by Park et al. [37] who have reported that the concentration of proteins and polysaccharides in the sludge supernatant can be improved after cell lysis, and the presence of proteins and polysaccharides can promote sludge particle adsorption, interparticle bridging, and double-layer compression. Further, they can shorten the distance between sludge particles, and finally, they can produce compact flocs with good settleability, which has been verified by Zhen et al. [38].

3.2.5. Dewaterability

The above experiments demonstrated the effect of electrochemical cell lysis on particle size and sludge settleability. Generally, improved sludge settleability means decreased dewaterability. This study used capillary suction time (CST) as an evaluation parameter to investigate the relationship between sludge dewaterability and duration of electrolysis to verify this conclusion. The results indicate an inverse relationship between CST value and sludge dewaterability. The experimental results are shown in Figure S4. The CST value increased continuously with the duration of electrolysis, from 20.7 s at the outset to 66.2 s at 90 min, indicating that the sludge dewaterability decreased. The reason for this might be that, during the process of electrolytic sludge cell lysis, some of the sludge was oxidized and disintegrated, resulting in a large amount of sludge debris, which then negatively affected sludge dewaterability.

3.2.6. Microstructure

Based on the analysis of changes in sludge physical properties, we inferred that floc disruption and cell lysis of sludge occurred in the electrochemical cell lysis process. This would inevitably change the apparent morphology and microstructure of sludge. To confirm these changes, an optical microscope and a scanning electron microscope were used in this study to observe the raw sludge and the sludge treated by electrochemical cell lysis. The experimental results are shown in Figure 4.

In Figure 4a,b, the raw sludge showed a complete micelles structure under an optical microscope, and the sludge floc structure was closely related to each other, while after electrolysis the sludge floc structure was obviously loose, and most of the free sludge debris were in the field of vision, indicating that electrochemical action could effectively destroy the structure of the sludge floc and micelles.

In Figure 4c,d, it could be observed that the surface of the original sludge before electrolysis was relatively smooth and the sludge cells were relatively complete, while the surface of the sludge after electrolysis was very rough and damaged cells could be observed at the same time, indicating that electrochemical action could effectively destroy the microscopic structure of sludge and dissolve the sludge cells.

3.3. Effect of Electrolytic Cell Lysis on Biological Activity Using the Sn-Sb Electrode

We can observe the lysis and destruction of sludge cells in the SEM images, and we can confirm the destruction of sludge cells according to the analysis of sludge biological activity. In this study, the specific oxygen uptake rate (SOUR) of sludge was used to characterize

its biological activity. SOUR changes in the electrochemical cell lysis process are shown in Figure 5.

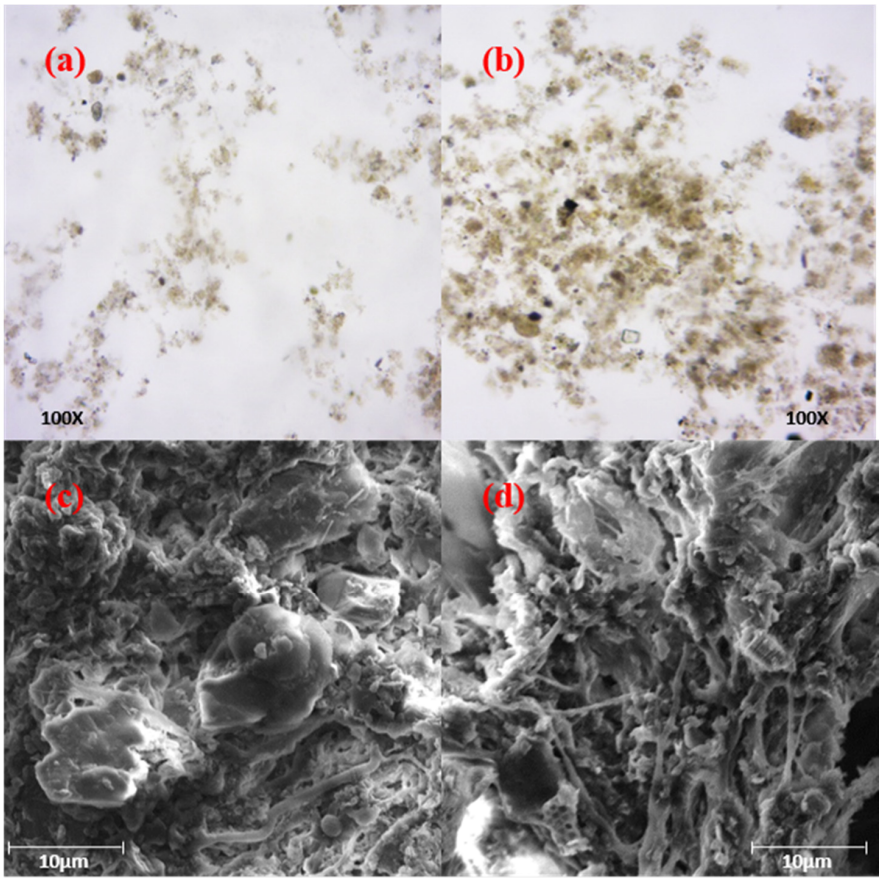

Figure 4. (**a**) An optical microscope image of raw sludge morphology (100×), (**b**) An optical microscope image of electrolyzed sludge morphology (100×), (**c**) A scanning electron micrograph (SEM) image of raw sludge morphology, and (**d**) An SEM image of electrolyzed sludge morphology.

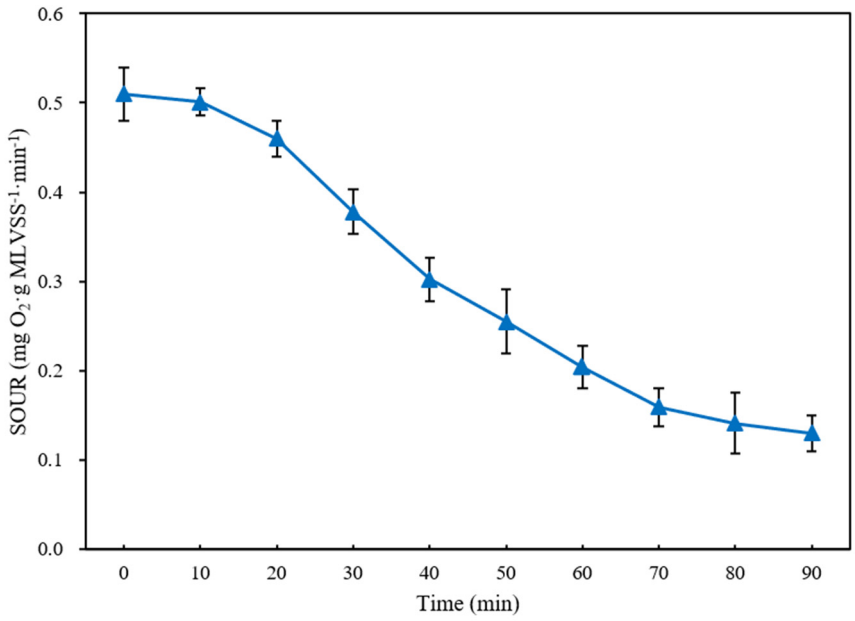

Figure 5. The variation of sludge specific oxygen uptake rate (SOUR) during electrochemical cell lysis.

The SOUR rate decreased along with the electrochemical cell lysis, indicating that the biological activity of sludge decreased continuously. At 90 min, the SOUR of sludge treated by electrochemical cell lysis was only 25.49% of that of the raw sludge, indicating that nearly 75% of the sludge was inactivated, and the electrochemical technique could effectively lyse sludge cells.

3.4. Effect of Electrolytic Cell Lysis on Sludge Chemical Composition Using the Sn-Sb Electrode

3.4.1. COD

In the process of electrochemical cell lysis, the change in COD solubilization is an important indicator for the evaluation of cell lysis. Further, the oxidability of the strong oxidizing free radicals produced during catalytic oxidation is particularly strong, and some organic matter could be directly oxidized to end-products, namely CO_2 and H_2O. This could lead to a change in the total COD of the mixture. Therefore, this study investigated the changes in total COD, particulate COD, and soluble COD in the electrochemical cell lysis process, and the experimental results are shown in Figure 6a.

The total COD of the sludge mixture decreased continuously for the duration of electrolysis, suggesting that electrochemical oxidation directly mineralized some organic matter. By 90 min, the total COD of the sludge mixture had decreased from 3668.25 mg/L to 3043.70 mg/L, decreasing by 17.03%. Meanwhile, the concentration of solubilized COD in the supernatant of sludge rose from 34.50 mg/L at 0 min to 503.31 mg/L at 90 min. The reason might be that some extracellular polymers and intracellular substances, such as volatile fatty acids, proteins, and polysaccharides, were released into the supernatant owing to the disruption of sludge flocs and cell lysis caused by electrochemical oxidation.

Song et al. [36] demonstrated that electrochemical pretreatment can effectively disrupt sludge cells, and the sludge intracellular substances were then dissolved. Compared with untreated sludge, sludge treated by electrochemical cell lysis showed better digestion performance and produced more biological gases.

3.4.2. Concentrations of Nitrogen and Phosphorus

During the electrochemical cell lysis process, the concentration of nitrogen and phosphorus in the sludge supernatant can also partly reflect the status of sludge disintegration. As can be seen from Figure 6b, during electrolysis from 0–90 min, the TN concentration increased from 6.65 mg/L to 36.40 mg/L, while the organonitrogen accounted for 49.75%, reaching 18.11 mg/L. The concentrations of NH_4^+-N and NO_3^--N also increased, which were 9.12 mg /L and 9.06 mg /L, respectively. The concentration of NO_2^--N did not change significantly, which may be owing to the strong electrochemical oxidation of the electrode. NO_2^--N can be easily oxidized to NO_3^--N; thus, there was a limited accumulation of NO_2^--N. There are two potential reasons for the increase in TN. One is that proteins were released from extracellular polymers in the process of electrochemical oxidation, thus destroying the sludge bacteria micelles. The other reason is that some sludge cell walls and membranes were disrupted, leading to the outflow of intracellular nitrogen-containing substances.

As shown in Figure 6c, during electrolysis from 0 to 90 min, the concentration of TP in the sludge supernatant increased over time, rising from 2.49 mg/L to 12.64 mg/L, and the orthophosphate accounted for 92.64% of the electrolyzed product, reaching 11.71 mg/L. There are many phosphorous-containing substances, such as EPS, phospholipid bilayer of cell membranes, intracellular polysaccharides, and nucleic acids, in sludge. The increase in TP concentration in the supernatant was also attributed to these substances. In the overall cell lysis process, the growing trend in orthophosphate was very similar to that in TP, and the proportion was more than 90% for the duration of the process, illustrating that orthophosphate comprised the majority of the total phosphorus released by electrochemical cell lysis. Yang et al. [39] demonstrated that the electrochemical treatment promoted the release of phosphorus from waste activated sludge in the liquid phase.

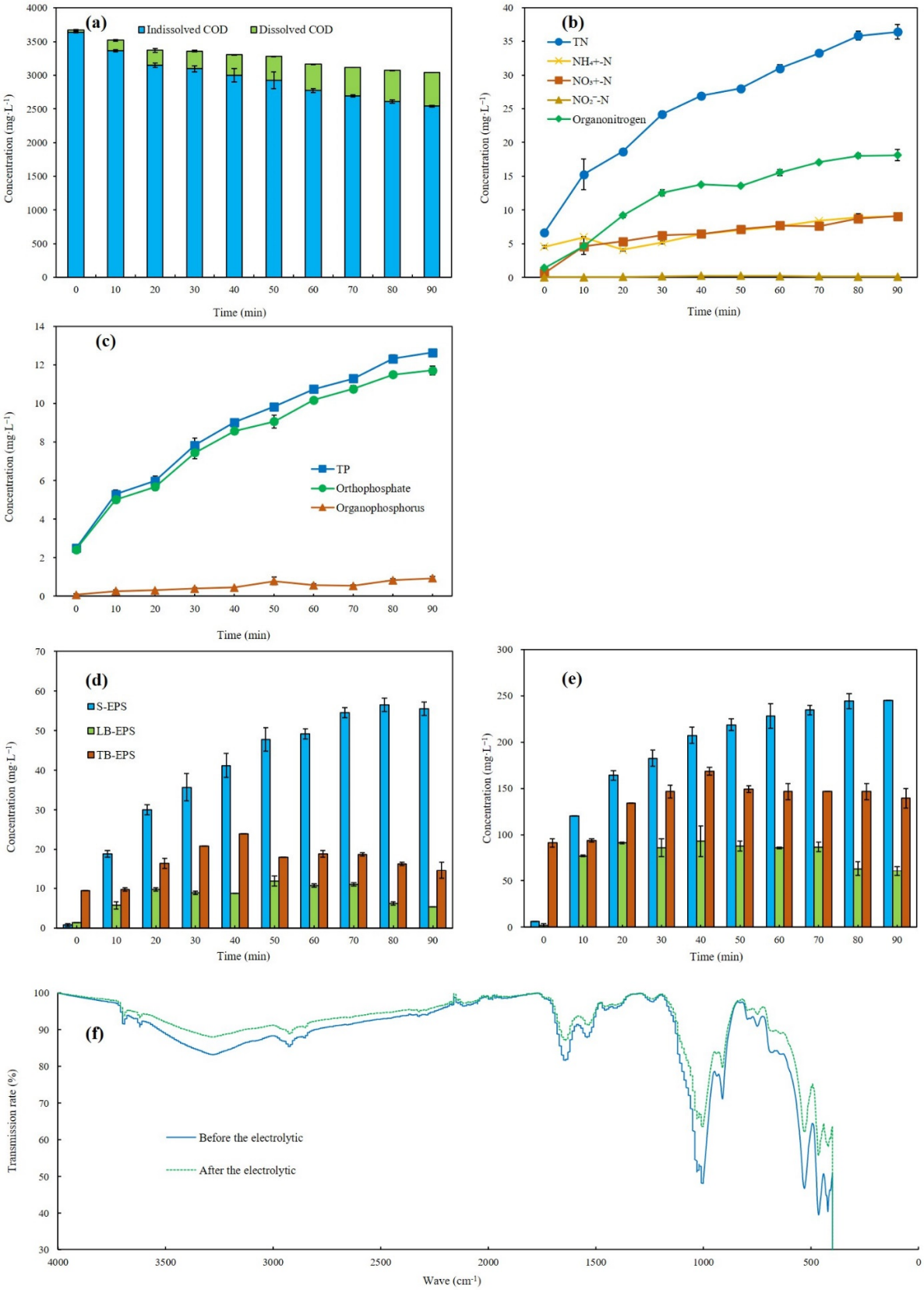

Figure 6. Changes in the concentrations of (**a**) Different types of chemical oxygen demand (COD), (**b**) Different types of nitrogen, (**c**) Different types of phosphorus, (**d**) Protein, (**e**) Polysaccharide, and (**f**) An Fourier transform infrared spectrometer (FTIR) image of sludge extracellular polymeric substances (EPS) level before and after electrolysis.

3.4.3. EPS Composition

The previous data showed that the concentration of COD, TN, and TP in the sludge supernatant increased significantly after electrolytic treatment, accompanied by changes in sludge settleability and dewaterability. These were related to the changes in sludge extracellular polymeric substances (EPS). During the electrolysis process, the force interactions between EPS components, which was very important for the colloidal stability of the floc, were disrupted [40]. EPS were mainly composed of proteins and polysaccharides, and the changes in EPS content in this study were characterized by the content of proteins and polysaccharides. In the process of electrochemical cell lysis, EPS were disrupted before cells were lysed by strong oxidation, which meant that, theoretically, the content of proteins and polysaccharides in bound EPS would continue to decrease. These substances would continue to dissolve into the supernatant with cell lysis proceeding, and the solubility of EPS would also increase on an ongoing basis. The variation trend of EPS for the in situ experiment is shown in Figure 6d,e.

S-EPS refers to soluble EPS, and LB-EPS and TB-EPS refer to loosely and tightly bound EPS, respectively. From the results for S-EPS, the content of both proteins and polysaccharides increased significantly, from 5.98 mg/L and 0.73 mg/L (before electrolysis) to 244.99 mg/L and 55.55 mg/L (after 90 min electrolysis), respectively. The increase in protein and polysaccharide content in the supernatant was one of the reasons why the sludge dewaterability decreased after electrochemical cell lysis, which has been proved by Yu et al. [41].

Three main factors affected the content of proteins and polysaccharides in the supernatant: (1) bound EPS were destroyed, and the proteins and polysaccharides released into the supernatant increased their content; (2) the cell lysis resulted in the release of intracellular substances into the supernatant, which also increased the content of proteins and polysaccharides in the supernatant; and (3) the strong oxidation of the electrode continuously oxidized large molecules of organic matter (e.g., proteins and polysaccharides) into small molecules of organic matter or even inorganic matter (CO_2 and H_2O), reducing the concentration of proteins and polysaccharides in the supernatant.

As shown in the variation trend, the content of proteins and polysaccharides increased rapidly in the first 60 min and slowed down significantly thereafter. By 90 min, the concentration of polysaccharide had decreased. It might be that electrochemical oxidation was only effective at the processing of oxidizing proteins and polysaccharides into small molecules of organic matter, but it could not cause further disruption of the sludge flocs. Therefore, the concentration of protein and polysaccharide in the supernatant increased slowly or even decreased slightly.

For LB-EPS and TB-EPS, the content of proteins and polysaccharides showed a trend of first rising and then falling, and the concentration of proteins and polysaccharides after 90 min electrolysis was higher than that before electrolysis, which differed from the theoretical prediction. Such a trend might be related to the mechanism of electrochemical oxidation on sludge: electrochemical oxidation first disrupted the structure of sludge flocs, which made EPS exposed and easier to be extracted, and then destroyed the cell structure, and finally made EPS dissolved and intracellular substances released.

Therefore, the quantitative analysis results of EPS could not fully reflect the changes in EPS composition. To further reveal the effect of electrochemical cell lysis on the sludge EPS composition, the FTIR technique was used in this study to analyze the composition of organic matter in EPS in the raw sludge and the sludge treated by electrolysis for 90 min. The experimental results are shown in Figure 6f.

The organic matter in sludge samples before and after electrolysis showed obvious absorption peaks near 3300 cm^{-1}, referring to the stretching vibration of hydroxyl (-OH) [8]; absorption peaks ranging from 1643 to 1454 cm^{-1} were related to the stretching and deformation vibrations of the amide bond C=O, C-N, and N-H in amino acids, which were peptide bonds of protein secondary structure; absorption peaks between 1027 and 1004 cm^{-1} are related to the stretching and bending vibration of C-C, C-O-C, and C-OH of

polysaccharides or carbohydrates [42]. After electrochemical cell lysis, the relative intensity of absorption peaks between 1643 and 1454 cm^{-1}, and between 1027 and 1004 cm^{-1} decreased, indicating the degradation of proteins and polysaccharides in EPS.

3.4.4. Analysis of the Mechanism of Action of Cell Lysis Using an Electrode

Based on the previously mentioned study on the changes in sludge characteristics in the electrochemical cell lysis process, this process can be illustrated as shown in Figure 7.

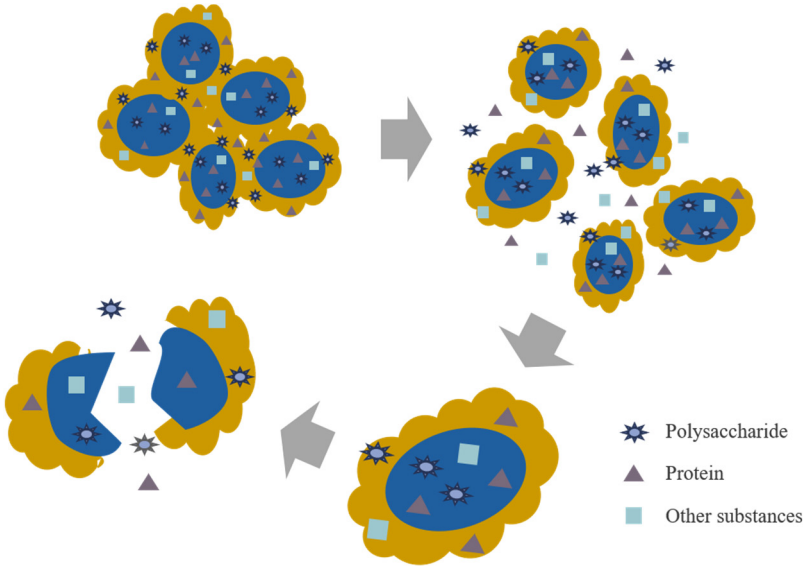

Figure 7. Schematic diagram of the electrochemical cell lysis process.

Thus, at the start of the electrochemical reaction, first, the electrocatalytic oxidation on the anode surface disrupts the sludge floc structure and the microbial micelles disintegrate into smaller particles or single cells, leading to the reduction of sludge particle size. This then affects the sludge settleability and dewaterability. In this process, a part of the bound EPS is released, leading to an increase in the concentration of proteins, polysaccharides, and other organic matter in the sludge supernatant [42]. With the electrochemical oxidation proceeding, bound EPS continues to be oxidized and dissolved, and the sludge cell wall and cell membrane are also affected [43]. Therefore, the bacterial structure is destroyed, and the destroyed cell membrane and intracellular substances are then released into the sludge supernatant, leading to a further increase in the concentration of organic substances in the supernatant. This is represented by a rapid increase in the concentration of COD, TN, and TP. Specific to reactions with various substances, electrochemical oxidation can oxidize some large molecules of carbon-containing substances to small molecules of volatile fatty acids (VFAs) [44], thereby reducing the pH value of the mixture. Regarding the change in nitrogen content in the supernatant, part of the organic nitrogen can be oxidized to inorganic nitrogen, such as NH_4^+-N, NO_2-N, and NO_3-N. NO_2^--N can be further oxidized to NO_3^--N. Regarding increased TP in the supernatant, most of the released organic phosphorus can be oxidized to PO_4^{3-} [42]. Some organic substances released into the supernatant in the electrochemical cell lysis process can be directly mineralized to end products, CO_2 and H_2O [20].

4. Conclusions

In this study, the Ti/SnO_2-Sb electrode was determined to be more suitable for sludge cell lysis. Under the optimal electrolysis parameters, the sludge disintegration rate and cell lysis rate reached 25.35% and 20.15%, respectively, effectively improving sludge dewaterability. Based on the changes in sludge characteristics, the mechanism of the electrochemical cell lysis was concluded to be (1) the disruption of sludge floc and micelle structure and (2)

the destruction of sludge cells. These two processes led to an increase in the concentration of organic matter in the sludge supernatant which was simultaneously accompanied by some mineralization of organic matter.

Supplementary Materials: The following are available online at https://www.mdpi.com/2073-4441/13/5/616/s1, Figure S1: Effect of electrochemical cell lysis on MLSS, MLVSS and IS of sludge, Figure S2: Change of pH of sludge mixture during electrochemical cell lysis, Figure S3: Changes of sludge settling performance during electrochemical cell lysis, Figure S4: Changes of sludge dewatering performance in electrochemical cell lysis process.

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