Development of Novel Anaerobic Digestion System for Multipurpose Sewage Sludge Treatment

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Abstract

Increasing amount of sewage sludge is produced annually from wastewater treatment plants (WWTPs) with the rapid urbanization and population growth worldwide, while its treatment and disposal has become one of the serious challenges due to the increasingly stringent regulation and the high disposal cost. Anaerobic digestion (AD) has been widely applied to stabilize organic components and recover energy (CH_4) simultaneously from sewage sludge before sludge dewatering and final disposal. Although sludge volume can be reduced to some extent, AD of sludge is still facing challenges due to its characteristics like high water content and poor dewaterability of digestate, and difficulty in its posttreatment due to the high nitrogen (N) and phosphorus (P) levels, which could also be recovered for agricultural use. Biogas recirculation has been utilized as a mixing strategy for AD process. Its utilization in the internal circulation anaerobic reactor for wastewater treatment has been proven to have positive effect on anaerobic sludge granulation, and wastewater treatment efficiency with high quality biogas production (~ 80%CH_4). On the other hand, multivalent cations have been demonstrated their effectiveness on binding with bacteria, resulting in enhanced sludge dewaterability. In addition, struvite precipitation by adding Mg salt is a commercialized method for simultaneous P and N recovery from liquid digestate in WWTPs due to its marketable value as slow releasing fertilizer, and $FeCl_3$ is commonly used in WWTPs for various purposes including organics, P and suspended solids removal, and sludge conditioning.

This dissertation attempted the combination of biogas recirculation with chemical addition ($MgCl_2$ or $FeCl_3$) in one AD reactor, aiming to realize biogas upgrading, nutrients conservation and sludge conditioning simultaneously. The effects of biogas recirculation and chemical addition on biogas production, methane content, nutrients conservation and sludge dewaterability were investigated through both batch and semi-continuous AD experiments. Comparison was also conducted on the effects of divalent Mg^{2+} and trivalent Fe^{3+} addition at the same equivalent concentration on the performance of the AD system.

The main results from this study can be summarized as follows:

(1) The intermittent biogas recirculation coupling with $MgCl_2$ addition enhanced methane content to 86% after 17 days' batch AD test. The results of carbon profiles and increased pH values revealed that the reduced carbon dioxide might be simply dissolved in sludge liquor or adsorbed on sludge particles. The bioavailable P fraction in sludge was not affected by $MgCl_2$ addition during AD. The $MgCl_2$ addition at a Mg:P_{ortho} molar ratio of 1:1 conserved 87% of soluble P and 19% of ammonia N in the solid phase of digestate. The struvite formed in the

reactor was identified and confirmed by X-ray diffraction (XRD) and scanning electron microscope (SEM). The dewaterability of digested sludge was enhanced by 37% under $MgCl_2$ addition (35.8 mg-Mg/g-TS) together with biogas recirculation. A strong correlation between extracellular polymeric substances (EPS) and sludge dewaterability was also found according to Pearson's correlation analysis. A shorter hydraulic retention time (HRT, 17 days) with biogas recirculation was proposed to enhance methane content and improve sludge dewaterability.

(2) $FeCl_3$ addition at a dosage of > 600 mg-Fe/L (49 mg-Fe/g-TS) further enhanced methane content to 88% during batch AD with biogas recirculation. A $FeCl_3$ addition of 900 mg-Fe/L did not affect the methane yield during 30 days' batch AD but significantly reduced the biogas production rate constant (k) and maximum methane production rate (μ) estimated from the first-order kinetic model and modified Gompertz model. The formation of carbonate precipitates might be the reason for the further increased methane content with the increase in $FeCl_3$ dosage. $FeCl_3$ addition at an Fe:P_{ortho} molar ratio of 1.5:1 (900 mg-Fe/L) achieved 98.7% conservation of P in the solid phase of digestate. The sludge dewaterability and settleability were enhanced by 79% and 56% when $FeCl_3$ addition was conducted at 900 mg-Fe/L (68 mg-Fe/g-TS) together with biogas recirculation during the batch AD test.

(3) The intermittent biogas recirculation during the semi-continuous AD test enhanced both methane content and methane production. $MgCl_2$ addition almost did not contribute to P conservation during this semi-continuous AD due to the lower pH value (7.00) and relatively low ortho-P concentration (~ 60 mg/L) in influent sludge, while $FeCl_3$ addition achieved 97% of P conservation in the solid phase of digestate. The sludge dewaterability was enhanced by 76% and 94% under the addition of $MgCl_2$ (12 mg-Mg/g-TS) and $FeCl_3$ (19 mg-Fe/g-TS), respectively. The analysis on archaeal and microbial communities revealed that biogas recirculation enhanced the relative abundance of both acetoclastic and hydrogenotrophic methanogens, contributing to the increased methane yield and methane content in biogas.

The results from this novel AD system suggest the possibility to simplify the sludge treatment facilities in WWTPs, which can potentially realize simultaneous biogas upgrading, P conservation and sludge conditioning in one AD reactor. This study also provides scientific reference for a new concept of sludge management in WWTPs.

Key words: Sewage sludge; Anaerobic digestion; Biogas recirculation; Phosphorus conservation; Sludge dewaterability

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Abbreviations and acronyms

AD	Anaerobic digestion
AHR	Anaerobic hybrid reactor
ANOVA	Analysis of variance
AP	Apatite phosphorus
BSA	Bovine serum albumin
COD	Chemical oxygen demand
CST	Capillary suction time
DNA	Deoxyribonucleic acid
EBPR	Enhanced biological phosphorus removal
EPS	Extracellular polymeric substances
FIT	Feed-in-tariff
FNA	Free nitrous acid
GAOs	Glycogen accumulating organisms
HRT	Hydraulic retention time
IC	Internal circulation
ICP	Inductive coupled plasma
IP	Inorganic phosphorus
K_{sp}	Solubility product
LB-EPS	Loosely bound extracellular polymeric substances
MLIT	Ministry of Land, Infrastructure, Transportation and Tourism
MOE	Ministry of the Environment
N	Nitrogen
NAIP	Non-apatite inorganic phosphorus
OLR	Organic loading rate
OP	Organic phosphorus
Ortho-P	Orthophosphate
P	phosphorus
PAC	Polyaluminum chloride
PAM	Polyacrylamide
PAOs	Polyphosphate accumulating organisms
PCR	Polymerase chain reaction
PN	Proteins

PS	Polysaccharides
SAO	Syntrophic acetate-oxidizing
SEM	Scanning electron microscope
S-EPS	Soluble extracellular polymeric substances
SI	Saturation index
SMT	Standards, Measurements and Testing
SRF	Specific resistance to filtration
SVI_{30}	Sludge volume index in 30 min
TB-EPS	Tightly bound extracellular polymeric substances
TKN	Total Kjeldahl nitrogen
TN	Total nitrogen
TOC	Total organic carbon
TP	Total phosphorus
TS	Total solids
VFAs	Volatile fatty acids
VS	Volatile solids
WWTPs	Wastewater treatment plants
XRD	X-ray diffraction

Chapter 1 Introduction

1.1 Sewage sludge treatment

1.1.1 Current situation

Increasing amount of sewage sludge is produced annually from wastewater treatment plants (WWTPs) with the rapid urbanization. The treatment and management of sewage sludge has become one of the serious challenges due to the increasingly stringent regulations and the high disposal cost, which usually accounts for 50% of total operation cost in WWTPs (Zhen *et al.*, 2017). According to the latest data released by the Ministry of the Environment (MOE) in Japan, around 167.32 million tons of sludge were generated in 2016, which accounted for 43.2% of the total industrial wastes (MOE, 2019). The recycle ratio of sewage sludge reached to 73% (dry solid basis) in 2017; however, only 32% was used as biomass (biogas, biofuel and land application) (MLIT, 2017). In China, even serious problem is that 83% of raw sludge are improperly dumped without any treatment (Yang *et al.*, 2015). Besides, large amount of sludge is disposed directly after mechanical dewatering with moisture content higher than 80%, while the water content of sludge cake for safety disposal should be lower than 60% (Wang *et al.*, 2010). The improper treatment would result in severe environmental pollution due to the high nutrients content, pathogens and heavy metals containing in sewage sludge. An increasing attention paid to environmental issues also promoted the development of various technologies and policies for resources recovery and pollutants removal from sewage sludge. As cost-effectiveness is one of the most important factors dictating the practical application of the technologies to a great extent, both environmental and economic aspects should be considered in the real world of sewage sludge management.

1.1.2 Problems unsolved

To make efforts on the development of novel technologies for sustainable sewage sludge treatment, the existing difficulties and problems should be clearly addressed. Taking a local WWTP in Chikusei, Ibaraki, Japan as an example, the primary sludge collected from primary clarifier and excess sludge collected from secondary clarifier are mixed together as thickened sewage sludge and treated by anaerobic digestion (AD). Then, the digested sludge is conditioned by chemical coagulants ($FeCl_3$ and CaO) and mechanically dewatered to reduce sludge volume. The dewatered sludge cake is finally transported to specific places for final disposal, such as landfill, incineration or composting. The liquid fraction of digested sludge after dewatering usually flows back to aeration tank for nutrients removal.

AD of sewage sludge is the most studied treatment among the above-mentioned processes, since it can not only stabilize sludge, reduce sludge solids, remove odor and pathogens, but also produce biogas that can compensate part of energy required for sewage sludge treatment (Appels *et al.*, 2008). AD mainly consists of several successive steps, i.e. hydrolysis, acidogenesis, acetogenesis and methanogenesis. The complex large molecular organics are decomposed by different groups of microbes to smaller molecules and finally converted to methane (CH_4) and carbon dioxide (CO_2). Sewage sludge has very complex composition, which usually consists of flocs formed by microbial aggregates, extracellular polymeric substances (EPS), organic and inorganic particles, etc. The microbes are protected by the cell wall and their secretions, EPS, which makes it difficult to be hydrolyzed during AD (Zhen *et al.*, 2017). The presence of EPS also largely influences sludge dewaterability since EPS are negatively charged and have strong affinity to water. They bind with cells and water through complex interaction to form a stable gel-like structure which resists to dewatering and degradation (Sheng *et al.*, 2010). In the last decades, numerous studies have investigated various treatment methods to accelerate hydrolysis and enhance biogas production during AD, or to improve sludge dewaterability, thus easing the disposal of sewage sludge and lowering the operation cost by the enhanced biogas production (Kim *et al.*, 2003; Neyens and Baeyens, 2003).

On the other hand, the large proportion (40-50%) of CO_2 in biogas reduces the caloric value and increases biogas volume for storage or transportation. Though the biogas containing CH_4 higher than 60% could be directly used for heating or electricity generation, the conversion efficiency was relatively low due to the large proportion of incombustible gas (Hakawati *et al.*, 2017). The upgrading or cleaning of biogas could also generate new possibilities for its uses as substitute of fossil fuel, such as vehicle fuel, domestic stoves, fuel cells or being injected into natural gas grids (Sun *et al.*, 2015). However, the total cost for biogas upgrading is extremely high regardless of upgrading methods when raw biogas capacity of the plant is lower than 1000 m^3_{STP}/h (Miltner *et al.*, 2017). There are averagely 330 million m^3/year biogas produced from sewage sludge in Japan, in which around 82% is utilized for electricity generation (37%) or digester heating (23%), and the other 18% is burned and wasted (MLIT, 2018). The government has also developed various strategies for promoting the energy recovery efficiency from biogas produced in WWTPs, including biogas upgrading for injection into natural gas grid or supply vehicle fuel for public buses (MLIT, 2010a). The real application is still limited due to the high costs for biogas upgrading and network buildup.

Furthermore, there are 10% of phosphorus flows in sewage system annually in Japan, where is totally reliant on imported phosphorus sources (MLIT, 2017). Phosphorus is one of the essential nutrients for food production. It is mainly extracted from phosphorus rock, which is non-renewable and mainly reserves in China, the US and Morocco. It was estimated that the global phosphorus reserves will be depleted in 50-100 years (Cordell *et al.*, 2009). While rejected liquid, the liquid fraction of digested sludge after dewatering, usually contains high concentrations of phosphorus and nitrogen, which need to be further treated by activated sludge process, thus increasing the operation cost in WWTPs. It was reported that even several WWTPs abolished AD tank every year before 2004 in Japan due to the high cost involved (MLIT, 2004). Until 2015, only around 10% of phosphorus flowed into sewage system was recovered as fertilizer in Japan.

From the above, sewage sludge contains abundant resources that could be recovered as energy and nutrients, while the high operation cost of over 50% involved in sludge treatment in WWTPs slowed down the application of existing technologies for sustainable sewage sludge treatment. Sludge dewatering process is the most difficult and costly part in WWTPs, while of great importance to reduce the sludge volume for final disposal. Biogas upgrading and phosphorus recovery could enhance the energy recovery efficiency and compensate the depletion of phosphorus sources in Japan, where has no domestic phosphorus rock. Thus, it is requisite to develop novel technologies for sustainable sludge treatment with low cost, mainly on sludge dewatering, biogas upgrading and phosphorus recovery.

1.2 Sludge dewatering

1.2.1 Current methods for sludge dewatering

The sludge after thickening and stabilization in WWTPs generally needs to be dewatered by natural or artificial methods according to the dimension and financial level of WWTPs. The natural methods use the principle of gravitation and evaporation to reduce water content in the sludge (Gurjar and Tyagi, 2017). However, borrowing the forces from nature is time consuming. Large land area is also required for the treatment of huge amount of sewage sludge. In addition, the leachate, which is rich in nitrogen, phosphorus, and perhaps heavy metals and pathogen, would cause severe pollution on farmland and water system. Air pollution also might be occurred due to the evaporation of odorous and harmful gases, such as NH_3, H_2S, etc. The artificial methods are using the principle of pressure difference (filter press) and artificial gravity (centrifuge). Chamber filter press, belt filter press and centrifuge/decanter are the mostly used technologies for sludge dewatering. Chamber filter press can produce filter cake with highest solid content of 40-70%, followed by 25-35% solid content by centrifuge/decanter and 18-25% solid content by belt filter press (Powell, 2016). Though the solid content obtained from belt filter press is the lowest, it is generally used in WWTPs because of its effectiveness on high-volume waste streams. Besides, in order to reduce the water content of sludge cake to the required value (i.e. 50% for incineration, in China) for further disposal, sludge conditioning is usually prerequisite before dewatering process in WWTPs.

1.2.2 Sludge conditioning

The water fraction of sludge can be divided into free water and bound water. The free water can be easily removed by thickening and dewatering process. However, the bound water is closely combined with the sludge particles by physical or chemical forces, which makes the dewatering process difficult and costly. Thus, sludge conditioning is often used to easier the dewatering process. Chemical addition and thermal treatment are the general methods that has applied in WWTPs. However, the improper use, especially overuse of the conditioners, such as ferric salts, lime and organic polymers may pose negative effect on our environment during disposal process of sludge. The high energy consumption involved in thermal treatment also limits its applicability at industrial scale. Up to now, still many researchers are focusing on various methods that can enhance the sludge dewaterability. In general, sludge conditioning methods can be divided into physical, biological and chemical methods. The literatures review and the corresponding mechanisms involved in each method are summarized as below.

(1) Physical conditioning

Physical conditioning, namely, is to enhance sludge dewaterability through physical treatment, such as thermal, ultrasonic and microwave treatments, etc., or via addition of physical conditioners. Thermal treatment is the most commonly used physical method for enhancing dewaterability at industrial scale (Wang *et al.*, 2010). It was reported that dewaterability can be significantly improved when treatment temperature higher than the threshold value (120-150°C). The high temperature can destroy microbial cell structure, release

EPS, bound water and intracellular materials. The soluble organic content in sludge can be also increased, which favors biogas production via anaerobic digestion. The produced biogas can counteract part of high energy input during thermal treatment, making the process applicable at industrial scale. Li *et al.* (2018) studied the energy balance of a pilot scale process combined with hydrothermal pretreatment, AD and pyrolysis for sludge dewatering and co-production of biogas and biochar. This work demonstrates that the process is effective for full resource reuse of sewage sludge. Other physical treatments, like ultrasonic (Ruiz-Hernando *et al.*, 2013), or microwave treatment (Wojciechowska, 2005), have been also investigated, however, its effect on dewaterability enhancement was not so significant unless being used together with other conditioning method (Zhu *et al.*, 2018).

Nowadays, physical conditioners have drawn more and more attention, due to its environmental friendliness. Industrial and agricultural residues, such as cement, coal fly ash, rice husk, sawdust, etc., have been widely studied. They can work as skeleton builders for sludge, reducing compressibility of sludge and forming water release channel. Besides, the addition of agricultural residues, other than chemical conditioners, can avoid the decrease of calorific value of sludge cake, favoring the incineration disposal process (Wang *et al.*, 2017b). However, using physical conditioners solely usually has limited impact on dewaterability enhancement because of the little amount of free water existed in sludge, unless using chemical or physical treatment together to release bound water and EPS from sludge. “Cells disintegration-floc reconstruction-skeleton building” system has been proven as an effective method for sludge conditioning. Zhu *et al.* (2018) used the combination of ultrasound, traditional cationic polyacrylamide (PAM) and rice husk conditioning, resulting in 90.50% decrease of specific resistance to filtration (SRF) compared with that of raw sludge. Wu *et al.* (2017) reported a 99.03% decrease of SRF when using $KMnO_4$, $FeCl_3$ together with sludge cake biochar. The modification on physical conditioners by using acid, $FeCl_3$, etc. also have been widely studied (Wu *et al.*, 2016b; Zhong *et al.*, 2017).

(2) Microbial conditioning

Thomas *et al.* (1993) first reported the possibility by using enzymatic treatment to improve sludge dewaterability. Ayol (2005) found that only 10 mg/L of enzyme addition in polymer (300 mg/L) conditioned sludge could double the solids concentration in sludge cake. The EPS analyses of this study indicated that enzyme product additions improved the degradation of extracellular protein and polysaccharide. Furthermore, Dursun *et al.* (2006) revealed that the enzyme treatment improved dewaterability by weakening the gel structure of the sludge floc through the hydrolysis of EPS, while only in case of filtration processes with low shear stress. On the contrary, the higher shear force involved in centrifugation leads to floc deterioration.

In recent years, bioleaching also has been found as a potential method for improving sludge dewaterability. Originally, bioleaching is used for the extraction of metals from minerals through microbial oxidation process. Acidophilic iron-oxidizing and sulfur-oxidizing bacteria are involved in bioleaching, thus, Fe^{3+} and S^0 usually need to be added as energy substrate. The process is also quite efficient in removing metals from sewage sludge. Song and Zhou (2008) claimed that the specific resistance was reduced by 4.6 times after bioleaching. Liu *et al.* (2012) compared bioleaching with other physical and chemical conditioning methods. The results showed that the SRF or capillary suction time (CST) was decreased by 93.1% or 74.1% after bioleaching, which is similar with chemical conditioning by the addition of $FeCl_3$ and CaO, but much more effective than physical conditioning methods. Besides, the filtrate of bioleached sludge contains lowest concentration of chroma, chemical oxygen demand (COD), total nitrogen (TN) and total phosphorus (TP), further reducing the cost for filtrate disposal after dewatering process. Further work by using *Acidithiobacillus ferrooxidans* culture as sludge conditioner was studied by Murugesan *et al.* (2014), in order to avoid the over-acidification, which would deteriorate sludge dewaterability during bioleaching process. The results indicated that the culture and filtrate of *A. ferrooxidans* can be used as an effective sludge conditioner. The polymeric inorganic ferric flocculant secreted in the culture has high affinity to the sludge particles, thus resulting in rapid aggregation of sludge particles.

On the other hand, bioflocculants produced from sludge (Guo and Ma, 2015), rice stover (Guo and Chen, 2017) or potato starch wastewater (Guo *et al.*, 2015) by using specific microbial strains are raising attention nowadays due to its environmental-friendliness and negligible secondary pollution. The utilization of waste residues could further lower the operation cost for sludge dewatering process.

(3) Chemical conditioning

Because of the advantages of low cost and high efficiency, chemical conditioning is the most common method used for sludge conditioning and dewatering process. Briefly, the process improves dewaterability by simply adding chemicals. From the simplest acids (HCl) to metal salts (e.g. $FeCl_3$, $Al_2(SO_4)_3$, lime, polyaluminum chloride (PAC)), and synthetic polyelectrolytes (e.g. PAM), they have already been utilized in many WWTPs. The ferric and calcium salts are the most common chemicals used in WWTPs. According to the mechanisms involved in chemical conditioning, the chemicals used for sludge conditioning can be divided into two types. Firstly, acids and strong oxidants ($KMnO_4$, H_2O_2, Fenton's reagent, etc.), which could help release EPS from sludge particles or break cell walls to release intracellular materials. Simultaneously, the bound water trapped with EPS and in microbial cells also can be released as free water, becoming easily separated from sludge particles. Wang *et al.* (2017a) mentioned that acid conditioning also can release more metal cations from intracellular material, which can provide positive charge for sludge particles, resulting in increased zeta potential and particle size. They obtained 64% decrease of CST and 74.8% decrease of SRF when adjusting the pH to 2.6 by HCl. Chen *et al.* (2001) reported that the water content of sludge dewatered with filtration decreased from 83.4% (no treatment) to 76.08% when treated with sulfuric acid at pH 2.5, which showed better dewatering efficiency than the conventional chemical conditioners (1.2 g/L $FeCl_3$ or 3 g/L CaO addition). Secondly, inorganic coagulants, including multivalent metal salts and polymeric flocculants, can enhance sludge dewaterability by charge neutralization. Metal cations can provide positive charge, while the long-chain molecular structure of polymeric flocculants can adsorb and bridge small sludge particles into large particles and neutralize their negative charges (Guan *et al.*, 2016; Wang *et al.*, 2018b).

Recently, polymeric flocculants, especially PAM, have been proven as the most efficient flocculant for sludge conditioning. Guo and Ma (2015) compared the dewatering efficiency among $FeCl_3$, $Al_2(SO_4)_3$, PAC and PAM. The results showed that only 0.15 g/L addition of PAM can decrease SRF by 71.68%, while 8.0 g/L $FeCl_3$, 8.0 g/L $Al_2(SO_4)_3$, and 4.0 g/L PAC addition decreased SRF by 55.28%, 58.41% or 66.37%, respectively. Besides its high charge density and coagulation efficiency, the stable hydrolysis, and alleviative impact on equipment and environment also make it procure more recognition and wider application compared to inorganic chemicals.

1.3 Phosphorus recovery/removal technologies

Phosphorus is essential for human being. However, the global phosphorus reserves are limited, and the remaining phosphorus rock was estimated to be exhausted within next 50-100 years (Cordell *et al.*, 2009). It has been reported that 15-20% of world phosphorus demand could be satisfied by recovering phosphorus from domestic wastewater (Yuan *et al.*, 2012). On the other hand, phosphorus is the main factor that could result in severe eutrophication in waterbodies even at very low concentration of 0.02 mg/L (Correll, 1999). As a result, phosphorus removal from wastewater to an increasingly stringent discharge standard has become an enormous challenge in WWTPs. From above, the definitions of phosphorus recovery or removal could be clarified. Many review papers have also summarized various phosphorus recovery/removal technologies that are commercially used or studied in laboratory (Morse *et al.*, 1998; Melia *et al.*, 2017). Basically, phosphorus recovery/removal technologies could be classified as biological, physical and chemical methods.

1.3.1 Enhanced biological phosphorus removal (EBPR)

EBPR process was developed by adding an anaerobic/anoxic zone before activated sludge process. The main functional bacteria are called as polyphosphate accumulating organisms (PAOs), which could take up volatile fatty acids (VFAs), release phosphate under anaerobic condition and absorb excess phosphate as polyphosphate from wastewater under aerobic condition. EBPR process has been considered as a more sustainable and cost-effective method for phosphorus removal in WWTPs due to the high phosphorus bioavailability in the produced sludge and less or no chemical addition. However, the stability of the microbial process has become a problem in full-scale application. Process inefficiency usually happened due to the competition of glycogen accumulating organisms (GAOs) for organic carbon sources (Yuan *et al.*, 2012). Consequently, many researches have been focused on finding the optimized conditions for PAOs growth. It has been found that PAOs activity could be maintained at pH 8 (Oehmen *et al.*, 2005), relatively lower temperature (e.g. 20°C) (Panswad *et al.*, 2003) with propionate as carbon source (Oehmen *et al.*, 2006), while the growth of GAOs was restricted. Under the desired condition, phosphorus removal efficiency could reach to over 90% (Yuan *et al.*, 2012).

The produced phosphorus enriched sludge from EBPR process could be directly applied as fertilizer after dewatering or be solubilized and followed by other phosphorus recovery technologies, such as crystallization. Though the dewatered EBPR sludge has been demonstrated as effective as the mineral phosphorus fertilizer, the easily degradable organics and possible contaminants in sludge might affect soil properties and food quality (Singh and Agrawal, 2008). Thus, the most commercialized methods, crystallization, has usually been combined with EBPR process to maximum resources recovery efficiency (Yuan *et al.*, 2012). Magnesium and calcium salts are mostly used for crystallization from digested sludge or the separated liquid digestate in the developed commercial processes, such as Airprex, NuReSys, Phosnix, FIX-Phos, and so on. The phosphorus in EBPR sludge could be released by AD or chemical extraction methods. Through AD process, the biologically bound phosphorus in EBPR sludge could be released. Besides, the easily degradable organics could be stabilized, with renewable energy produced as biogas. However, the released phosphate is also easily precipitated with metal ions in sludge, reducing the phosphorus recovery efficiency. Chemical extraction by using acid is more efficient; however, the separation of heavy metals and phosphate is also necessary. The chemical addition and nanofiltration for cations removal makes the process costly (Melia *et al.*, 2017). In order to increase the phosphorus recovery efficiency, various technologies have been investigated in recent years. For example, hydrothermal treatment of digested sludge has been demonstrated as an efficient method for phosphorus and nitrogen release (Yu *et al.*, 2018). Incineration of sludge at around 1000°C with $MgCl_2$ or $CaCl_2$ addition could increase phosphorus bioavailability in sludge ash, and simultaneously remove organic pollutants and heavy metals volatilized as the chlorinated salts (Adam *et al.*, 2009).

1.3.2 Adsorption

Physical methods for phosphorus removal include ion exchange, membrane filtration, magnetic attraction, adsorption and so on. Among these technologies, adsorption is the most studied one. Some newly developed adsorbents have shown high sorption capacity over 100 mg-P/g (Choi *et al.*, 2012; Lin *et al.*, 2018). Although the adsorption process was rarely applied in industrial scale due to the high cost, the excellent performance may promote its application in the future with consideration of the increasingly stringent requirement on phosphorus discharge standard. In addition, the increasing trends by using agricultural by-products or industrial wastes as the raw material for adsorbents could potentially increase the feasibility of adsorption technologies for industrial-scale application (Yao *et al.*, 2011). Yet, more research needs to be done to balance the adsorption capacity and the potential environmental risk brought about by used adsorbents, since the adsorbents with excellent adsorption capacity usually contains high content of heavy metals which are used for adsorbent modification.

1.3.3 Chemical precipitation and crystallization

Phosphorus removal from wastewater was first achieved by chemical precipitation in the 1950s (Morse *et al.*, 1998). By simply adding metal salts of Fe, Al or Ca, the soluble phosphate ions in wastewater could be precipitated with the divalent or trivalent cations. Fe salts have been most commonly used since their coagulation effects also could increase sludge settleability, reduce suspended solids and organic content in wastewater (De Gregorio *et al.*, 2010). Chemical precipitation is still the most widely used process for phosphorus removal in WWTPs nowadays due to its simplicity and flexibility. It can be applied in any stages during wastewater treatment. However, excess amount of chemicals usually need to be added to remove phosphorus efficiently, also resulting in the generation of large amount of chemical sludge that is difficult to be reused and disposed (Melia *et al.*, 2017). Then, crystallization was developed for the controlled chemical precipitation through adding seed materials. Through this process, a more marketable end product with high purity could be produced at optimized conditions. Most of the commercial processes for phosphorus recovery in WWTPs are using crystallization reactor as a side stream of sludge treatment to produce struvite or Ca-bound phosphorus for commercial use.

1.4 Biogas upgrading

Biogas mainly contains methane and carbon dioxide with small amounts of water, H_2S, NH_3, N_2, O_2 and so on. Biogas production has been rapidly increased in recent years due to the increasing concern on utilization of bioenergy and the wide application of AD for organic wastes treatment. To demand various utilization ways, the removal of incombustible and harmful gases in biogas became more and more important. The number of upgrading plants in the world was rapidly increased from around 20 before 2001 to over 200 in 2012 (Bauer *et al.*, 2013). Several researchers already summarized the existing biogas upgrading technologies that have been utilized in industries and the novel technologies under development (Sun *et al.*, 2015; Miltner *et al.*, 2017).

The separation of CO_2 from biogas is mainly based on the different physical properties, such as the solubility (water/organic solvent absorption), molecular size and condensing temperature. Though most of biogas upgrading technologies could remove CO_2 and other trace gases simultaneously, the pre-removal of trace gases, especially H_2S, is usually necessary to avoid the corrosion or other problems on upgrading equipment. However, the high capital cost and operation and maintenance cost of all the technologies limited their application in small-scale industries with raw biogas capacity lower than 500 Nm^3/h (Miltner *et al.*, 2017). The concept of in-situ methane enrichment was first proposed in 1990s (Hayes *et al.*, 1990), while the application is still under development at pilot scale. Due to the high solubility of CO_2 in water, the digestate was used as the absorbents in this concept. Through recirculating digestate in AD tank after CO_2 being desorbed by air stripping, a high CH_4 content in biogas could be realized. Nordberg *et al.* (2012) examined the pilot-scale in-situ methane enrichment most recently, achieving 87% of CH_4 content with a CH_4 loss of 8%. Though the investment cost of in-situ methane enrichment system has not been investigated, the additional equipment for CO_2 desorption and energy consumption for digestate pumping should be still expensive. Usually, the upgraded biogas is used as vehicle or injection into natural gas grid, which have strict requirement on methane content in biogas (Sun *et al.*, 2015). In small-scale industries, the produced biogas is mainly used for heating or gas engine. The raw biogas with around 60% of CH_4 content could be directly used. Although the increased methane content could enhance the engine or heating efficiency, the investment on building additional facilities for biogas upgrading are not really necessary. With all the above considerations, biogas recirculation inside the AD tank may be more economically feasible to achieve the similar targets.

1.5 Biogas recirculation

The concept of biogas recirculation (or sparging or lift) has been utilized in the internal circulation (IC) reactor system for wastewater treatment, which can produce high quality biogas (80% CH_4) and generate well settleable granular sludge (Pereboom and Vereijken, 1994). Biogas recirculation has also been used as a mixing alternative for enhanced biogas production in AD reactor, enhanced biomass accumulation in anaerobic hybrid reactor (AHR) or alleviated fouling problem in anaerobic membrane bioreactor, which has been summarized in Table 1-1 (Suvajittanont and Chaiprasert, 2003; Ersahin *et al.*, 2016). The biogas production rate was increased from 1.5 to 1.8 L/(L·d) when the mechanical agitation of 30 rpm was replaced by biogas recirculation of 24 L/10 min both with 6-h intervals during AD of piggery slurry at an organic loading rate (OLR) of 3.9 g-VS/(L·d) (Lee *et al.*, 1995). The attached biomass on the media in AHR was increased by 16.3% with biogas recirculation of 2.6 L/(L·d) (Suvajittanont and Chaiprasert, 2003). Both biogas yield and COD removal were enhanced during AD of dairy manure at an OLR of 4.6 g-VS/(L·d) under continuous biogas recirculation of 750 mL/min (Demirer and Chen, 2005). Karim *et al.* (2005a) indicated that the continuous biogas recirculation at 1 L/min could significantly improve the methane production from animal waste at a higher OLR (3.24 g-VS/(L·d)). Siddique *et al.* (2015) obtained 29% increase in methane yield with biogas recirculation rate of 24.14 L/d during the anaerobic co-digestion of petrochemical wastewater and activated manure at an OLR of 13.3 g-COD/(L·d). Most recently, Latha *et al.* (2019) investigated the different mixing modes during AD at a high OLR of 6 g-VS/(L·d). The results indicated that intermittent biogas recirculation of 2000 mL/min could significantly enhance the biogas yield with a higher methane content of 88% achieved. From the above reports, biogas recirculation could be a promising cost-efficient mixing strategy during AD due to the possibility on enhanced biogas yield and methane content in biogas. However, the information relating to the changes of sludge properties and CH_4 content in the AD system with biogas recirculation for sewage sludge treatment is still limited.

1.6 Research objective and thesis structure

The treatment and disposal of sewage sludge, the main by-product during wastewater treatment, accounts for more than half of the total operation cost in WWTPs. On the other hand, the enriched nutrients in sewage sludge have attracted more and more attention nowadays. The increasing stringent environmental regulation also promoted the development of various technologies for sustainable sewage sludge treatment. However, for small-scale WWTPs, it would be financially deficit if building additional facilities for other purposes, such as phosphorus crystallization, sludge conditioning, biogas upgrading and so on. Thus, it’s requisite to develop new and simple systems for multipurpose sewage sludge treatment with low operation cost. The objective of this research is to establish a novel AD system for sewage sludge treatment, aiming to increase methane content in the biogas, maintain highly bioavailable resources (especially P) in the treated sludge, and enhance sludge dewaterability to serve multiple purposes. The concept of the novel AD system and the thesis structure are presented in Fig. 1-1. The thesis was divided into the following 6 chapters:

(1) Chapter 1 Introduction

Firstly, the current situation and the existing problems during sewage sludge treatment was stated. Secondly, the current methods for sludge dewatering and the state-of-the-art of sludge conditioning was summarized in detail. Thirdly, the technologies for phosphorus recovery or removal from wastewater or sewage sludge and their pros and cons were addressed. Further, the current technologies and development trends for biogas upgrading were put forward. The literatures relating to biogas recirculation in AD process was reviewed. Finally, the objective of this study and the thesis structure were arrived.

(2) Chapter 2 Effect of biogas recirculation coupling with $MgCl_2$ addition on anaerobic digestion of sewage sludge

The batch AD experiment was conducted to investigate the effect of biogas recirculation coupling with $MgCl_2$ addition on biogas production, methane content, nutrients conservation and sludge properties during AD of primary sludge. Mg could easily precipitate as struvite ($MgNH_4PO_4 \cdot 6H_2O$) with ammonia and orthophosphate (ortho-P), which are rich in digested sludge. Struvite crystallization has also been utilized in most of the commercial phosphorus recovery processes, since the ammonia and ortho-P could be recovered simultaneously, and the formed struvite could be used as slow releasing fertilizer with marketable value (Melia *et al.*, 2017). Thus, divalent cation Mg^{2+} was chosen in this Chapter to realize the nutrients conservation and enhance sludge dewaterability in one AD reactor.

(3) Chapter 3 Effect of biogas recirculation coupling with $FeCl_3$ addition on anaerobic digestion of sewage sludge

The effect of biogas recirculation coupling with $FeCl_3$ addition on biogas production, methane content, nutrients conservation and sludge properties during AD of primary sludge was explored through batch AD experiment. Different concentration of $FeCl_3$ addition during AD with or without biogas recirculation was also studied to explore the changes on methane content and sludge properties. $FeCl_3$ is one of the most commonly used chemical coagulants in WWTPs for various purposes. In addition, many researchers have stated that vivianite ($Fe_3(PO_4)_2 \bullet 8H_2O$) is the most easily formed phosphate precipitation during AD and the fertilizer value of vivianite was already reported from more than 20 years ago (Eynard *et al.*, 1992; Wilfert *et al.*, 2018). Due to the above reasons, the addition of trivalent cation Fe^{3+} in the AD system with biogas recirculation was tested in this Chapter.

(4) Chapter 4 Semi-continuous operation of the novel anaerobic digestion system with biogas recirculation

The long-term effect of biogas recirculation on performance of the AD system was investigated through semi-continuous AD of excess sludge. The effects of $MgCl_2$ or $FeCl_3$ addition on AD performance, nutrients conservation and sludge dewaterability were also compared. In addition, the mechanism of biogas recirculation was further clarified through the analysis of archaeal and microbial community.

(5) Chapter 5 Mechanisms and economic analysis

The possible mechanisms involved in the novel AD system for biogas upgrading, P conservation and sludge conditioning were summarized. Then, the economic analysis in industrial scale and national scale was conducted.

(6) Chapter 6 Conclusion and future research

The results of this research were summarized, and the future research was pointed out.

Table 1-1 Summary of previous works on utilization of biogas recirculation in anaerobic digestion process.

Reactors	Biogas recirculation rate	Substrates	Organic Loading Rate	Achievement	References
Stirred tank reactor/1.5L	24 L/10 min with 6 h interval	Swine slurry	3.9 g-VS/(L·d)	Increased biogas production	Lee *et al.*, 1995
Anaerobic hybrid reactor/5.5L	1.44 L/d (day 7) to 14.4 L/d (day 15)	Starch wastewater	5.5 g-COD/(L·d)	Enhanced biomass accumulation and methane content	Suvajittanont and Chaiprasert, 2003
Anaerobic digestor/3.73L	1 L/min	Manure slurry	3.24 g-VS/(L·d) (15% TS)	Increased methane production	Karim et al., 2005a
Anaerobic hybrid reactor/14.5L	38.88 L/d (27 ml/min)	Dairy manure	6.89 g-COD/(L·d) (9.87 % VS)	Increased biogas and methane production	Demirer and Chen, 2005
Semi-continuous reactor/15L	2 L/h, 120min before AD	Cow manure	43 g-COD/(L·d)	Increased biogas and methane production	Sánchez-Hernández et al., 2013
Continuous stirrer tank reactor (CSTR)/2.7L	24.14 L/d	Petrochemical wastewater with activated manure	13.3 g-COD/(L·d)	Increased biogas and methane production	Siddique et al., 2015
Gas-lift AnDMBR	35 m/h	High strength wastewater	2 g-COD/(L·d)	Alleviative membrane fouling problem	Ersahin et al., 2016
Anaerobic membrane reactor	3 L/min, 30s; 5 L/min, 30s	Synthetic wastewater	2 g-COD/(L·d)		Zhang et al., 2017

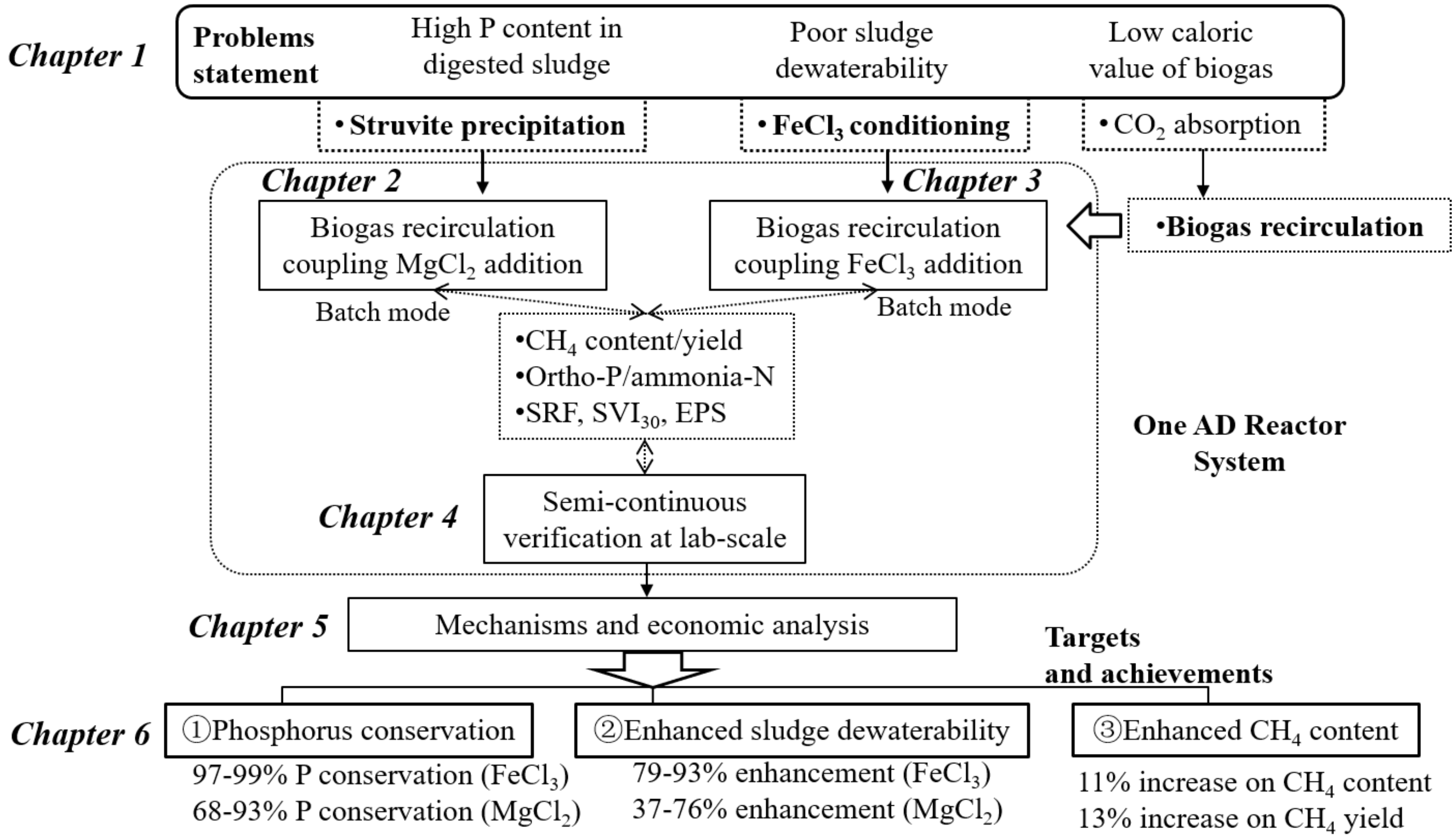

Fig. 1-1 The framework of this thesis. SRF, specific resistance to filtration; SVI_{30}, sludge volume index within 30 min; EPS, extracellular polymeric substances; AD, anaerobic digestion.

Chapter 2 Effect of biogas recirculation coupling with $MgCl_2$ addition on anaerobic digestion of sewage sludge

2.1 Background

Large quantity of ortho-P and ammonia N are released into the liquid phase of the digested sludge during AD (i.e. fermentation liquor), increasing the burden on WWTPs when the liquor is returned to the aeration tank(s) after digested sludge being dewatered. Nevertheless, they are also the main essential nutrients for plant growth. As it is known, forced struvite ($MgNH_4PO_4 \cdot 6H_2O$) precipitation can be applied in industries as a side stream for simultaneous P and N recovery from the liquid digestate by adding Mg salt, and the recovered struvite is regarded as a slow-releasing fertilizer (Yetilmezsoy *et al.*, 2017). Besides, Mg addition is also helpful for the enhancement of sludge dewaterability due to the effectiveness of divalent cations in binding with negatively charged sludge particles or biopolymers (Zhai *et al.*, 2012). To further reduce the operation cost for sustainable sewage sludge treatment, Mg salt was directly added into the AD reactor in this study to recover nutrients and enhance sludge dewaterability simultaneously. Most recently, Tarragó *et al.* (2018) reported that suspended solids favored the aggregation of struvite crystals, who further claimed that the presence of solids was beneficial for meeting crop demands and restoring soil fertility. Thus, adding Mg salt directly into sludge might be a better choice for nutrients recovery targeting land application. Previous studies have mainly focused on P recovery from digestate liquor via struvite precipitation or mitigation of ammonia-N inhibition during AD (Table 2-1). Little information is available on the changes of sludge properties and resources conservation when coupling biogas recirculation with Mg salt addition, which is crucial for the sustainable management of sewage sludge in WWTPs.

2.2 Materials and methods

2.2.1 Sludge samples

Concentrated primary sludge, sampled from the Shimodate WWTP in Chikusei, Ibarak, Japan was used as the feedstock for AD in this study. Sludge samples were stored in 20 L polypropylene tanks at 4°C in laboratory. Anaerobically digested sludge was sampled from the anaerobic digestion tank in the same WWTP, which is operated under mesophilic condition and fed with the mixture of the concentrated primary sludge and the excess sludge from the secondary clarifier. Before being used as the inoculum in the AD system in this study, the sampled digested sludge was incubated by adding the concentrated PS at a ratio of 4:1 (v/v) at 37±2°C in the laboratory for one week. The characteristics of the sampled sludges are shown in Table 2-2.

2.2.2 Establishment of AD system

Four identical reactors as shown in Fig. 2-1 were fabricated, each with a working volume of 0.7 L (D×H=8.5 cm × 12.34 cm) and being connected to a 1 L graduated cylinder. The produced biogas was collected in an inverted cylinder, and the biogas volume could be read directly from the scale on the cylinder. Saturated $NaHCO_3$ solution was prepared and used in the biogas collection part to avoid the dissolution of CO_2 in water. After biogas production became stable, biogas recirculation was started at a constant flowrate controlled by a gas pump and a gas flowmeter. Then the biogas was further distributed into the AD reactor via two ceramic gas spargers with the same pore size (selected from the same batch products of the manufacturer). Before AD experiments, the reactors were tested for their gas tightness for 5 days' continuous operation of air recirculation after being filled with tap water (instead of sewage sludge). Another four reactors, with the same size but without installation of gas spargers, gas flowmeter and air pump, were also set up and used for AD without biogas recirculation.

The eight reactors were operated in parallel using batch AD experiments: 1) two control reactors (Cont.) were operated under no biogas recirculation and no $MgCl_2$ addition; 2) two reactors (+Mg) were run with $MgCl_2$ addition while no biogas recirculation; 3) two reactors (Cont.+biogasR) were under biogas recirculation while no $MgCl_2$ addition; and 4) two reactors (+Mg+biogasR) were examined under biogas recirculation and $MgCl_2$ addition, respectively.

The feedstock and the inoculum were mixed at a ratio of 2:1 (volatile solids (VS) basis). The initial total solids (TS) and VS concentrations in each reactor were about 16.04 g/L and 10.8 g/L, respectively. For the reactors with $MgCl_2$ addition, $MgCl_2$ was directly added into the mixed sludge at 1428 mg/L (15 mmol/L), according to the molar concentration of PO_4^{3-} detected in the preliminary experiments. The initial pH was adjusted to 7.00±0.02 using 4 mol/L NaOH solution. After being loaded with 0.7 L of sludge mixture, each AD reactor was purged with N_2

gas for 5 min to create anaerobic condition. All the AD reactors were placed in a temperature-controlled thermostat (37±2°C) and the AD experiments lasted for 30 days. The biogas recirculation was started from day 4 using the biogas produced by itself at a flowrate of 200 mL/min in an intermittent mode of 60 min-on/60 min-off according to the preliminary experiments. On day 17, one bottle of each condition was opened and the sludge samples were taken for parameters' determination.

2.2.3 Procedures and analytical methods for general samples

TS and VS contents, TN and TP concentrations of the sludge samples were measured according to the standard methods (APHA, 2012). Biogas composition including CH_4 and CO_2 was analyzed by a gas chromatography (GC-8A, Shimadzu) equipped with a thermal conductivity detector (TCD). Dissolved biogas was detected as described elsewhere (Souza *et al.*, 2011).

To determine other parameters, 20 mL of each sample was centrifuged at 9900 rpm for 15 min. After the resultant supernatant being filtered through 0.22 μm filter membrane, the solid residue was dried at 60°C for 72 h and grounded into powder for insoluble organic carbon analysis using a total organic carbon (TOC) analyzer (TOC-V_{CSN} with SSM-5000A, Shimadzu). Soluble organic carbon and soluble inorganic carbon in the filtrate were determined by the same TOC analyzer equipped with an ASI-V autosampler. Soluble ammonia N was detected using the indophenol-blue method (Ivančič and Degobbis, 1984). Soluble ortho-P was measured according to the standard methods (APHA, 2012). Insoluble N, including organic N and precipitated inorganic N in the solid phase, was calculated by subtracting soluble TN from the general TN. Soluble organic N was calculated by subtracting soluble ammonia N from the soluble TN, as very little nitrate and nitrite were detected in the filtrates.

As for P fractionation, the Standards, Measurements and Testing (SMT) harmonized procedure developed by the European Commission was used for sequential extraction of P from the solid and liquid phases (García-Albacete *et al.*, 2012). Through this method, P can be categorized as 5 fractions, including total P (TP), organic P (OP), inorganic P (IP), non-apatite inorganic P (NAIP, associated with Al, Fe, or Mn oxides or hydroxides) and apatite P (AP, Ca-bound). NAIP and OP are deemed as bioavailable P since they can be potentially released, decomposed and utilized by plants and microorganisms.

The sample for metals content analysis was obtained by digesting the sludge with an equivalent volume of *aqua regia* solution at 150°C. After complete digestion, the volume was adjusted to its original value with deionized water. Soluble metal ions were obtained by digesting the filtrate using the mixture of HNO_3 and H_2O_2 (1:1, v/v) at 98°C. Then, the liquor containing metal ions was filtered through 0.22 μm filter membrane and quantified by inductive coupled plasma (ICP) emission spectroscopy (ICPS-8100, Shimadzu).

The sludge samples used for surface morphology observation were taken from the Cont. and +Mg+biogasR reactors on day 30 and freeze-dried, and then characterized by scanning electron microscopy (SEM, JEOL JSM6330F, Japan). The existence of struvite crystals in the sludge samples were identified by X-ray diffraction (XRD, Bruker AXS D8 ADVANCE/TSM, USA).

2.2.4 Determination of sludge dewaterability and settleability

Specific resistance to filtration (SRF) and diluted sludge volume index (SVI_{30}) were used to evaluate sludge dewaterability and settleability, respectively. SRF was determined using the vacuum filtration method as described by Wang *et al.* (2017b). In this study, a Buchner funnel placed with a filter paper (Whatman No. 1) was used for vacuum filtration at a constant pressure of 0.07 MPa. In the determination of diluted SVI_{30}, the sludge sample was diluted by 5 times to adjust the total suspended solids (TSS) in the range of 2000 - 4000 mg/L (Liao *et al.*, 2001). The diluted sludge sample (after being mixed) was placed in a 100 mL graduated cylinder for 30 min settlement. Then the volume of settled sludge and the TSS of the mixed liquor were recorded and used for the calculation of diluted SVI_{30} according to the standard methods (APHA, 2012).

2.2.5 Extraction and determination of extracellular polymeric substances (EPS)

EPS in sludge were extracted using heat extraction method (Zhen *et al.*, 2012). In brief, the sludge sample was firstly centrifuged at 5000 rpm for 10 min, and the supernatant was collected to determine soluble EPS (S-EPS). After being washed with deionized water for three times, the sludge residue was re-suspended in 0.05% NaCl solution to its original volume. After being well shaken, it was heated at 70°C for 1 min, and then centrifuged at 5000 rpm for 10 min. The resultant supernatant was used for analysis of loosely bound EPS (LB-EPS). Later, the sludge residue was again re-suspended in 0.05% NaCl solution after being washed with deionized water for three times. After the mixture being heated at 60°C for 30 min and centrifuged at 5000 rpm for 15 min, the supernatant was collected as tightly bound EPS (TB-

EPS). After these extraction procedures, all the liquid samples were filtered through 0.45 μm filter membrane. Proteins (PN) and polysaccharides (PS) contents in the EPS extracts were then quantified by using the phenol-sulfuric acid method and Lowry-Folin method with glucose and bovine serum albumin (BSA) as the standard, respectively (Lowry *et al.*, 1951; Dubois *et al.*, 1956).

2.2.6 Statistical analysis

All the experiments in this study were conducted in triplicate and the data were presented as their average values. One-way analysis of variance (ANOVA) was applied to analyze the statistical difference of the experimental data using Microsoft Office Excel 2016. Pearson's correlation analysis was performed by using OriginPro, Version 2018C (OriginLab Corporation, Northampton, MA, USA) to clarify the correlation between EPS and SRF. Significant difference was assumed at $p < 0.05$.

2.3 Results and discussion

2.3.1 Effect of biogas recirculation on biogas upgrading

The CH_4 contents in the reactors with biogas recirculation (+biogasR) significantly increased during AD (Fig. 2-2a). As shown, the CH_4 contents in all the reactors were similar before starting the biogas recirculation (day 4), which increased from 63% to 72%. After day 4, the CH_4 contents in the reactors (+biogasR) gradually increased and reached the highest (85.4-86.0%,) on day 18, and then some decrease in CH_4 content was detected when the reactors were operated for a longer time. As a contrast, the CH_4 contents in the reactors without biogas recirculation kept stable, around 75% till day 30. Only addition of $MgCl_2$ showed no significant effect on CH_4 content compared with the reactor Cont.

Fig. 2-2b illustrates the cumulative CH_4 production from the reactors during the 30 days' AD. Almost the same variation trend was observed on cumulative CH_4 production from all the reactors, indicating that the AD performance was not significantly affected by biogas recirculation and $MgCl_2$ addition. Similar results have been reported by Karim *et al.* (2005a), who claimed that biogas recirculation did not affect the methane production during AD when manure slurry was fed at low TS (5%). The cumulative CH_4 productions on day 17 amounted to 88%, 90%, 94% and 96% of the total CH_4 yield on day 30 in the reactors Cont.,

Cont.+biogasR, +Mg, and +Mg+biogasR, respectively. Generally, the effective biogas production duration is determined as the duration for achieving 80% of the total biogas (Huang *et al.*, 2017). Thus, the effective biogas production duration should be shorter than 17 days for the primary sludge used in this study, which would be further shortened when biogas recirculation and/or $MgCl_2$ addition being applied. A prolonged operation duration was adopted in this study because another target was to reveal the effect of biogas recirculation on the sludge properties.

Up to now, very few reports could be found on the obvious increase in CH_4 content through biogas recirculation during the AD process of sewage sludge. Most recently, Latha *et al.* (2019) reported an in-situ methane enrichment (88%) during AD with an intermittent biogas recirculation intensity of 2 L/min, which also significantly enhanced the biogas yield from co-digestion of food waste and sewage sludge at high solids content. Previous works mainly focused on the effect of biogas recirculation in a semi-continuous/continuous AD for wastewater treatment. An increase of CH_4 content from 56% to 59% was obtained by Suvajittanont and Chaiprasert (2003) during starch wastewater treatment using biogas recirculation at 14.4 L/d. The increased CH_4 content obtained in this study during the AD of sewage sludge was comparable to others. Restated, the continuous mode of AD with biogas recirculation for sludge treatment needs further investigation to confirm the feasibility of this system, since the dissolved CO_2 in sludge liquor might be again stripped out if a longer operation of biogas recirculation being applied (30 days in this study).

In order to further elucidate the C conversion during AD, the insoluble organic C, soluble organic C, soluble inorganic C, C converted to biogas (CH_4-C, CO_2-C) and dissolved biogas C were measured and calculated. These C fractions and the pH in each reactor during AD are illustrated in Fig. 2-3. The reduction of insoluble organic C did not show much difference among all the reactors during the experimental period. On day 17, the total organic C (TOC, soluble + insoluble) reductions were 20% (Cont.), 20% (Cont.+biogasR), 23% (+Mg) and 23% (+Mg+biogasR), in which 15-17% of TOC were detected to convert to CH_4. On day 30, the TOC reductions increased to 33%, 33%, 32% and 30% in the four reactors, respectively, and the conversion ratios of TOC to CH_4 further increased to 18-19%. This phenomenon again indicated that the effective methane production duration could be shorter than 17 days, and a longer operation duration could not achieve much higher conversion of TOC in the sludge to CH_4 under the tested conditions. The C as CO_2 was detected slightly lower in the reactors with biogas recirculation, decreasing by around 2% and 3% on day 17 and day 30, respectively. On the other hand, the proportions of soluble inorganic C and dissolved biogas C were 12% and

11% in the reactors Cont.+biogasR and +Mg+biogasR on day 17, which increased to 14% and 13% on day 30, respectively. While little change in these two parts of C was detected in the reactors without biogas recirculation. The increased amount was comparable to the reduced amount of CO_2, implying that the reduced CO_2 in the biogas might be dissolved in the fermentation liquor or adsorbed onto the sludge particles during biogas recirculation. A higher pH value in the reactors with biogas recirculation may also be partially contributed by the increase of carbonates in the fermentation liquor. Whereas the decrease in CH_4 content after 17 days' operation (Fig. 2-2) might be brought about by the escaped CO_2 which was initially dissolved or as inorganic C in the fermentation liquor, and then stripped out after reaching its saturation state in the liquid phase due to the long-term biogas recirculation.

2.3.2 Nutrients conservation

(1) Phosphorus

In this study, $MgCl_2$ was added directly into the AD reactors with no additional phosphate addition to recover P and N simultaneously. The P fractions are presented in Fig. 2-4 as OP, NAIP and AP in the solid phase and soluble P in the liquid phase. The sum of NAIP+OP in the solid and soluble P were defined as the total bioavailable P, and the amount of NAIP+OP in the solid was termed as solid bioavailable P. The results indicated that the total bioavailable P (87% of TP) in the sewage sludge remained almost no change before and after AD no matter whether biogas recirculation or $MgCl_2$ addition was applied or not. The OP fraction obviously decreased on day 17 and day 30 by about 46% and 50%, respectively, probably due to the hydrolysis of OP during AD. However, the soluble P was not detected to increase correspondingly, as most of the hydrolyzed OP might be transformed into NAIP in the solid phase. Biogas recirculation was found to have slightly positive effect on the solid bioavailable P. The NAIP in the solid phase was reduced in the reactor +Mg on day 30 when compared with that on day 17, which was slightly increased in the reactor +Mg+biogasR. In addition, a slight increase of NAIP in the solid phase was also observed in the reactor Cont.+biogasR in comparison to that in the reactor Cont. on both day 17 and day 30. This observation indicates that AD process together with biogas recirculation and $MgCl_2$ addition could greatly promote P conservation in the solid phase of digested sludge. Moreover, the enhanced P conservation in the solid phase through struvite precipitation might also bring about some scaling problems in the AD tanks in practice (Ohlinger *et al.*, 1998). Therefore, the configuration and operation conditions of the sludge treatment facilities should be further optimized to avoid scaling problems in the AD tanks and the pipelines associated with (Koga, 2019).

In short, by coupling biogas recirculation with $MgCl_2$ addition during AD process, 93% of TP could be preserved in the solid phase, in which 86% was bioavailable. Moreover, 87% and 86% of soluble P could be reduced in the reactor +Mg+biogasR on day 17 and day 30, respectively, when compared with the reactor Cont. The removal of soluble P in this study is comparable with those obtained from the fermentation liquor through struvite precipitation. For example, Münch and Barr (2001) achieved 94% of ortho-P removal by adding $Mg(OH)_2$ at a Mg:P molar ratio of 1.3:1 to the dewatering liquid from digested sludge. Wacławek *et al.* (2016) reported a 81% of ortho-P removal from the liquid phase of digested sludge by adding MgO and Na_3PO_4 at a Mg:P:N molar ratio of 1:1:1.

(2) Nitrogen

The changes of N species on day 0, day 17, and day 30 during AD are shown in Fig. 2-5. On day 17, about 19% decrease of ammonia N was detected in both reactors +Mg compared with the reactors Cont. and Cont.+biogasR, while the reduced amount of ammonia N in the reactor +Mg+biogasR (102.6 mg-N/L) was slightly higher than that in the reactor +Mg (97.4 mg-N/L). On day 30, the reduced amount of ammonia N was only 82.7 mg-N/L in the reactor +Mg, while it increased to 157.9 mg-N/L in the reactor +Mg+biogasR. A higher pH was always detected in the reactors +biogasR (Fig. 2-3), which may favor the struvite formation in these reactors. As a result, 69% and 72% of TN could be preserved in the solid phase of digestate after 17- and 30-days' AD with biogas recirculation and $MgCl_2$ addition, respectively. About 19% and 29% of ammonia N could be reduced in the reactor +Mg+biogasR, compared with those in the reactor Cont. on day 17 and day 30, respectively. This observation is comparable to the previous findings by using struvite precipitation in AD process. As pointed by Othman *et al.* (2009), 32% reduction in ammonia N could be achieved by adding $MgCl_2$ and Na_3PO_4 at a molar ratio of Mg:P:N =1.3:1:1 under neutral pH conditions. Uludag-Demirer *et al.* (2008) claimed a 23% reduction in ammonia N by adding 1750 mg/L $MgCl_2•6H_2O$ and 3000 mg/L Na_2HPO_4 during AD at pH 7.15, with no influence on biogas production being observed.

2.3.3 Struvite formation

The formation of struvite crystals in the reactor +Mg+biogasR was confirmed by XRD and SEM (Figs. 2-6 and 2-7). The peak positions in the XRD pattern of the sludge samples from the reactor +Mg+biogasR matched well with the reported XRD pattern of struvite crystals

(Prywer *et al.*, 2012; Chen *et al.*, 2018). The struvite-like crystals could also be clearly noticed from the SEM image of the sludge sample from the reactor +Mg+biogasR (Fig. 2-6d). To estimate the quantity of the formed struvite during AD under the condition of biogas recirculation and $MgCl_2$ addition, the concentrations of soluble Mg and total Mg were measured on day 0, day 17 and day 30 (Fig. 2-8a). The concentration change of soluble Mg was consistent with the variations of PO_4^{3-} and NH_4^+. This observation again proved that biogas recirculation could accelerate struvite precipitation. The changes of the molar concentrations of increased insoluble Mg^{2+}, decreased PO_4^{3-} and NH_4^+ were calculated based on the corresponding data from the reactor Cont. on day 0, day 17 and day 30 (Table 2-3). Since the Mg concentration was also noticed to decrease in the reactor Cont., the struvite concentration estimated in this study only represents the part that was increased by biogas recirculation and/or $MgCl_2$ addition during AD. In addition, the decreased PO_4^{3-} might be contributed by the precipitation with Mg^{2+} alone or the co-precipitation with other co-existing cations (e.g. Ca^{2+}), and Mg^{2+} might be precipitated with other anions (e.g. CO_3^{2-}). Thus, the decreased molar concentration of NH_4^+ in the liquid was assumed as the increased molar concentration of struvite in this study (as little NH_3 was detected in the biogas). On day 30, the increased insoluble Mg, decreased PO_4^{3-} and decreased NH_4^+ in the reactor +Mg+biogasR were 12.83, 11.43 and 11.42 mmol/L, respectively, indicating that most of the Mg added into the reactor might precipitate as struvite. On day 17, the decreased NH_4^+ seems to be lower than that on day 30; however, the value was still higher than that in the reactor without biogas recirculation (+Mg). It could be estimated that about 7.32 mmol/L (119.56 mg/g-TS) and 11.42 mmol/L (192.96 mg/g-TS) struvite formed in the reactor +Mg+biogasR after 17 and 30 days' AD, respectively, according to the amount of the decreased NH_4^+.

2.3.4 Changes in sludge properties

(1) Sludge dewaterability and settleability

The changes of SRF and diluted SVI_{30} during AD are shown in Fig. 2-9. The SRF was reduced by 18% after $MgCl_2$ being added in the sludge mixture on day 0, indicating its enhanced dewaterability, most probably due to the binding capacity of divalent cation (Mg^{2+}) with negatively charged sludge particles (Sobeck and Higgins, 2002). Liu *et al.* (2011) compared the effect of seawater and brine on sludge conditioning, indicating that Ca^{2+} and Mg^{2+} have similar effectiveness that is greater than monovalent cations like Na^+ on sludge dewaterability and flocculation. After 17 days' AD, the SRF was further decreased in the reactors +Mg and

+Mg+biogasR by 55% and 37% compared with the initial value without $MgCl_2$ addition. This observation to a greater extent agrees with Wang *et al.* (2018a) who found that the crystallized struvite in anaerobically digested sludge could bind with EPS fractions, resulting in enhanced sludge dewaterability. However, biogas recirculation seemed to worsen the sludge dewaterability to some extent in both reactors Cont. and +Mg. In contrast, little change in SRF was observed in the reactors without biogas recirculation on day 17 and day 30. A shorter biogas recirculation (< 17 days) along with Mg addition can remarkably enhance the dewaterability of the digested sludge. Compared to the 16% reduction in capillary suction time (CST) by alkaline pretreatment of AD sludge at pH 8 by Shao *et al.* (2012), and the 14% increase in dewatered solid content after free nitrous acid (FNA) pretreatment of AD sludge by Wei *et al.* (2018), this study achieved higher enhancement on sludge dewaterability by using this novel AD system.

On the contrary, an opposite effect of biogas recirculation was observed on sludge settleability within 30 min. The diluted SVI_{30} was found to decrease in all the sludges after AD, which was much lower in the reactors with biogas recirculation on day 30. However, after 30 min' SVI test, more small flocs were observed in the supernatant of sludge samples from the reactors with biogas recirculation on day 30. Jin *et al.* (2003) reported a positive correlation between floc size and SVI, indicating that a smaller floc size is beneficial for sludge settleability. While as pointed by Zhang *et al.* (2015), a large amount of fine colloidal particles is detrimental to sludge dewatering process. The small particles might be separated from sludge aggregates due to the shear force brought about by biogas recirculation, thus worsening the sludge dewaterability to some extent.

(2) Extracellular polymeric substances (EPS)

Many previous works suggest the important role of EPS on sludge dewaterability, flocculation, and sedimentation (Liu and Fang, 2003). EPS are a mixture of high-molecular-weight polymers excreted by microorganisms, and its major components are proteins (PN) and polysaccharides (PS). Some EPS are negatively charged and have strong affinity for water. The results from this study show that the PS and PN contents in TB-EPS decreased, while those in LB-EPS increased after AD process under all the tested conditions (Fig. 2-10). Higher PS and PN contents were detected in S-EPS in the reactors with biogas recirculation compared to no biogas recirculation counterparts, while those in LB-EPS and TB-EPS were relatively lower in the reactors with biogas recirculation. An intensive mixing during AD may result in decreased excretion of extracellular polymeric substances (EPS), and breakup of sludge flocs of bacteria and archaea with negative effect on biogas production (Karim *et al.*, 2005b). In addition, the increased S-EPS might not contribute to biogas production from AD of sludge. In this work, according to Pearson's correlation analysis, a strongly positive correlation was found between both PS and PN in S-EPS and SRF (R=0.841, p=0.009 for PS; R=0.964, $p=1.13\times10^{-4}$ for PN), while a strongly negative correlation was found between PN in TB-EPS and SRF (R=-0.957, $p=1.92\times10^{-4}$). Thus, during a long-term biogas recirculation, the PN in TB-EPS might be transferred to the liquid phase, probably due to the resultant high shear force from biogas recirculation. This might destroy the structure of sludge flocs, and further released small colloidal particles and then deteriorated sludge dewaterability to some extent. Moreover, an excessive S-EPS content is detrimental to cell cohesion and stability of sludge flocs, leading to poor dewaterability (Zhang *et al.*, 2015). A higher PN content was also detected in the TB-EPS from the sludges in the reactors with Mg addition. As expected, the added Mg^{2+} might bridge with the biopolymers in TB-EPS, partially avoiding being stripped off from the sludge particles during biogas recirculation.

2.4 Summary

In this study, a novel AD reactor coupling biogas recirculation with $MgCl_2$ addition was established for realizing biogas upgrading, resources conservation and sludge conditioning simultaneously. During 17 days' AD with $MgCl_2$ addition and intermittent biogas recirculation, the following results could be obtained: 1) CH_4 content was enhanced to 86% in comparison to 75% in the reactors without biogas recirculation; 2) 87% ortho-P and 19% ammonia N were conserved in the solid phase of digested sludge; 3) the sludge dewaterability was enhanced by 37%. In addition, a long-term biogas recirculation (30 days in this study) was found to worsen sludge dewaterability and decrease CH_4 content to some extent. Further optimization of operation conditions is necessary to achieve higher CH_4 content and enhanced dewaterability for practical application.

Table 2-1 Summary of previous works on struvite precipitation for P and N recovery/removal from anaerobically digested liquor.

Substrates	Mg source	PO_4^{3-} source	Mg:P (:N)	Removal efficiency	Ortho-P (mg/L)	Ammonia-N (mg/L)	Temp. (°C)	pH	References
Digested manure	MgO	-	1:1	95% (P)	1591.2	2360	35	8.5	Beal *et al.* (1999)
Digested sludge dewatering liquid	$Mg(OH)_2$	-	1.3:1	94% (P)	61	790	20	8.5	Münch and Barr (2001)
Anaerobic digested sludge	MgO	Na_2HPO_4	1:1:1	81% (P) 92.6% (N)	358	1449.6	20	9.5	Wacławek *et al.* (2016)
Food waste	$MgCl_2$	-		73% (P) 67% (N)	-	-	-	Neutral	Lee *et al.* (2004)
Waste activated sludge	$MgCl_2 \bullet 6H_2O$	Na_3PO_4	1.3:1:1	32% (N)	411.5	427.5	35	Neutral	Othman *et al.* (2009)
Dairy manure	$MgCl_2 \bullet 6H_2O$	Na_2HPO_4	1:2.45:3	23% (N)	-	359	35	Neutral	Uludag-Demirer *et al.* (2008)
Pig manure	Stabilization agent (MgO, P_2O_5, etc.)	K_2HPO_4	1.7:1:1	77% (N)	36	1785	37	7.5	Romero-Güiza *et al.* (2015)

Table 2-2 The characteristics of concentrated primary sludge and anaerobically digested sludge used in this chapter.

Parameters	Concentrated primary sludge	Anaerobically digested sludge
Total solids (TS, %)	1.82	1.26
Volatile solids (VS, %)	1.38	0.94
Total phosphorus (TP, mg-P/L)	990.01	728.98
Soluble ortho-P (mg-P/L)	561.14	263.74
Total nitrogen (TN, mg-N/L)	1285.59	989.74
Ammonia nitrogen (mg-N/L)	145.29	372.12
pH	5.98	7.01

Note: average values from triplicate determinations.

Table 2-3 Changes in molar concentrations of increased soluble Ca^{2+}, increased insoluble Mg^{2+}, decreased PO_4^{3-} and NH_4^+ on days 0, 17 and 30 after $MgCl_2$ addition during anaerobic digestion with (+Mg+biogasR) or without biogas recirculation (+Mg) (Unit: mmol/L except being specified).

Operation conditions	Operation duration (days)	Increased soluble Ca^{2+}	Increased insoluble Mg^{2+}	Decreased PO_4^{3-}	Decreased NH_4^+
+Mg	0	0.41	10.12	10.59	9.59
+Mg	17	0.39	12.39	12.19	6.95
	30	0.42	11.89	11.54	5.91
+Mg	17	0.21	12.62	12.16	7.32
+ biogasR	30	0.11	12.83	11.43	11.42

Note: The increased and decreased amount were calculated based on the corresponding data in the reactors Cont. and Cont.+biogasR on day 0, day 17 and day 30, respectively.

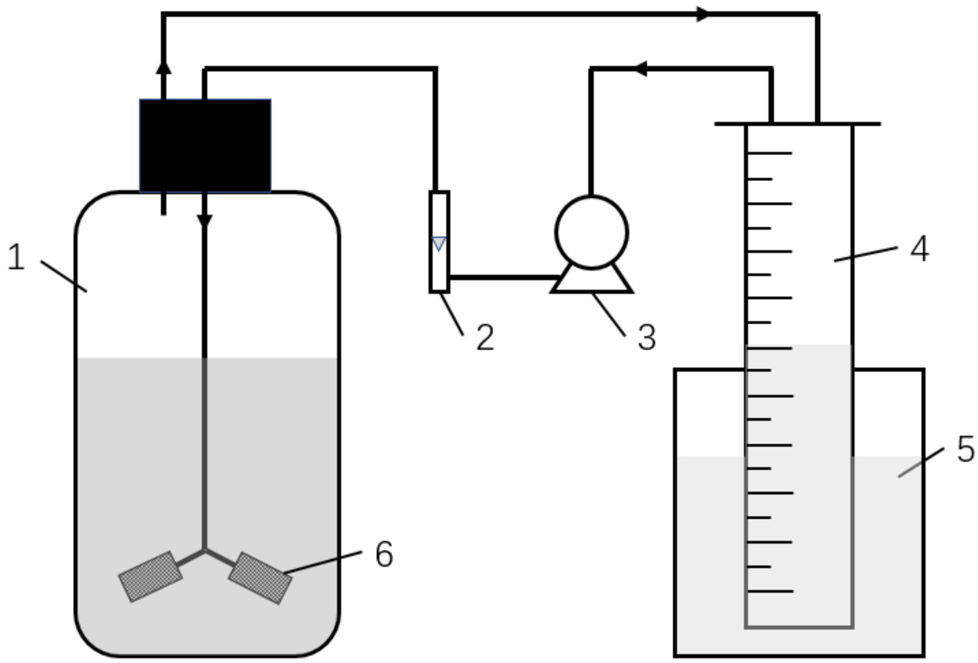

Fig. 2-1 Schematic of the batch anaerobic digestion system used in this study. 1. One-liter anaerobic digestion reactor; 2. Gas flowmeter; 3. Gas pump; 4. Graduated cylinder; 5. Saturated $NaHCO_3$ solution; 6. Gas sparger.

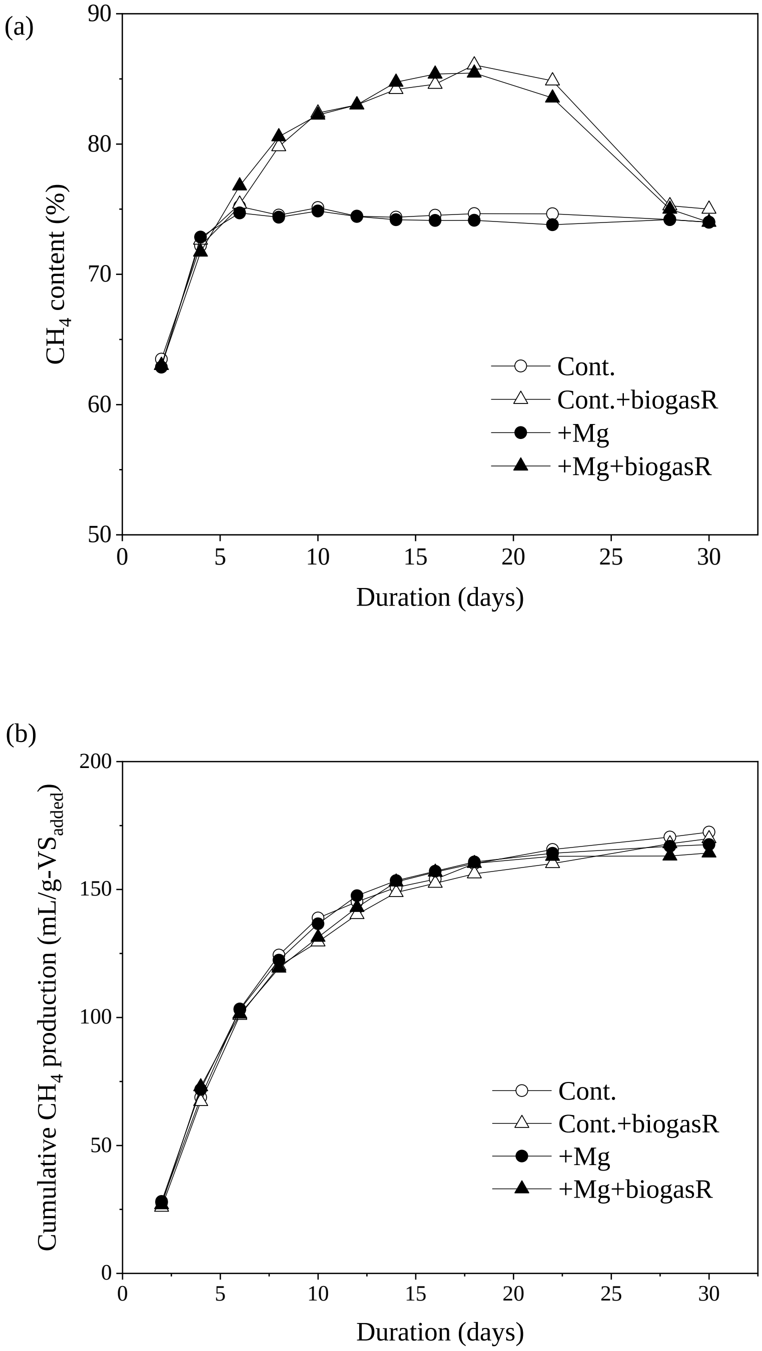

Fig. 2-2 Variations of CH_4 content (a) and cumulative CH_4 production (b) during 30 days' anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.

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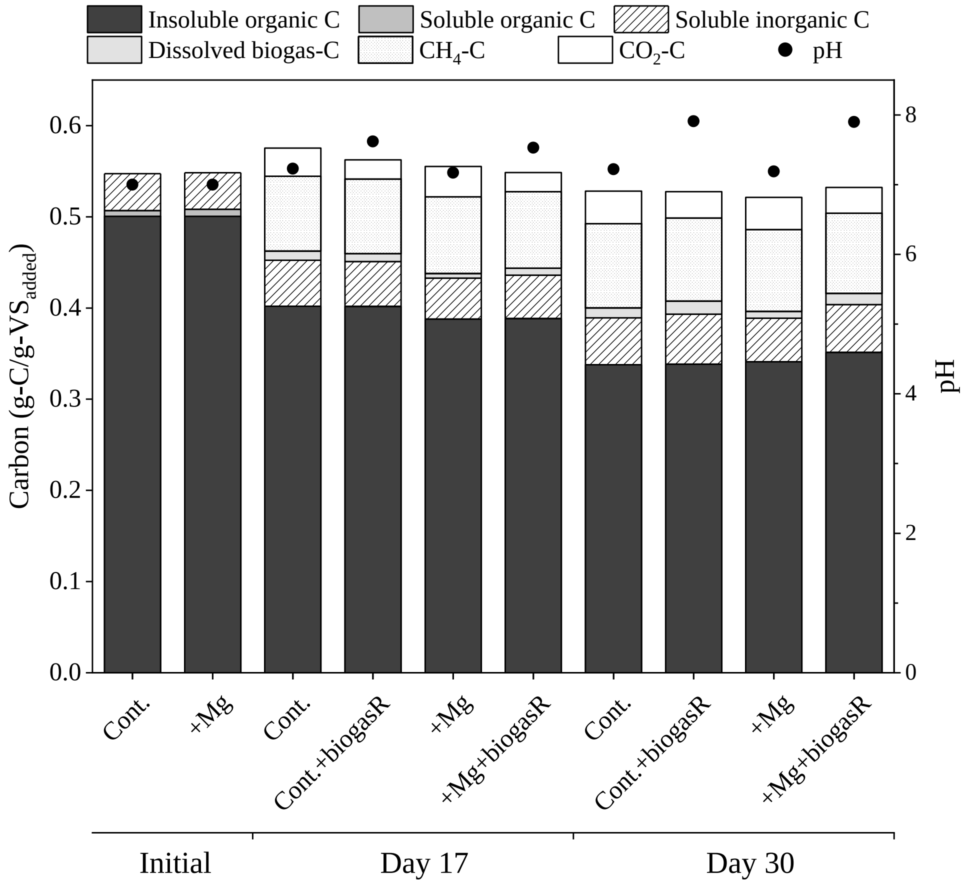


Fig. 2-3 Carbon (C) profiles and digestate pHs in the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.

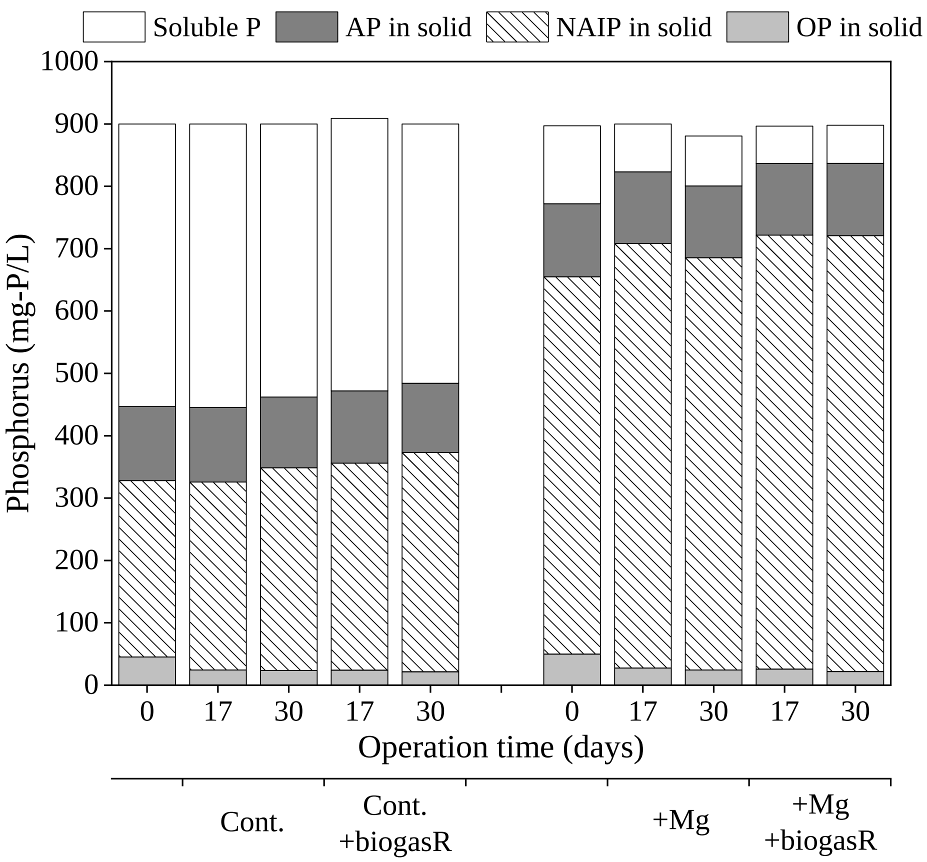


Fig. 2-4 Phosphorus (P) profiles in the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). The concentration of each fraction (AP, NAIP or OP in the solid phase) was calculated based on their contents in the solid and the solids concentration in the sludge sample. Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.

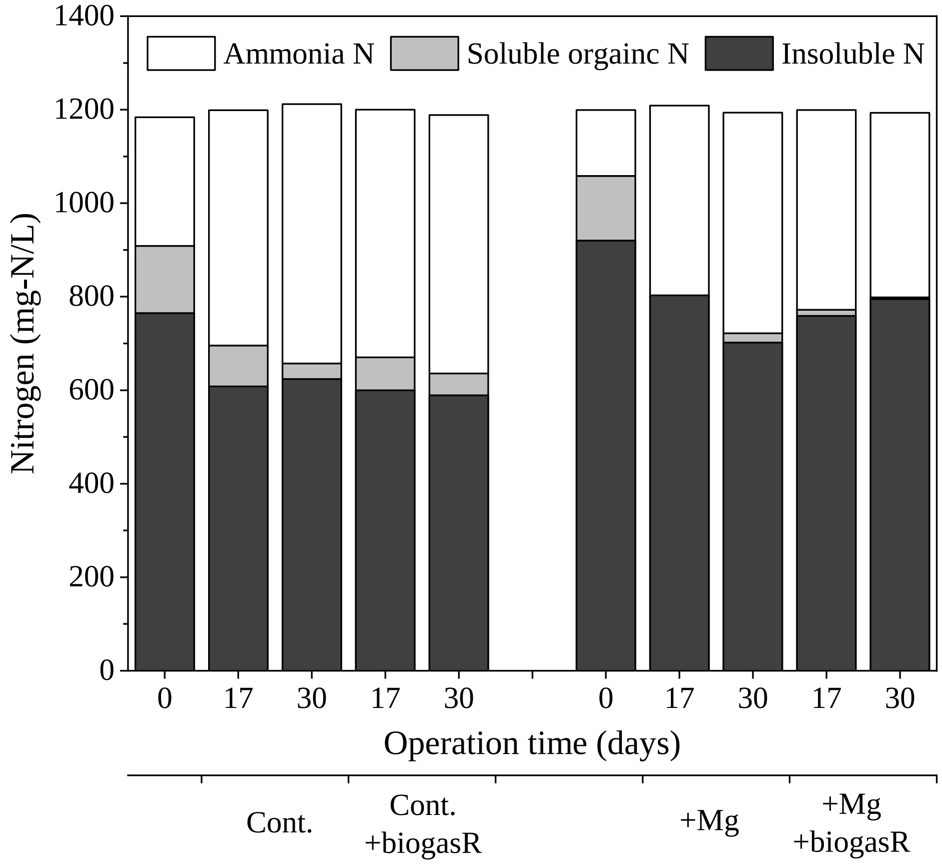


Fig. 2-5 Nitrogen (N) profiles in the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). The concentration of insoluble N was calculated based on its N content in the solid and the solids concentration in the sludge sample. Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.

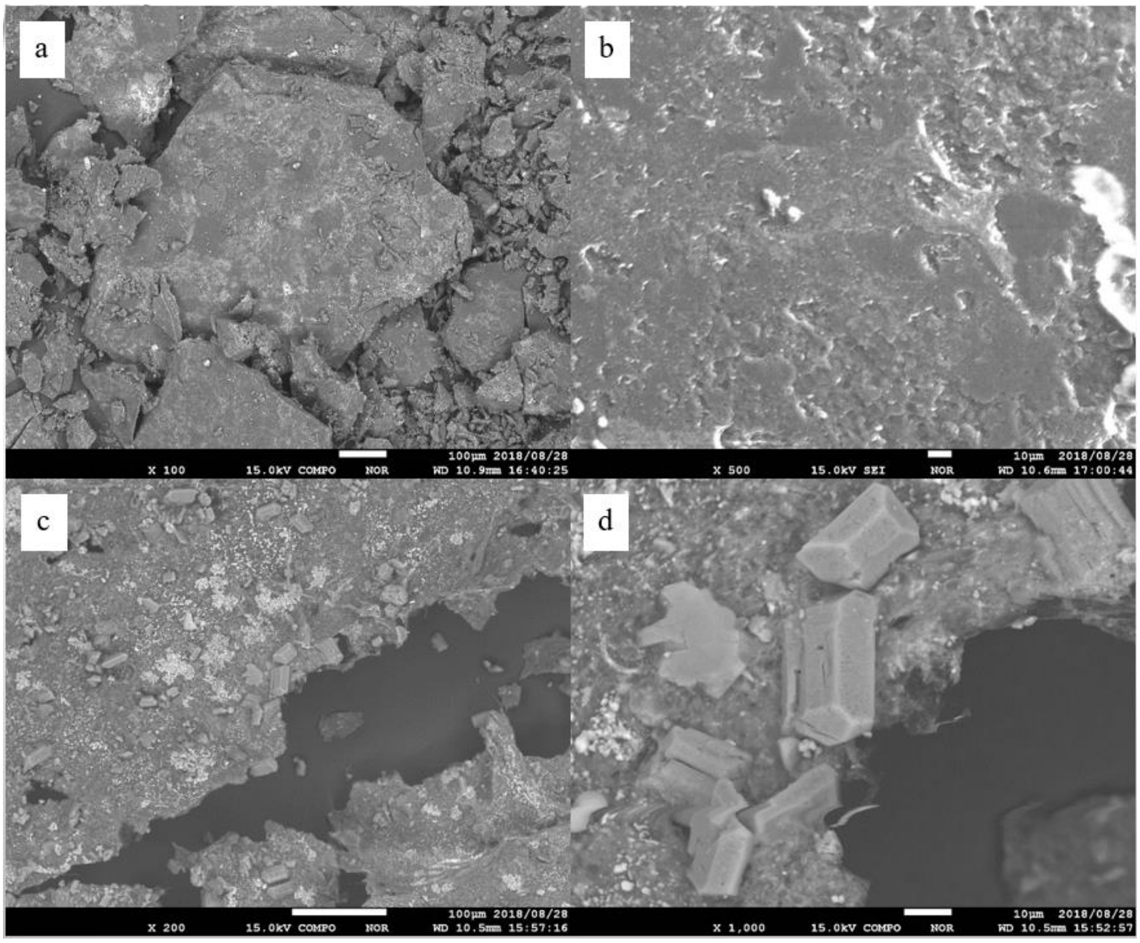


Fig. 2-6 Scanning electron microscope images of sludge samples from the reactors Cont. (a, b) and +Mg+biogasR (c, d) on day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.

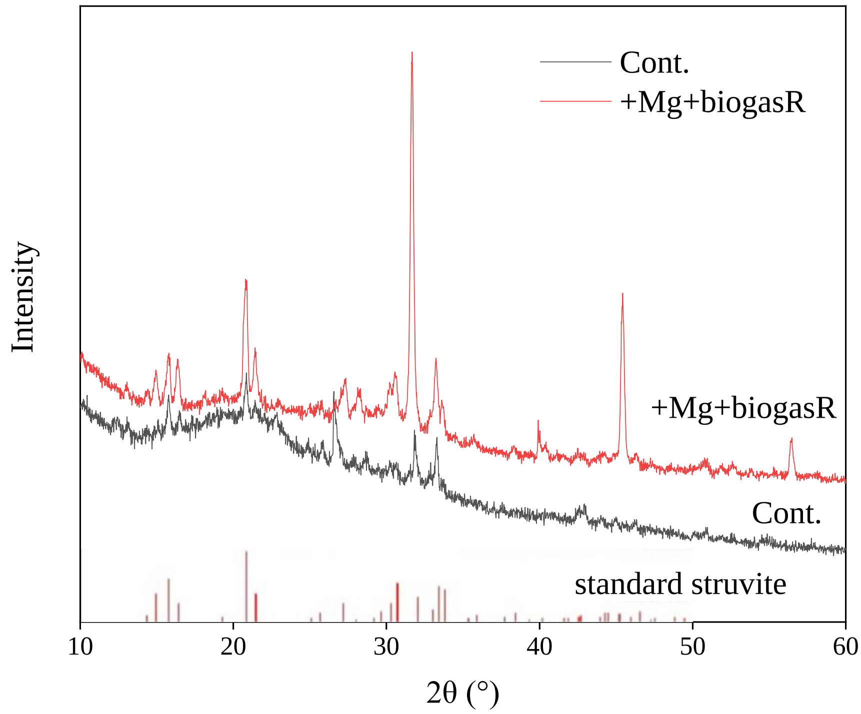


Fig. 2-7 X-Ray diffraction patterns of standard struvite crystal and the sludge samples from the reactors Cont. and +Mg+biogasR on day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition. The standard curve of struvite crystal was referred to Wang *et al.* (2019).

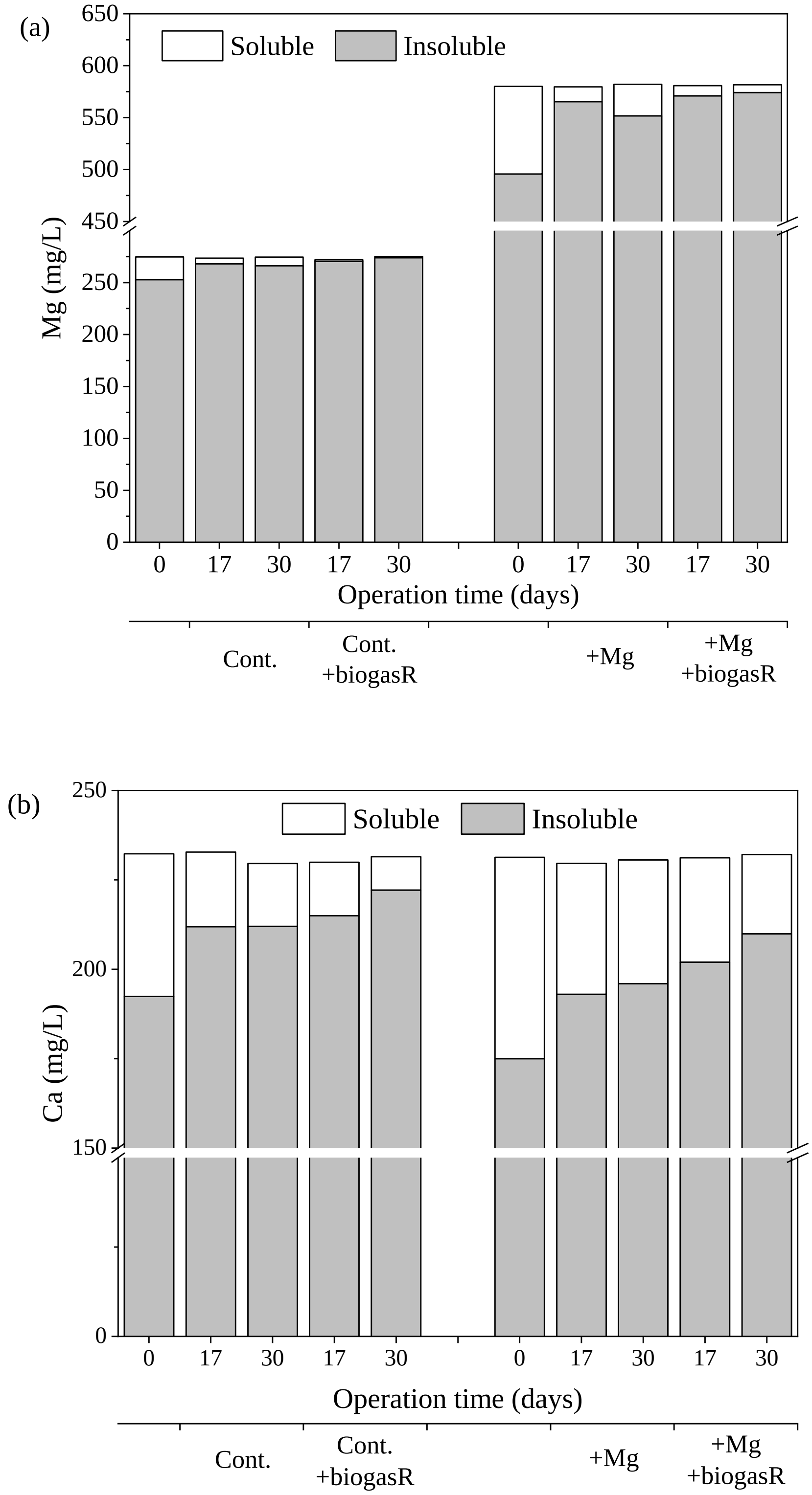


Fig. 2-8 Changes in Mg (a) and Ca (b) concentrations in the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg).

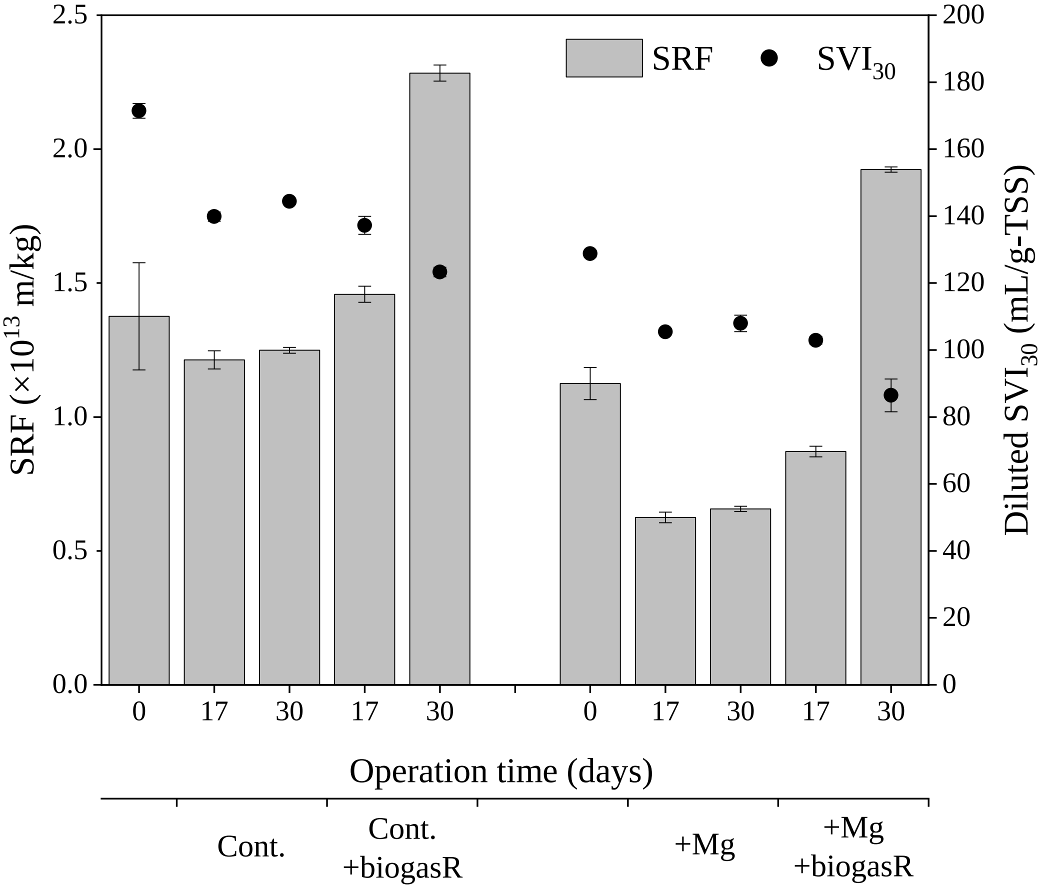
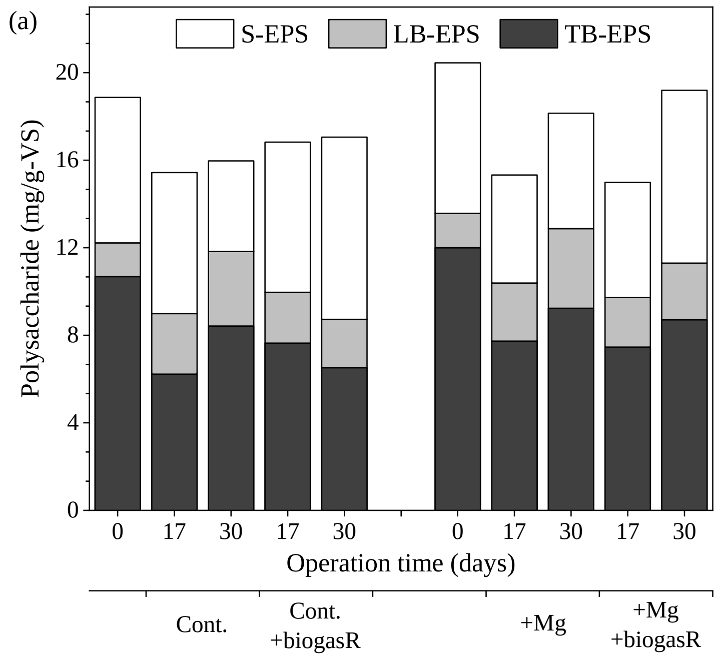


Fig. 2-9 Specific resistance to filtration (SRF) and diluted sludge volume index within 30 min (SVI_{30}) in the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg). Cont.-Control reactor with no biogas recirculation and no $MgCl_2$ addition.



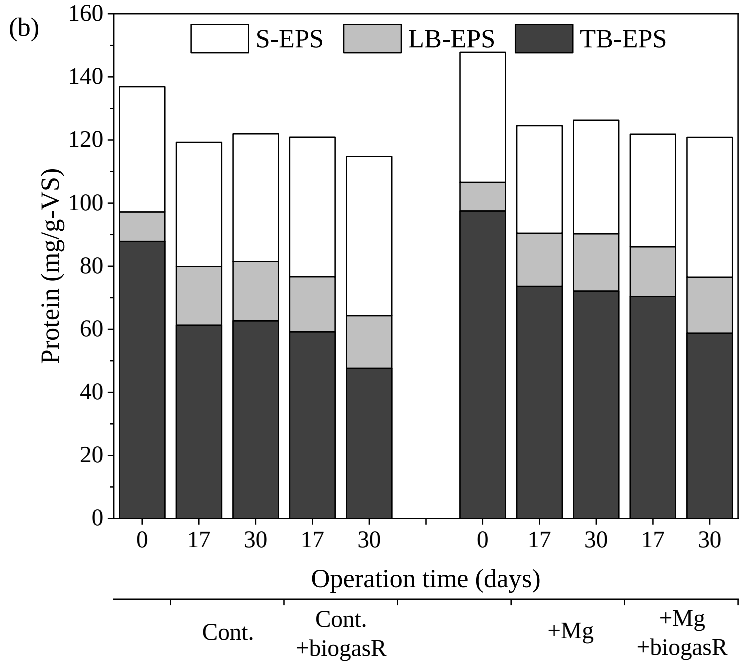


Fig. 2-10 Changes in extracellular polymeric substances (EPS) content including polysaccharides (a) and proteins (b) in the soluble EPS (S-EPS), loosely bound EPS (LB-EPS) and tightly bound EPS (TB-EPS) in the sludges from the reactors on day 0, day 17 and day 30 during anaerobic digestion with biogas recirculation (+biogasR) and $MgCl_2$ addition (+Mg).

Chapter 3 Effect of biogas recirculation coupling with $FeCl_3$ addition on anaerobic digestion of sewage sludge

3.1 Background

$FeCl_3$ is the most commonly used coagulants in WWTPs due to its low-cost and high efficiency. $FeCl_3$ could be used in any step during sewage or sludge treatment, achieving various purposes efficiently (Table 3-1). For instance, 1) the addition of $FeCl_3$ for pre-precipitation of municipal sewage at a Fe:P molar ratio of 0.8 obtained 64% of suspended solids removal, 50% of COD removal, 22% of total Kjeldahl nitrogen (TKN) removal and 43% of TP removal (Ghyoot and Verstraete, 1997); 2) the application of $FeCl_3$ at a Fe:P molar ratio of 1.5-1.9:1 to an activated sludge reactor achieved soluble P removal up to 98% with improved sludge sedimentation properties (De Gregorio *et al.*, 2010); 3) 1.25% w/w $FeCl_3$ addition in anaerobic digester showed positive effect on digestion performance with higher VS reduction and odor control with reduced hydrogen sulfide in the headspace gas of dewatered biosolids (Park and Novak, 2013); 4) the sludge conditioning with 10% $FeCl_3$ (dry basis) was tested in both laboratory and pilot tests, resulting in a reduction of 47.5% in the sludge volume (Lin *et al.*, 2014). The addition of Fe during wastewater treatment is often considered to limit phosphorus recovery, however, the wide use of Fe in WWTPs has also promoted the attention on phosphorus recovery in the form of Fe-P. Wilfert *et al.* (2015) first pointed out that the poor understanding of the Fe and P interaction in WWTPs limited the efficient phosphorus recovery from Fe-P rich wastes. They also found that vivianite ($Fe_3(PO_4)_2$) is the dominant inorganic solid phosphorus compound not only in anaerobic digesters but also in waste activated sludge due to its very low solubility (K_{sp}=10^{-36}) and slow oxidation kinetics (Wilfert *et al.*, 2016; Wilfert *et al.*, 2018). Vivianite has been occasionally used as a phosphorus fertilizer (Nelipa, 1961) and its effectiveness in reducing iron chlorosis in various plants has been proven for more than 20 years (Eynard *et al.*, 1992). Though vivianite is sparklingly soluble in normal soil pH conditions, the quick uptake of phosphorus by plants could favor its dissolution. In addition, the oxidation of Fe may cause net release of phosphorus bound in vivianite (Wilfert *et al.*, 2015). Further, the presence of organic matters could significantly influence the Fe and P interaction, possibly promoting the formation of amorphous iron-organic-phosphorus complexes, which show high ortho-P adsorption capacities and Fe-P bioavailability (Wilfert *et al.*, 2015). Thus, the effect of $FeCl_3$ addition on AD performance, P removal and sludge dewatering in anaerobic digestion system with biogas recirculation was investigated in this chapter.

3.2 Materials and methods

3.2.1 Sludge samples

The sludge samples' preparation was as the same as in Chapter 2 (2.2.1).

3.2.2 Establishment of AD system

The reactor design and preparation for batch AD test were the same as those mentioned in section 2.2.2. Eight reactors were prepared in this experiment for different concentrations of $FeCl_3$ addition with or without biogas recirculation. After well mixing the feedstock and inoculum at a ratio of 2:1 (VS basis), 300 (R1), 600 (R2) or 900 (R3) mg-Fe/L of $FeCl_3$ was added into the mixed sludge, respectively. The corresponding ferric ion contents in sludge were detected as 27.74, 49.32 and 68.63 mg-Fe/g-TS, which are within the reported optimal range for enhancing sludge dewaterability (Table 3-1). The molar ratios of Fe:P_{ortho} were 0.6, 1 and 1.5, respectively. The reactors without Fe addition were used as the control (R0, 7.43 mg-Fe/g-TS). After adjusting pH to 7.00±0.02, 0.7 L of the mixed sludge was loaded into each reactor, which was then purged with N_2 gas for 5 min to create anaerobic condition. The AD experiments were conducted at 37±2°C and lasted for 30 days.

3.2.3 First-order kinetic and modified Gompertz models for methane production

The first-order kinetic model (Eq. 3-1) and modified Gompertz model (Eq. 3-2) were applied to fit the cumulative methane production in this study.

$$\mathrm{y} = M_T(1 - \exp(-kt)) \qquad 3\text{-}1$$

$$y = M_T\, exp\left\{-exp\left[\frac{\mu(\lambda - t)e}{M_T} - 1\right]\right\} \qquad 3\text{-}2$$

where y (mL/g-VS) is the cumulative methane production at time t (d); M_T (mL/g-VS) is the total methane production during the 30 days' AD; e is Euler's number that equals to 2.718282; k (d^{-1}) is the specific biogas production rate constant estimated from the first-order kinetic model (Syaichurrozi and Sumardiono, 2014); μ (mL/(g-VS·d)) and λ (d) are the maximum methane production rate and lag phase time estimated from the Gompertz model, respectively (He *et al.*, 2017).

3.2.4 Analytical methods

The analytical methods were as the same as in Chapter 2 (2.2.3 and 2.2.4).

In addition, the saturation index (SI) of oversaturated CO_3^{2-} precipitates that were possibly formed during AD were estimated using Visual MINTEQ ver. 3.1 (developed by Jon Petter Gustafsson at KTH, Sweden) according to the soluble concentrations of metal cations, soluble inorganic carbon and the pH values and temperature of digestate at each condition.

3.3 Results and discussion

3.3.1 Biogas upgrading

The changes of methane content during 30 days' AD with or without biogas recirculation at the increasing dosage of $FeCl_3$ are illustrated in Fig. 3-1a. With the increase of $FeCl_3$ addition, the initial methane content was lowered compared with the Control (R0). The methane contents in R0-R3 became stable after day 6, while the average methane content was obviously enhanced with the increasing dosage of $FeCl_3$, maintaining at around 77% in R0 and R1, 78% in R2 and 80% in R3. In the reactors with biogas recirculation, the methane content gradually increased, while different phenomenon was observed in the reactors with different amount of $FeCl_3$ addition. In R0+biogasR and R1+biogasR, the methane content increased to the highest (85-86%) on day 14, then it began to decline afterwards, to the same levels as in corresponding reactors without biogas recirculation after day 26. In R2+biogasR and R3+biogasR, the methane content continuously increased until day 30 to around 87% and 88%, respectively. The above results indicated that the added $FeCl_3$ might contribute to the enhanced methane content in the produced biogas during AD with or without biogas recirculation, probably due to the formation of $FeCO_3$ precipitate (K_{sp}=3.13×10^{-11}).

Fig. 3-1b shows the cumulative methane production during 30 days' AD. The methane yield on day 30 at each condition showed no significant difference, indicating that the $FeCl_3$ addition up to 900 mg-Fe/L was not toxic to methanogenesis during AD. However, the methane production rate seems to reduce with the increasing amount of $FeCl_3$ addition. The cumulative methane production at each condition was fitted to the first order kinetic and modified Gompertz models to better understand the effect of biogas recirculation and $FeCl_3$ addition on the performance of AD. The former model is generally used to explain the hydrolysis process, while the latter can be used to estimate the maximum methane production rate and lag phase during AD. The estimated parameters from the two models are shown in Table 3-2. The modified Gompertz model fitted all the experimental data much better than the first order kinetic model, especially under higher $FeCl_3$ addition conditions. The k value calculated from the first order kinetic model could represent the hydrolysis rate during AD (Syaichurrozi and Sumardiono, 2014). Obviously, the k value was significantly lower in R3 and R3+biogasR with 900 mg-Fe/L of $FeCl_3$ addition, in agreement with the lowered maximum methane production rate (μ) and prolonged lag phase (λ) estimated by the modified Gompertz model. Fe^{3+} could be efficiently bounded with organic compounds, probably decreasing the degradation rate of organics with resultant slow methane production rate during AD (Dentel and Gossett, 1982). When comparing the estimated data in the reactors with and without biogas recirculation, lower k and μ values were also obtained in the reactors with biogas recirculation. Although biogas recirculation did not affect the total methane yield and organic carbon degradation (Fig. 3-1c) during AD, the long-term effect of biogas recirculation on AD performance remains unclear, which should be further confirmed.

As seen from Fig. 3-1c, the organic carbon degradation during AD was not significantly affected even by addition of $FeCl_3$ up to 900 mg-Fe/L, which was consistent with the similar methane yield obtained under each condition. Compared with the results from Chapter 2, $FeCl_3$ showed a better performance on biogas upgrading with the similar molar concentration of Fe^{3+} or Mg^{2+} addition (15 mmol/L), probably due to the excellent flocculation ability of trivalent Fe^{3+} and the lower solubility product (K_{sp}) of Fe-related precipitates. In Chapter 2, the results also indicated the possibility that the decreased CO_2 in biogas was dissolved in sludge liquor or adsorbed on sludge particles during biogas recirculation. In this chapter, the effect of biogas recirculation on biogas upgrading was evaluated at different concentrations of $FeCl_3$ addition and the possible precipitates of CO_3^{2-} was estimated by the saturation index (SI) using Minteq v3.1 database according to Huang *et al.* (2015). Fig. 3-2 represents the SI of oversaturated CO_3^{2-} related precipitates in digestates after AD with or without biogas recirculation and $FeCl_3$ addition. The SI of all precipitates increased with the increasing dosage of $FeCl_3$, and biogas recirculation further increased the SI values. The dissolution of Ca^{2+} and Mg^{2+} was promoted by the increasing dosage of $FeCl_3$ (Fig. 3-3), which also contributed to the higher SI of $CaCO_3$ and $CaMg(CO_3)_2$.

3.3.2 Phosphorus conservation

The digestate liquor with high P and N concentrations generated after digested sludge being dewatered is usually returned to aerobic tank. The P concentration up to hundreds ppm contributes to 10-50% of nutrients and organics loading in the main stream of WWTPs (Ivanov *et al.*, 2009). Thus, it's essential to reduce the nutrients concentrations in the soluble phase of digested sludge. On the other hand, P recovery from digestate liquor would be more efficient compared with wastewater due to its high concentration of ortho-P. $FeCl_3$ is the common chemical used for phosphorus removal in WWTPs. Though many researchers argued that Fe-P has low bioavailability, thus reducing the P recovery efficiency (Melia *et al.*, 2017). However, the most recent publications stated that the main form of precipitated phosphate in sewage sludge of WWTPs using Fe flocculant is vivianite, of which the fertilizer values has been reported by many researchers (Eynard *et al.*, 1992; Rosado *et al.*, 2002; Wilfert *et al.*, 2018). Except for the formation of Fe-P precipitates, the ortho-P also could be adsorbed on the amorphous ferric hydroxides (oxides) formed under neutral pH condition or involved in iron-organic-phosphorus complex in the sludge (Wilfert *et al.*, 2015). The initial and final values of soluble ortho-P in sewage sludge during AD with or without biogas recirculation and $FeCl_3$ addition are shown in Fig. 3-4. With the increase amount of $FeCl_3$ addition, the ortho-P were linearly decreased. The lowest soluble ortho-P was 10.15 mg-P/L, obtained on day 30 with biogas recirculation and $FeCl_3$ addition at a Fe:P_{ortho} molar ratio of 1.5, which has been reported as the optimal molar ratio for P removal (Caravelli *et al.*, 2010; De Gregorio *et al.*, 2010). The final concentration of soluble ortho-P in the reactors with biogas recirculation showed slightly lower value compared with those without biogas recirculation. The percentages of P recovered in solid were 48.2%, 64.6%, 84.5% and 98% in R0-R3, respectively, while they were 50.1%, 67.0%, 87.2% and 98.7% in the corresponding reactors with biogas recirculation. The results were consistent with those in Chapter 2, indicating that biogas recirculation might promote the P precipitation during AD.

3.3.3 Sludge properties

The sludge dewaterability and settleability were evaluated during AD with or without biogas recirculation and $FeCl_3$ addition. As seen in Fig. 3-5a, the SRF decreased from 2.26×10^{13} m/kg to 1.43×10^{13}, 0.85×10^{13} and 0.5983×10^{13} m/kg after adding $FeCl_3$ in the mixed sludge to 27.74, 49.32 and 68.63 mg-Fe/g-TS, respectively. After AD without biogas recirculation, the SRF did not show significant difference compared with their initial values except for the control without $FeCl_3$ addition (R0). However, the AD with biogas recirculation worsened sludge dewaterability in R0+biogasR and R1+biogasR, while in R2+biogasR and R3+biogasR, the SRF did not show significant difference compared with the corresponding initial and final values in the reactors without biogas recirculation. The results indicated that the $FeCl_3$ content over 49 mg-Fe/g-TS could prevent the deterioration of sludge dewaterability brought about by biogas recirculation during AD. The reported optimal dosage of $FeCl_3$ for enhanced sludge dewaterability varied from 34 to 110 mg-Fe/g-TS, resulting in SRF reduction by 60-94% as shown in Table 3-1 (Liu *et al.*, 2012; Niu *et al.*, 2013; Guo and Ma, 2015; Wu *et al.*, 2016a). The $FeCl_3$ dosage in this study was in the optimal range reported in previous works.

The sludge settleability represented by diluted SVI_{30} was also enhanced with the increasing dosage of $FeCl_3$ (Fig. 3-5b). The diluted SVI_{30} decreased from initial 140.5 mL/g-TS to 75.3 mL/g-TS by adding 68.63 mg-Fe/g-TS. After 30 days' AD without biogas recirculation, the SVI_{30} slightly increased in R0 and R1, remained the same in R2, and decreased in R3 compared to their initial values. In addition, the sludge settleability was also significantly enhanced by biogas recirculation in all the reactors with biogas recirculation. The same results were obtained in Chapter 2, where detailed explanations were given.

3.4 Summary

The effect of $FeCl_3$ addition on the performance of the novel AD system with intermittent biogas recirculation was investigated in this chapter. The addition of $FeCl_3$ up to 900 mg-Fe/L did not enhance or reduce the biogas yield and organic carbon reduction during 30 days' AD of sewage sludge, while the dosage of 900 mg-Fe/L obviously reduced the biogas production rate. The intermittent biogas recirculation coupling $FeCl_3$ addition of 900 mg-Fe/L during 30 days' AD achieved the followings: 1) enhanced methane content to 88% compared to 77% in the control reactor (R0); 2) 98.7% of P conservation in the solid phase of digested sludge (Fe:P_{ortho} molar ratio=1.5); 3) enhanced sludge dewaterability and settleability by 78.6% and 56.5% compared to the control (R0).

Table 3-1 Summary of previous works on utilization of Fe^{3+} in wastewater and waste treatment process.

Substrates	Fe sources	Dosages	Achievement	References
Wastewater in primary clarifier	$FeCl_3$	Fe:P_S=0.8:1	P removal by 43%, COD removal by 50%, SS removal by 64%	Ghyoot and Verstraete (1997)
		20 mg-Fe/L	Ortho-P removal by 99.3% (3.93 mg/L ortho-P in raw sewage), TOC removal by 75.6%	Lin *et al.* (2017)
Wastewater in Activated sludge reactor	$FeCl_3$	Fe:P_S=1.5-1.9:1	P removal by 95%, improved COD and SS removal	De Gregorio *et al.* (2010)
Sewage sludge in AD reactor	Ferrihydrite	Fe:P_T=1.5:1	P removal by 53%	Cheng *et al.* (2015)
Sewage sludge in AD reactor	$FeCl_3$	172.5 mg-Fe/g-DS	Control of H_2S and odor causing compounds	Park and Novak (2013)
Sewage sludge in Thermophilic AD reactor	$FeCl_3$	200 mg-Fe/L	Increased methane yield by 98.9%	Yu *et al.* (2015)
Reject water after dewatering of digested sludge	Fe_2O_3	Fe:P_S=0.75-1.15:1	P removal by 90%, TOC removal by 90%	Ivanov *et al.* (2009)
Sewage sludge before dewatering	$FeCl_3$	34.5 mg-Fe/g-DS	SRF reduction by 94%	Niu *et al.* (2013)
		110 mg-Fe/g-DS	SRF reduction by 60%	Guo and Ma (2015)
		81.8 mg-Fe/g-DS	SRF reduction by 93.8%	Liu *et al.* (2012)
		39.7 mg-Fe/g-DS	SRF reduction by 80%	Wu *et al.* (2016a)
		34.5 mg-Fe/g-DS	SRF reduction by 50%	Lin *et al.* (2014)

P_S, soluble phosphorus; P_T, total phosphorus; COD, chemical oxygen demand; SS, suspended solids content; TOC, total organic carbon; AD, anaerobic digestion; DS, dry solids content; SRF, specific resistance to filtration.

Table 3-2 Parameters estimated from the first-order kinetic and modified Gompertz models for methane production from primary sludge under different conditions.

Models	Parameters	Reactors							
		R0	R1	R2	R3	R0+biogasR	R1+biogasR	R2+biogasR	R3+biogasR
First-order	k (d^{-1})	0.163	0.151	0.133	0.091	0.151	0.131	0.126	0.083
	R^2	0.953	0.963	0.932	0.883	0.972	0.970	0.896	0.891
Gompertz	μ (mL/(g-VS·d))	22.37	19.67	19.51	16.42	18.68	16.14	19.76	14.25
	λ (d)	0.71	0.46	1.46	3.31	0.18	0.19	2.20	3.18
	R^2	0.993	0.993	0.998	0.998	0.996	0.993	0.998	0.997

k, specific biogas rate constant; μ, maximum methane production rate; λ, lag phase time; R^2, correlation coefficient.

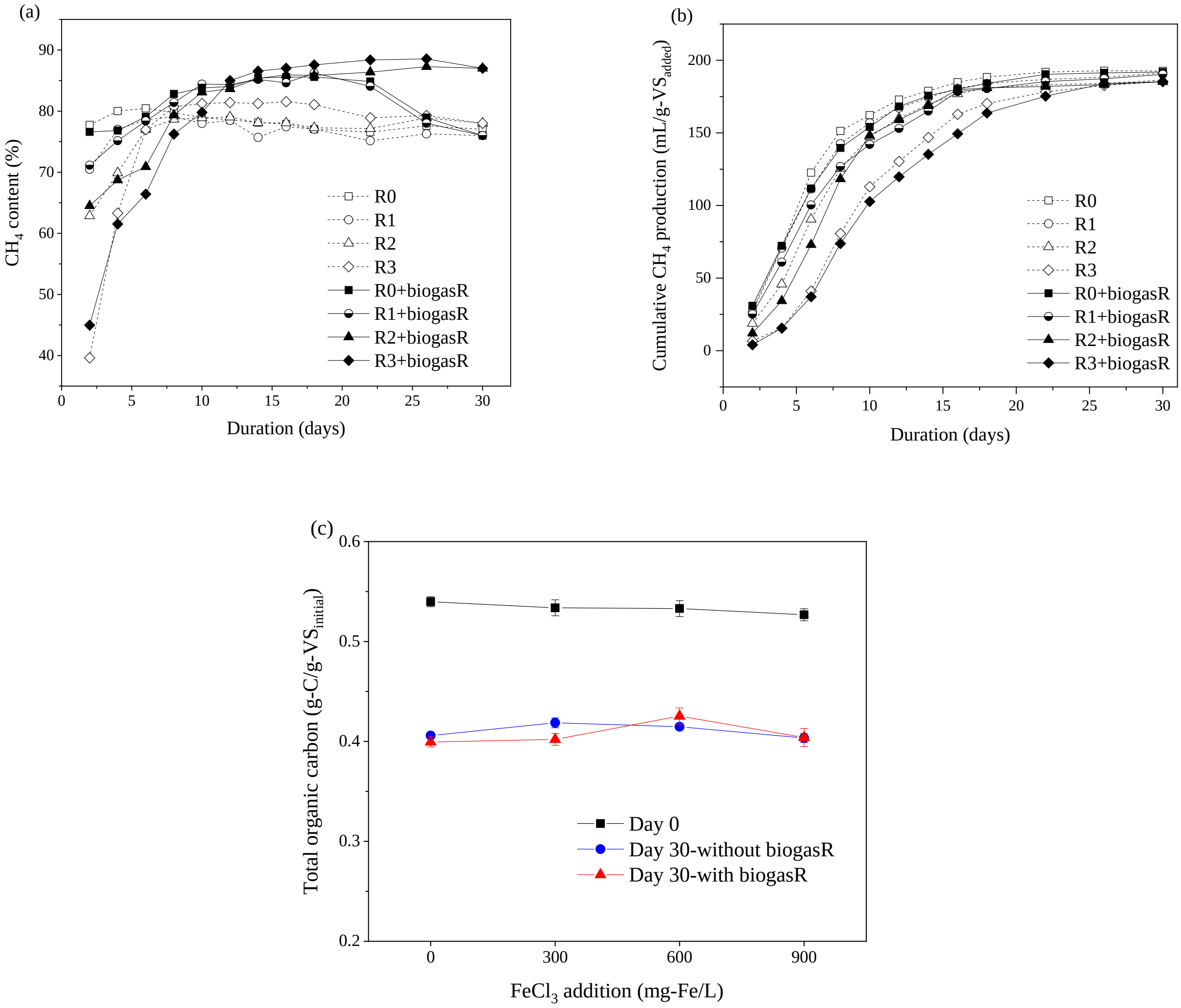


Fig. 3-1 The changes of CH_4 content (a), cumulative CH_4 production (b) and total organic carbon (c) during anaerobic digestion with or without biogas recirculation and $FeCl_3$ addition.

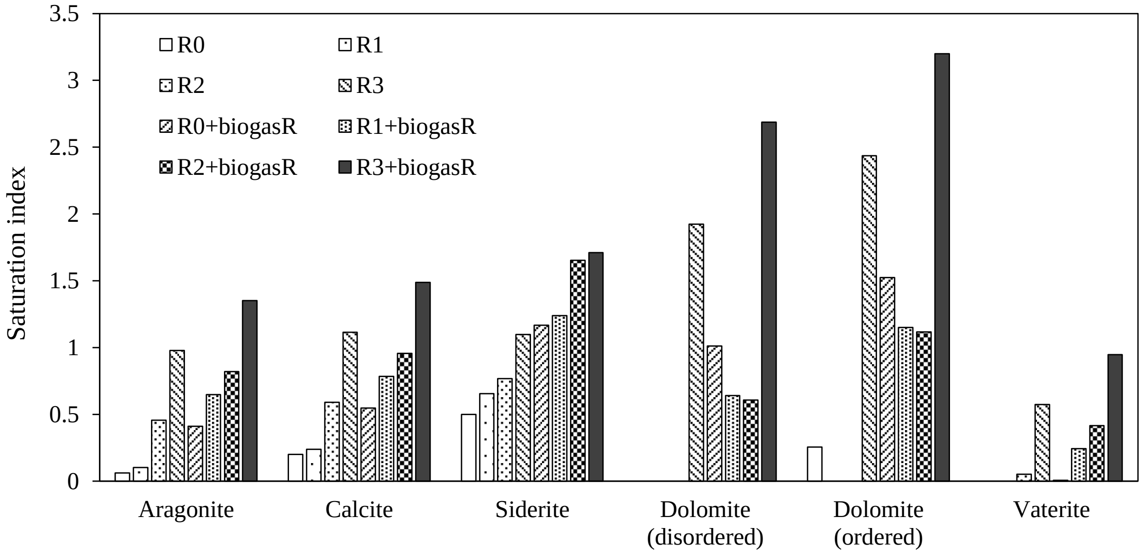


Fig. 3-2 Saturation index of oversaturated CO_3^{2-} related precipitation in digestate after anaerobic digestion with or without biogas recirculation and $FeCl_3$ addition. The data were estimated by using Minteq. V3.1.

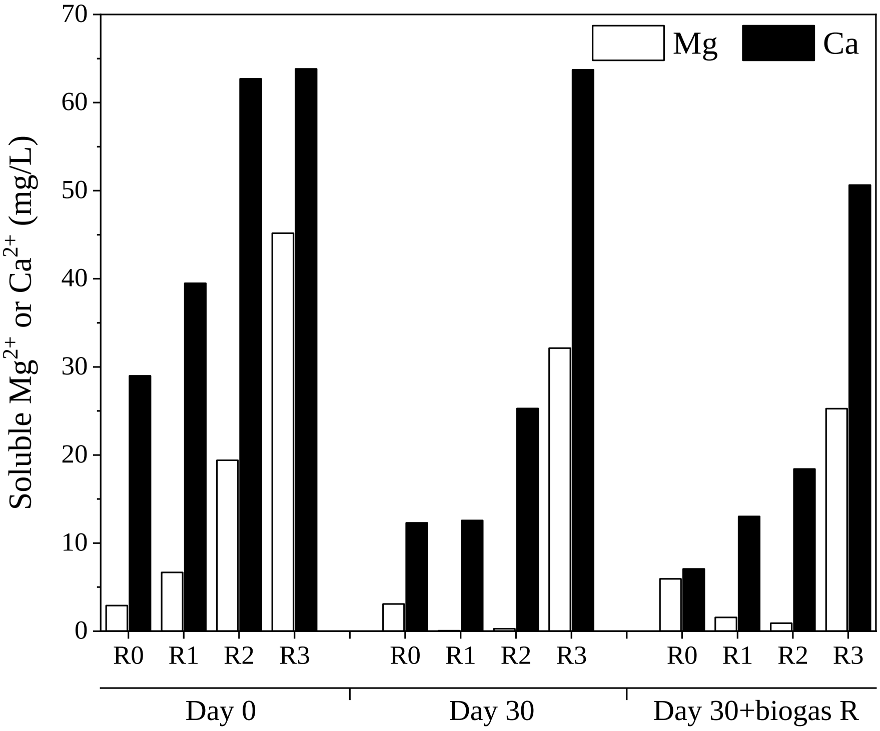


Fig. 3-3 The changes of Mg^{2+} and Ca^{2+} ions during anaerobic digestion with or without biogas recirculation and $FeCl_3$ addition.

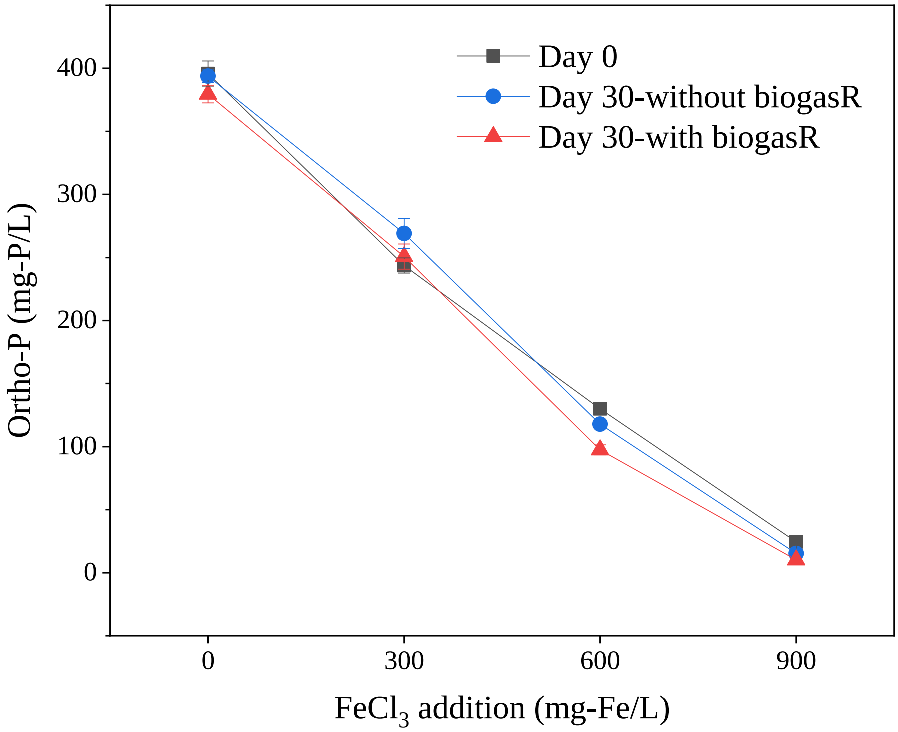


Fig. 3-4 The soluble ortho-P concentration on day 0 and day 30 during anaerobic digestion with or without biogas recirculation and $FeCl_3$ addition.

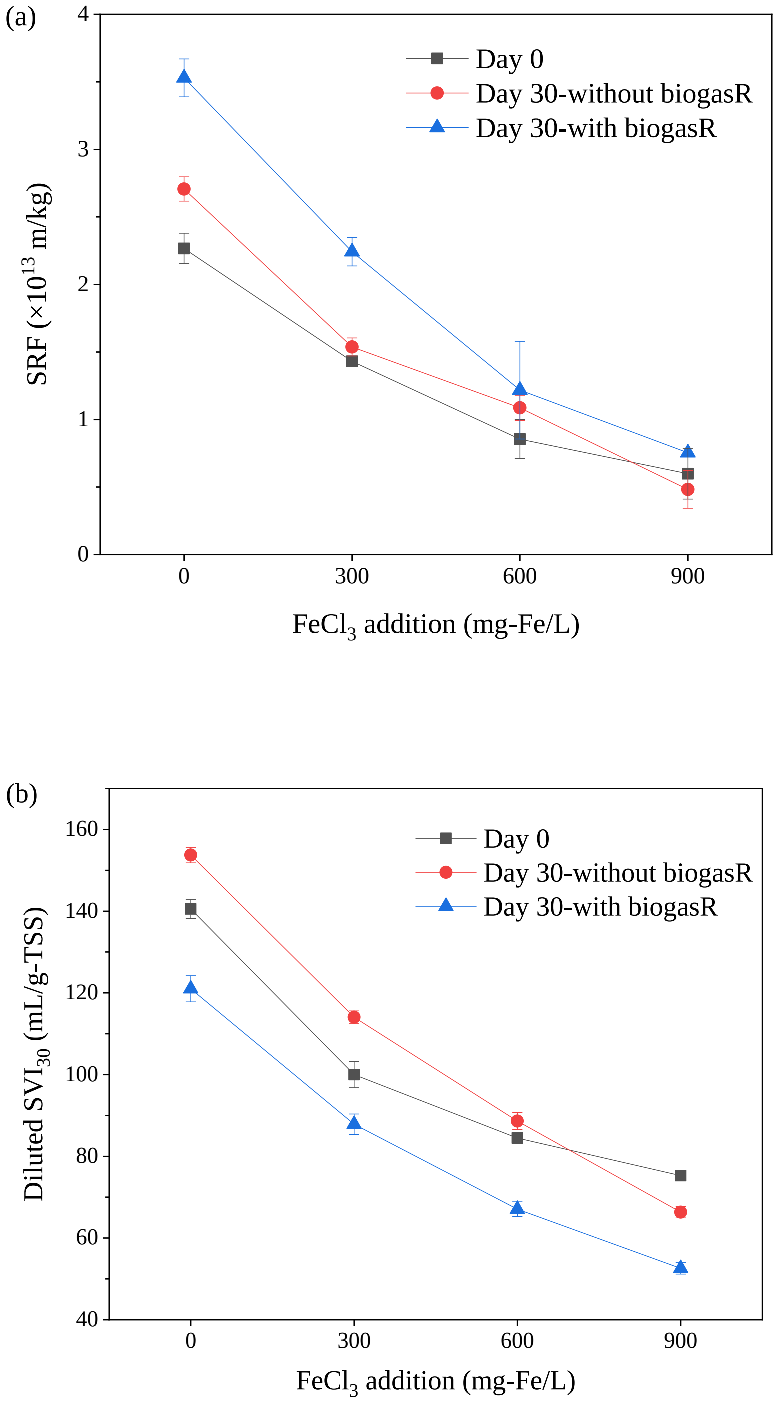


Fig. 3-5 The specific resistance to filtration (SRF, a) and diluted sludge volume index within 30 min (SVI_{30}, b) on day 0 and day 30 during anaerobic digestion with or without biogas recirculation and $FeCl_3$ addition.

Chapter 4 Semi-continuous operation of the novel anaerobic digestion system with biogas recirculation

4.1 Background

In this chapter, the long-term effect of biogas recirculation on AD performance was studied through semi-continuous mode. At the same time, the effects of $MgCl_2$ and $FeCl_3$ addition on phosphorus recovery and sludge dewaterability during semi-continuous AD was also compared.

Struvite crystallization is the most commercialized method in industries for phosphorus and nitrogen recovery from digested liquor, due to the marketable value of struvite as a slow-releasing fertilizer (Melia *et al.*, 2017). However, additional equipment is required to realize struvite crystallization. In addition, the phosphate addition is usually needed due to the low P/N molar ratio (<1) in digested liquor. As reported, the optimal pH for struvite precipitation is in the range of 8.5-10.0, which also increases the operation cost for pH adjustment (Nelson *et al.*, 2003; Yu *et al.*, 2017; Zeng *et al.*, 2018).

$FeCl_3$ is the most commonly used coagulant in WWTPs, due to its excellent capacity on phosphorus removal and sludge conditioning. Although the bioavailability of Fe-P precipitates has been argued by many researchers, more and more attention has been focused on phosphorus recovery in the form of Fe-P due to its low solubility of Fe-associated precipitates and the widely utilization of Fe (Wilfert *et al.*, 2018). The addition of $FeCl_3$ may be also beneficial for phosphorus recovery inside the AD reactor because of the neutral pH requirement for Fe-P precipitates.

In Chapters 2 and 3, the effect of $FeCl_3$ and $MgCl_2$ addition coupling with biogas recirculation during AD were investigated separately by using batch AD test. In this chapter, the effect of divalent Mg^{2+} and trivalent Fe^{3+} addition during AD on sludge dewaterability was compared by adding $FeCl_3$ and $MgCl_2$ at a same equivalent concentration.

4.2 Materials and methods

4.2.1 sludge samples

The concentrated excess sludge and digested sludge were sampled from the Shimodate WWTP in Chikusei, Ibaraki, Japan. The sludge samples were stored in 20 L polypropylene tanks at 4 °C in laboratory. The main characteristics of the concentrated excess sludge were as follows: TS 10.8±0.13 g/L, VS 8.8±0.14 g/L, TP 128.7±2.25 mg-P/L, soluble ortho-P 69.5±1.21 mg-P/L, TN 394.5±8.70 mg-N/L, ammonia N 69.2±2.13 mg-N/L, and pH 5.34±0.02.

4.2.2 Optimization of $MgCl_2$ or $FeCl_3$ dosage for sludge dewatering

Since one of the purposes by adding Mg^{2+} or Fe^{3+} during AD was to enhance the sludge dewaterability, the optimum dosages of $MgCl_2$ and $FeCl_3$ were determined before semi-continuous AD. An equivalent concentration was adopted to make divalent Mg^{2+} ions and trivalent Fe^{3+} ions comparable. $MgCl_2$ and $FeCl_3$ salts were added to 100 mL of concentrated excess sludge at 4.62, 9.24 or 13.9 meq./L, respectively, with the corresponding concentrations of $MgCl_2$ being 220, 440 and 660 mg/L and those of $FeCl_3$ being 250, 500 and 750 mg/L, respectively. After a rapid shaking at 150 rpm for 1 min and a slow mix at 50 rpm for 4 min, the sludge dewaterability of the conditioned sludge was determined by vacuum filtration method and the data were represented as SRF. The detailed procedure for SRF determination could be found in section 2.2.4.

4.2.3 Start-up and operation of semi-continuous AD system

The semi-continuous AD reactors used in this study had the similar configuration as that in Chapter 2, with two additional ports as sludge inlet and outlet on the side wall of the reactors. The two reactors (namely R0 and R1) were fed with the mixture of 0.4 L concentrated excess sludge and 0.2 L digested sludge, then the reactors were purged by nitrogen gas for 5 min to create an anaerobic condition. After 20-day continuous operation at 37±2°C, 120 mL sludge exchange was performed every 3 days to realize the semi-continuous operation. During the exchange, 120 mL of treated sludge was extracted with another 120 mL of raw concentrated excess sludge being fed into each reactor after pH being adjusted to neutral. The hydraulic retention time (HRT) was controlled at 15 days according to the results of Chapter 2, and the organic loading rate (OLR) was 0.59 g-VS/(L·d). After 15-day semi-continuous operation, the reactors were put to data collection with operation conditions listed in Table 4-1. As seen, $MgCl_2$ was added to R1 at a concentration of 440 mg/L from phase 2 to phase 5. Since phase 3 and onward, an intermittent biogas recirculation (1 h-on/2 h-off) was started at a flowrate of 100 mL/min in both reactors. In phase 4 and phase 5, the HRT of both reactors was reduced to 12 days since the biogas production was noticed to accelerate in phase 3 with biogas recirculation. Thus during the last 2 phases, the sludge exchange interval was maintained as 3 days, while 150 mL of sludge was exchanged each interval (HRT=12 days). Additionally, $FeCl_3$ was added to R0 during phase 5 at a concentration of 500 mg/L in order to compare its effect on phosphorus recovery and sludge dewaterability with $MgCl_2$ at an identical equivalent concentration.

4.2.4 Analytical methods for the general parameters

All the analytical methods were as the same as in Chapter 2 (2.2.3 and 2.2.4).

4.2.5 Analysis of archaeal and bacterial communities

The effluent sludge samples in R0 were collected on day 45 in phase 2 and day 69 in phase 3 of semi-continuous AD tests to demonstrate the effects of biogas recirculation on archaeal and eubacterial communities. The samples were concentrated by centrifuge and used for deoxyribonucleic acid (DNA) extraction using DNeasy PowerMax Soil Kit (QIAGEN, USA) according to the manufacturer's instructions. The DNA samples were stored below -20°C before delivering to Majorbio Biotech Co., Ltd. (Shanghai, China) for diversity analysis of microbial communities. The archaeal and eubacterial 16S rDNA were amplified via polymerase chain reaction (PCR) with primer sets of 524F10extF/Arch958RmodR and 338F/806R, respectively (Cai *et al.*, 2019). High-throughput sequencing was performed after the PCR products were quantitated by Majorbio Biotech Co., Ltd. (Shanghai, China).

4.3 Results and discussion

4.3.1 Optimization of $MgCl_2$ and $FeCl_3$ dosages for sludge dewatering

The SRF of sludge was decreased by 58.6%, 87.6% and 87.1% with $MgCl_2$ addition, and by 91.4%, 97.2% and 98.2% with $FeCl_3$ addition, at equivalent concentrations of 4.62, 9.24 and 13.9 meq./L, respectively (Fig. 4-1). The primary mechanism for enhanced sludge dewaterability by multivalent cations is the charge neutralization (Sobeck and Higgins, 2002). $FeCl_3$ exhibits superior sludge dewaterability enhancement to $MgCl_2$, since the trivalent Fe^{3+} ions could not only more effectively neutralize the surface charge of sludge flocs, but also form amorphous ferric hydroxide species to sweep colloidal precipitates out from water (Kang, 1994). The optimal dosages of $MgCl_2$ and $FeCl_3$ were determined as 9.24 meq./L, corresponding to 440 mg/L $MgCl_2$ and 500 mg/L $FeCl_3$, respectively.

4.3.2 Sludge dewaterability

Fig. 4-2 shows the changes of average SRF value of effluent sludge samples in each phase during the semi-continuous AD. The average SRF of effluent sludge from both reactors during phase 1 was around 9.67×10^{12} m/kg, close to that of the feed sludge. Although TS and VS can be reduced in AD, the degraded extracellular polymeric substances (EPS) may reduce flocculation ability of sludge flocs and deteriorate sludge dewaterability (Wang *et al.*, 2018a). In this study, $MgCl_2$ was added in R1 from phase 2 to phase 5. In phase 2, the SRF of effluent sludge samples from R1 (with $MgCl_2$ addition) was 3.3×10^{12} m/kg, reducing by 65% compared to R0. In phase 3 with biogas recirculation, the SRF of effluent sludge samples was slightly decreased in both reactors, likely resulted from the reduced solids content during phase 3-5 (Fig. 4-7). In phase 4, the HRT was reduced to 12 days owing to the accelerated biogas production via biogas recirculation. The average SRF did not show significant change compared with that in phase 3. $FeCl_3$ was added in R0 during phase 5, where the SRF of effluent sludge samples was immediately decreased to 6×10^{11} m/kg, around 94% lower than that in phase 1. As shown in Fig. 4-3, almost all Fe was accumulated in sludge pellets in R0 after $FeCl_3$ addition during phase 5, while half of Mg was presented in soluble phase in R1 with $MgCl_2$ addition (phase 5). This observation also indicates the strong binding capacity of Fe^{3+} with negatively charged sludge particles, which likely contributed to the noted preferable sludge dewaterability enhancement (Li *et al.*, 2014).

4.3.3 Phosphorus conservation

Fig. 4-4 shows the average soluble ortho-P concentrations and pH of effluent sludge samples during the five phases of semi-continuous AD operation. The lowest soluble ortho-P (3.4 mg/L) was detected in R0 during phase 5 with $FeCl_3$ addition. The existence of vivianite-like crystal in the digestate was confirmed using XRD (Fig. 4-5): all the positions of relative peaks are close to those of standard vivianite, indicating the high possibility of vivianite formation during AD with $FeCl_3$ addition (Liu *et al.*, 2018).

In contrast, $MgCl_2$ addition in R1 from phase 2 to phase 5 showed little effect on soluble ortho-P reduction, except for that in phase 3 (soluble ortho-P=42 mg-P/L), in which pH was increased to 7.3. The pH during batch AD with biogas recirculation (chapter 2 and 3) was noted to increase to 7.9, which might be attributable to the dissolved carbonate in sludge liquor. However, the reduction of HRT did not alter the sludge pH in phases 4 and 5, which might be yielded by the increased sludge exchanging rate resulting in practical reduction of the duration for biogas recirculation through the sludge. Seen from Fig. 4-3, around half of Mg^{2+} was presented in the soluble phase of digested sludge, indicating its lower efficiency on sludge bridging and phosphorus precipitation compared with Fe^{3+} ions. This observation also correlates with the fact that struvite precipitation largely depends on pH value when the soluble ortho-P is lower than 60 mg/L (Nelson *et al.,* 2003). In fact, the previously reported final ortho-P concentration after struvite precipitation in digested liquid are commonly high. For instance, the final ortho-P concentrations were 89.26, 48.73 and 27.43 mg-P/L after struvite precipitation at initial pH of 8.0, 9.0 and 10.0, respectively, and a Mg:P molar ratio of 1:1 in hydrolysate with an initial ortho-P of 579.98 mg-P/L (Yu *et al.*, 2017). A final ortho-P of 20 mg-P/L was obtained in digested sludge after struvite precipitation at an initial pH 8.0, initial ortho-P of 80 mg-P/L and a Mg:P molar ratio of 1:0.8 (Wang *et al.*, 2018a). Several studies indicate that although the struvite precipitation is suggested as a promising method for phosphorus recovery, the recovery efficiency is relatively low due to the existence of other metal-P precipitates with lower solubility product (K_{sp}) than struvite in sewage sludge (Cornel and Schaum, 2009; Egle *et al.*, 2015)

4.3.4 AD performance

Fig. 4-6 reveals the specific biogas or methane yields and methane content during the 105 days' semi-continuous AD. In phase 1 (0-24 d), with neither biogas recirculation nor chemical addition, the average methane yields were 319.3 and 312.5 mL/g-VS_{fed} in R0 and R1, respectively. These two reactors showed similar performances. In phase 2 (25-45 d), the average specific methane yields were 325.7 (R0) and 327.3 (R1) mL/g-VS_{fed} and the methane content were both around 71.5%, suggesting that $MgCl_2$ addition in R1 did not significantly affect the methanogenesis during AD. However, the specific methane yields noted were more stable than phase 1. In phase 3, with the intermittent biogas recirculation, the average specific methane yields in R0 and R1 were 352.4 and 348.5 mL/g-VS_{fed}, respectively, increased by around 9% than phase 1 and phase 2. The methane contents were also gradually increased to around 82.4% after 6-day biogas recirculation, equivalent to 11% enhancement on methane contents. This enhancement is close to that in batch AD tests with biogas recirculation in chapter 2 and 3. In phase 4, the HRT was reduced from 15 days to 12 days due to the accelerated methane production. The average methane yields reached to 368.9 and 365.2 mL/g-VS_{fed} in R0 and R1, respectively, corresponding to 13% increase compared with those in phase 2. In phase 5, $FeCl_3$ was added into R0, with resultant average methane yields of 363.1 and 365. 3 mL/g-VS_{fed} in R0 and R1, respectively, reflecting no significant difference from those in phase 4.

In summary, neither $MgCl_2$ or $FeCl_3$ addition affected methane production or methane contents in biogas during the semi-continuous AD, while the intermittent biogas recirculation enhanced the methane yield by 13% and methane contents by 11%, respectively. The enhanced methane yield and methane contents by biogas recirculation in this study may be contributed by the following four aspects: 1) the promoted contact between substrate to bacteria (Lindmark *et al.*, 2014); 2) the enhanced dissolution of carbon dioxide in sludge liquor; 3) the promoted H_2/CO_2 pathway for methanogenesis; and 4) the efficient gas-liquid-solid (GLS) separation (Umaiyakunjaram and Shanmugam, 2016). Biogas recirculation has been claimed to be able to create chemical equilibrium and control alkalinity and NH_3 formation during AD (Latha *et al.*, 2019), which should not correlate with the present AD system since the digestate ammonia was low (69.2 mg-N/L).

The average VS contents of sludge samples from the semi-continuous AD reactors are shown in Fig. 4-7. In phase 1, the average VS content was 3.54 g/L, which was reduced by 59.8% compared with the feed as a result of biodegradability. In phase 2, the VS content was 3.47 g/L, which was reduced by 2% compared with phase 1, corresponding to the minor increase in methane yield noted in phase 2. In phase 3 to phase 5, the VS content was stabilized at around 3.3 g/L, 5.7% lower compared with phase 1 and 2; whereas the methane yield was increased by 13%. The reduced VS corresponded to the enhanced hydrolysis for increased acetate generation and therefore contributing to the enhanced methane production. The methane yield in R0 in phase 4 with biogas recirculation was around 580 mL/g-$VS_{reduced}$, which was higher than R0 during phase 1 without biogas recirculation (530 mL/g-$VS_{reduced}$), possibly resulting from the enhanced bacterial activity under biogas recirculation.

4.3.5 Archaeal and eubacterial communities

The relative abundances of archaeal community at order and genus levels in R0 during phase 2 (without biogas recirculation) and phase 3 (with biogas recirculation) are displayed in Fig. 4-8. The total sequences numbers of the detected samples were 41229 and 40420, respectively, with a mean length of around 429 base pairs. Methanosarcinales and Methanomicrobiales belonging to the phylum Euryarchaeota were the dominant archaeal orders in the semi-continuous AD reactor, indicating that the AD was driven by both acetoclastic and hydrogenotrophic methanogenesis (Liu *et al.*, 2009; Ziganshin *et al.*, 2013; Venkiteshwaran *et al.*, 2015). The relative abundance of these orders was increased from 52% in phase 2 to 76% in phase 3 when biogas recirculation was applied, corresponding to the noted and increased methane yield. Acetoclastic methanogens can convert acetate to CH_4 and CO_2, contributed by archaeal order Methanosarcinales and are usually dominant during AD. The hydrogenotrophic methanogens consisting of orders Methanomicrobiales, Methanobacteriales and Methanococcales can utilize H_2/CO_2 or formate to produce CH_4. In hydrogenotrophic pathway, acetate could be converted to H_2 and CO_2 by syntrophic acetate-oxidizing (SAO) bacteria and subsequently to CH_4 by methanogens (Liu and Whitman, 2008). Due to the positive Gibbs free energy of the conversion reaction from acetate to H_2, acetate oxidation can only occur when acetate concentration is sufficiently high and/or H_2 partial pressure is low (Lee *et al.*, 2013). Biogas recirculation during AD can reduce the H_2 partial pressure in the reactor, thus promoting the abundance of Methanomicrobiales. The enhanced abundance of Methanosarcinales was possibly due to the increased contact between substrate and microorganisms via biogas recirculation. The genus *Methanosaeta*, which belongs to order Methanosarcinales, was observed as one of the dominant genera in this study and its abundance increased from 28% in phase 2 to 42% in phase 3 with biogas recirculation. *Methanosaeta* shows high affinity for acetate and can dominate at low acetate concentration (Liu and Whitman, 2008), which is consistent with the low soluble organic content in the excess sludge used in this study (data not shown). Besides, the increased abundance of genera *Methanolinea* and *Methanospirillum* belonging to Methanomicrobiales was also observed. The results revealed that the increased methane yield with biogas recirculation was possibly attributable to the enrichment of both acetoclastic and hydrogenotrophic methanogens. In addition, large proportion of archaeal population was observed in phylum Crenarchaeota (33%), which are prevalent in extreme environment. Although their ecophysiological functions remain unclear, this phylum has been found in many AD reactors with high diversity of methanogens and usually co-occurred with Thermoplasmata (Chouari *et al.*, 2015).

The changes of bacterial community at phylum and class levels are presented in Fig. 4-9. The total sequences numbers of the detected samples were 56737 and 51005 in phase 2 without biogas recirculation and phase 3 with biogas recirculation, respectively, with a mean length of 422 base pairs. The eubacterial population was more diverse than archaeal, which mainly consisted of Bacteroidetes, Spirochaetae, Proteobacteria and Firmicutes. Most of them are known species of hydrolytic and acidogenesis bacteria (Venkiteshwaran *et al.*, 2015). The abundance of class Spirochaetes was increased from 21% in phase 2 to 31% in phase 3 with biogas recirculation. Spirochaetes are frequently detected in AD system and reported to be related to acetate oxidation (Lee *et al.*, 2013), which agrees with the increased abundance of hydrogenotrophic methanogens noted herein. The results again suggest that biogas recirculation can promote the utilization of H_2/CO_2 pathway for CH_4 production, which might also contribute to both increased CH_4 yield and content in biogas.

4.4 Summary

The long-term effect of biogas recirculation and chemical addition on biogas production, methane content, P recovery and sludge dewaterability was evaluated through semi-continuous AD experiments in this chapter. $FeCl_3$ addition during AD showed preferable sludge dewaterability and phosphorus recovery efficiencies than $MgCl_2$. The intermittent biogas recirculation enhanced specific methane yield of excess sludge by 13%, and enhanced methane content from 71.5% to 82.4%. Both acetoclastic and hydrogenotrophic methanogens were enhanced by biogas recirculation during AD, contributing to the enhanced methane yield and methane content.

Table 4-1 Operation conditions for the semi-continuous anaerobic digestion experiment.

Phases (days for start-end)	R0	R1	HRT (days)
Phase 1 (0-24)	Control	Control	15
Phase 2 (25-45)	Control	+Mg	15
Phase 3 (46-69)	+BiogasR	+Mg+BiogasR	15
Phase 4 (70-87)	+BiogasR	+Mg+BiogasR	12
Phase 5 (88-105)	+Fe+BiogasR	+Mg+BiogasR	12

HRT, hydraulic retention time; Control, without biogas recirculation and chemical addition; +BiogasR, biogas recirculation; +Mg, with $MgCl_2$ addition of 440 mg/L; +Fe, with $FeCl_3$ addition of 500 mg/L.

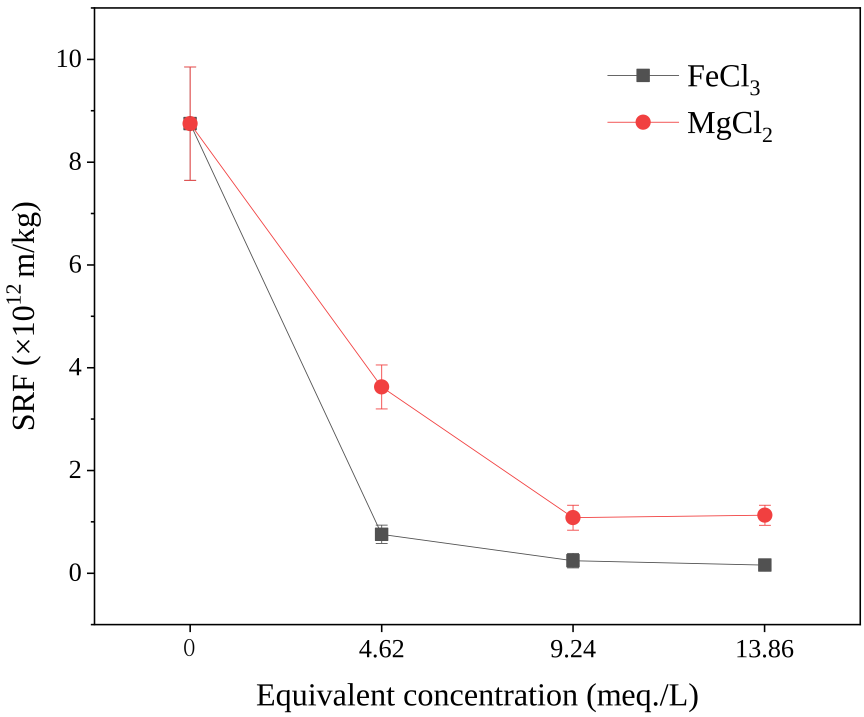
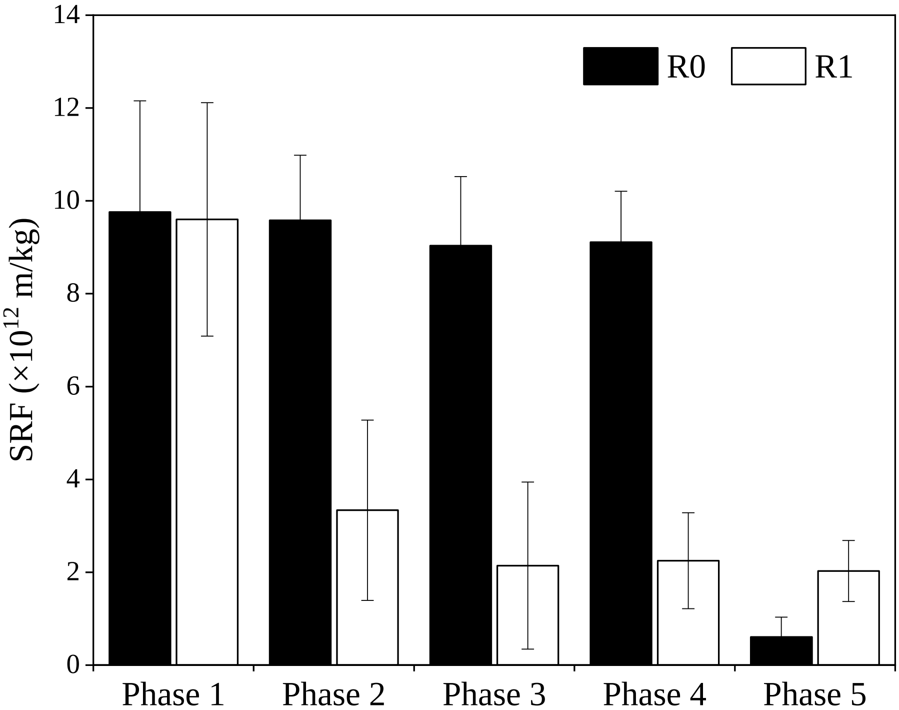


Fig. 4-1 Specific resistance to filtration (SRF) of concentrated excess sludge with different concentrations of $MgCl_2$ or $FeCl_3$ addition.



R0	Control	Control	+biogasR	+biogasR	+Fe+biogasR
R1	Control	+Mg	+Mg+biogasR	+Mg+biogasR	+Mg+biogasR
HRT	15 days			12 days	

Fig. 4-2 Average specific resistance to filtration (SRF) of effluent sludge from semi-continuous anaerobic digestion at different phases. HRT, hydraulic retention time; Control, with no biogas recirculation and no chemical addition; +biogasR, with biogas recirculation; +Mg, with $MgCl_2$ addition; +Fe, with $FeCl_3$ addition.

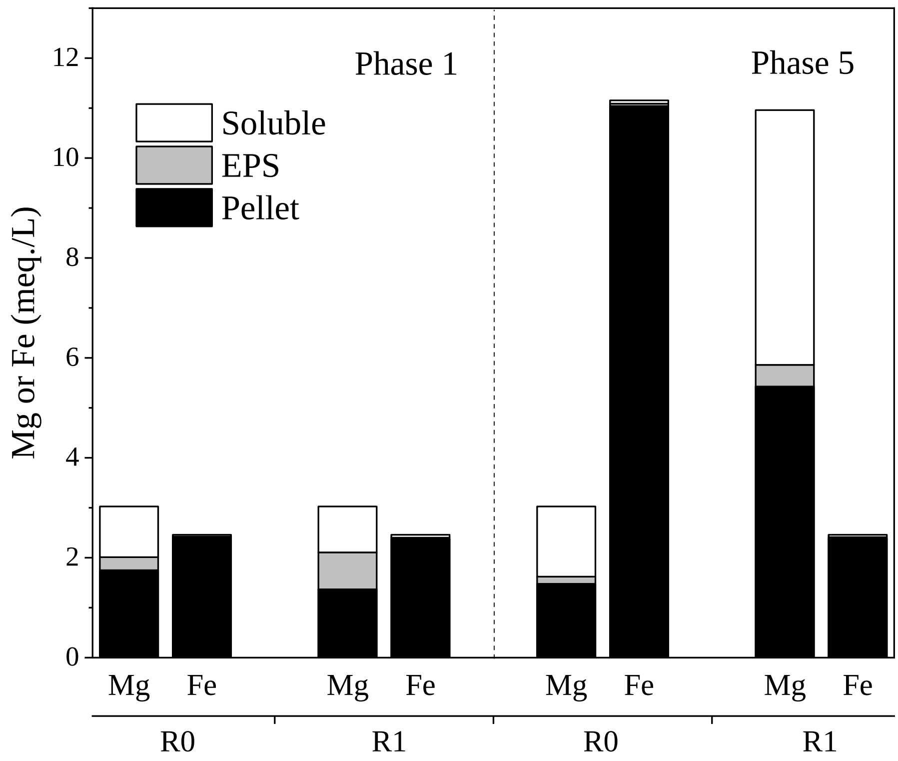
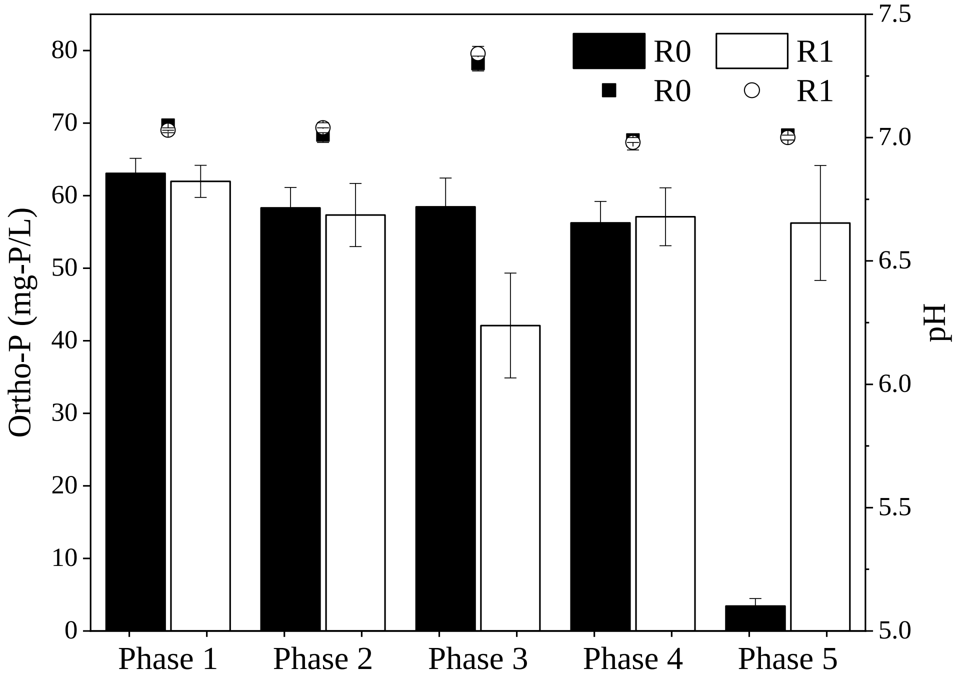


Fig. 4-3 Mg and Fe distribution in soluble phase, extracellular polymeric substances (EPS) and sludge pellet of effluent sludge in R0 and R1 during phase 1 and phase 5 under semi-continuous anaerobic digestion.



	Phase 1	Phase 2	Phase 3	Phase 4	Phase 5
R0	Control	Control	+biogasR	+biogasR	+Fe+biogasR
R1	Control	+Mg	+Mg+biogasR	+Mg+biogasR	+Mg+biogasR
HRT	15 days			12 days	

Fig. 4-4 Average soluble ortho-P concentrations (column) and the pHs (points) of effluent sludge from semi-continuous anaerobic digestion at different phases. HRT, hydraulic retention time; Control, with no biogas recirculation and no chemical addition; +biogasR, with biogas recirculation; +Mg, with $MgCl_2$ addition; +Fe, with $FeCl_3$ addition.

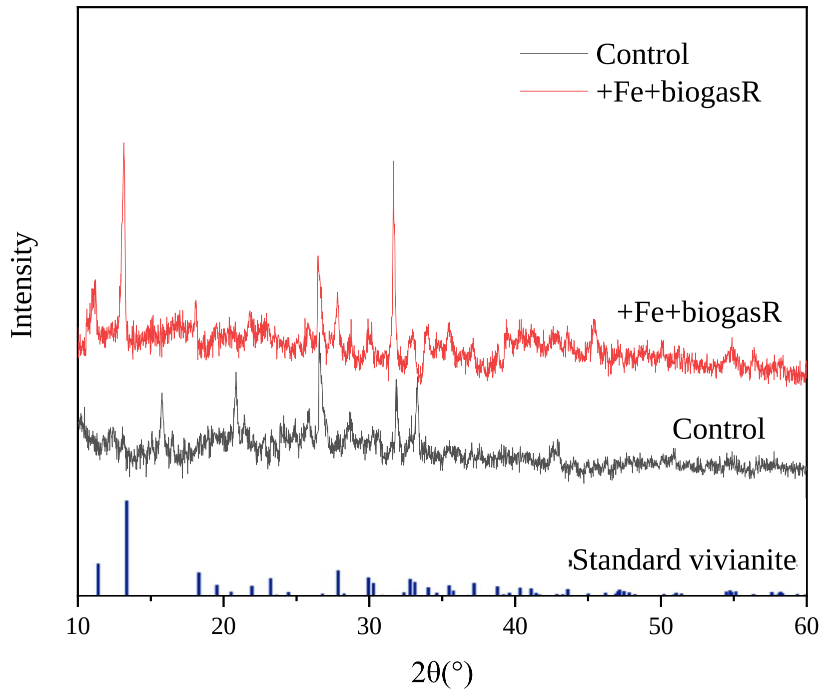


Fig. 4-5 X-ray diffraction patterns of standard vivianite and the sludge samples taken from R0 in phase 1 as control and phase 5 with $FeCl_3$ addition and biogas recirculation (+Fe+biogasR) during semi-continuous anaerobic digestion. The standard curve of vivianite was referred to Liu *et al.* (2018).

(a)

	Phase 1	Phase 2	Phase 3	Phase 4	Phase 5
R0	Control	Control	+biogasR	+biogasR	+Fe+biogasR
R1	Control	+Mg	+Mg+biogasR	+Mg+biogasR	+Mg+biogasR
HRT	15 days			12 days	

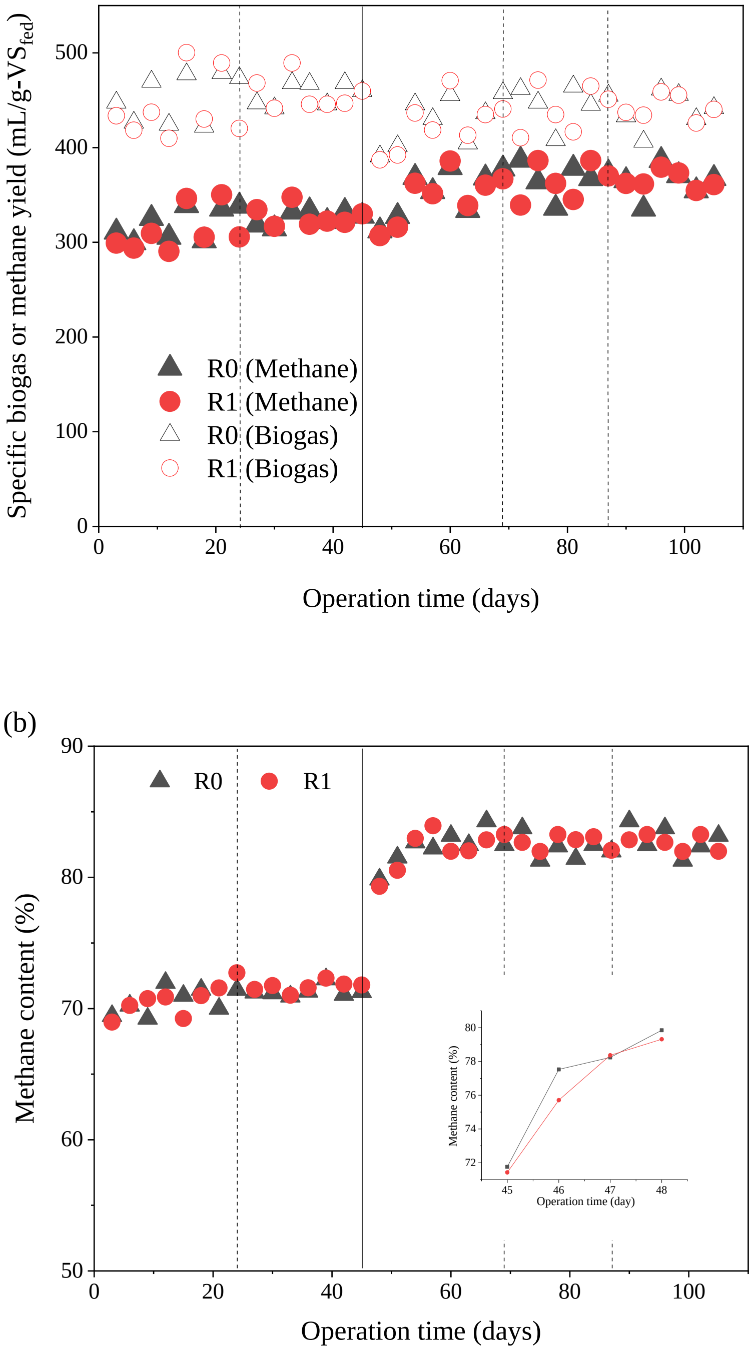
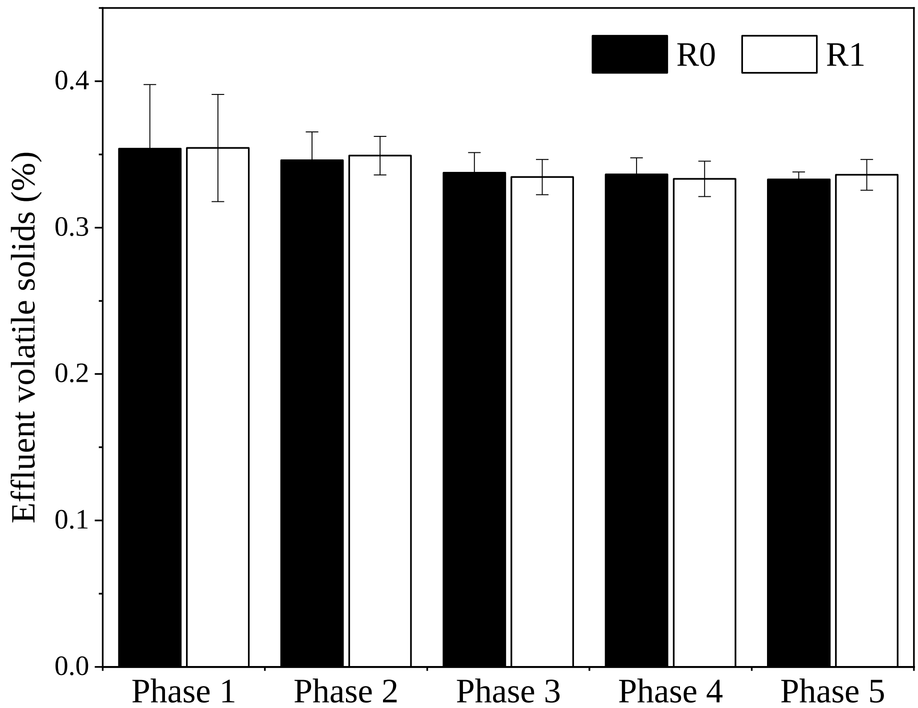


Fig. 4-6 Specific biogas or methane yield (a) and methane content (b) during semi-continuous anaerobic digestion with or without biogas recirculation and chemical addition. HRT, hydraulic retention time; Control, with no biogas recirculation and no chemical addition; +biogasR, with biogas recirculation; +Mg, with $MgCl_2$ addition; +Fe, with $FeCl_3$ addition.



R0	Control	Control	+biogasR	+biogasR	+Fe+biogasR
R1	Control	+Mg	+Mg+biogasR	+Mg+biogasR	+Mg+biogasR
HRT	15 days			12 days	

Fig. 4-7 Average volatile solids (VS) contents of effluent sludge from semi-continuous anaerobic digestion at different phases. HRT, hydraulic retention time; Control, with no biogas recirculation and no chemical addition; +biogasR, with biogas recirculation; +Mg, with $MgCl_2$ addition; +Fe, with $FeCl_3$ addition.

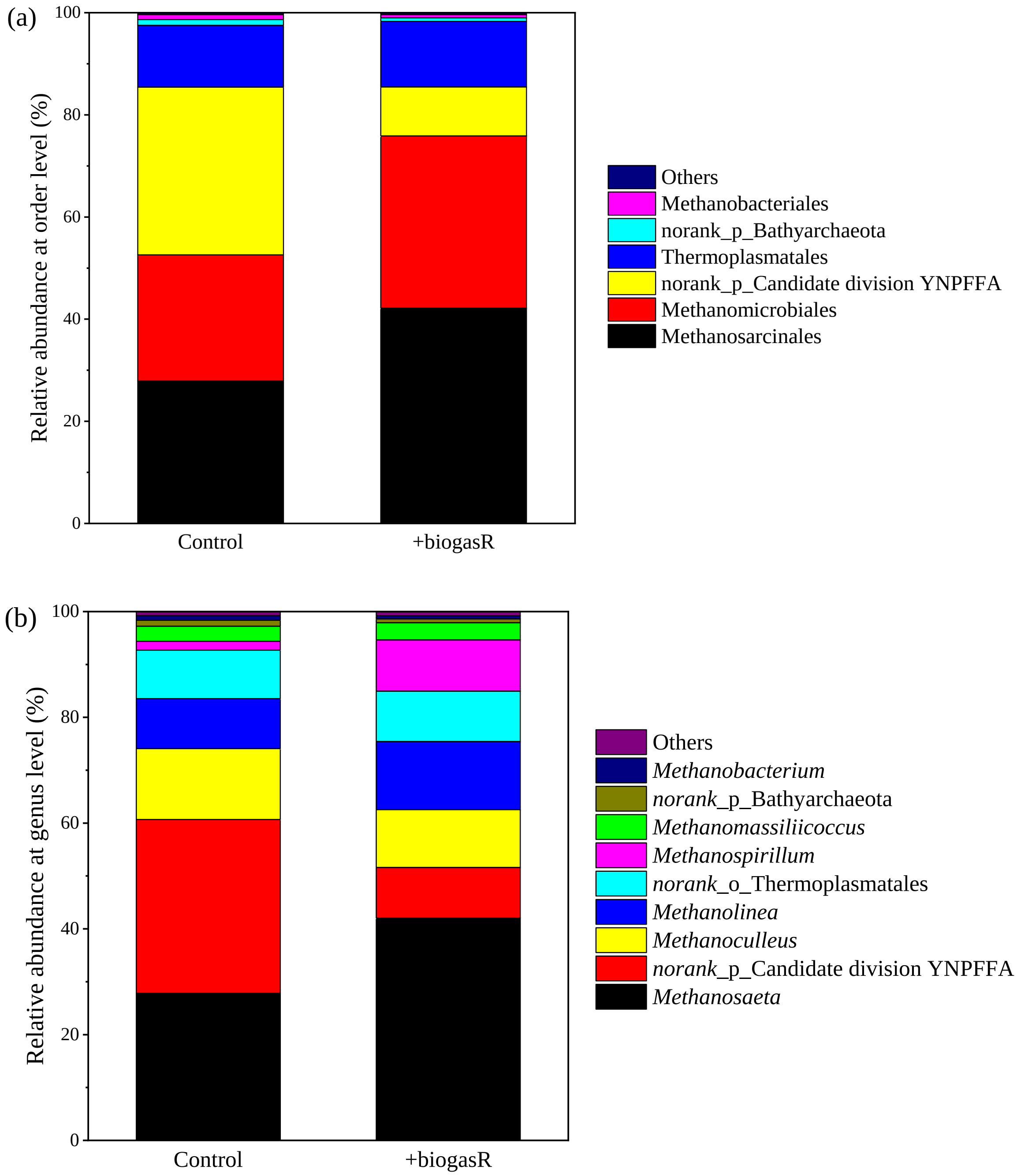
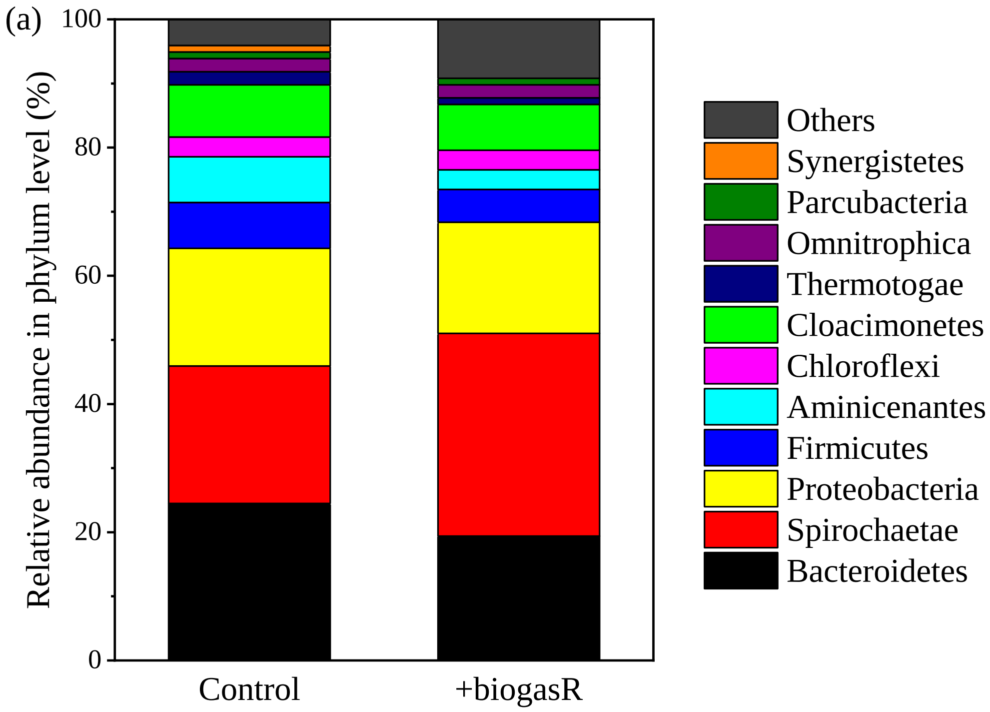


Fig. 4-8 Taxonomic composition of archaeal community at order (a) and genus (b) levels in R0 during phase 2 (day 45) without biogas recirculation (Control) and phase 3 (day 69) with biogas recirculation (+biogasR) of semi-continuous anaerobic digestion. Taxonomic compositions with relative abundance less than 0.05% was categorized as ‘Others’.



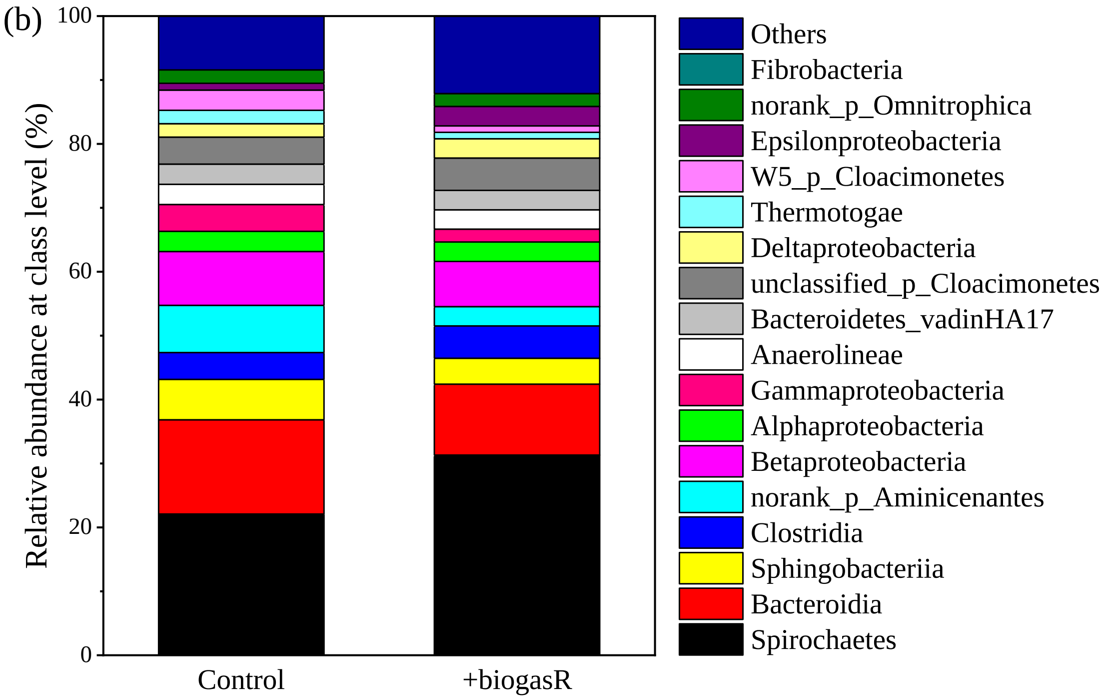


Fig. 4-9 Taxonomic composition of bacterial community at phylum (a) and class (b) levels in R0 during phase 2 (day 45) without biogas recirculation (Control) and phase 3 (day 69) with biogas recirculation (+biogasR) of semi-continuous anaerobic digestion. Taxonomic compositions with relative abundance less than 0.05% was categorized as 'Others'.

Chapter 5 Mechanisms and economic analysis

5.1 Mechanisms involved in the novel AD system

The novel AD system developed in this research realized biogas upgrading, P conservation and sludge conditioning simultaneously by coupling biogas recirculation and chemical addition. This chapter summarized the possible mechanisms involved in the novel AD system according to the results from chapters 2-4 (Fig. 5-1).

5.1.1 Biogas upgrading and AD performance

Biogas upgrading in the novel AD system was mainly realized by biogas recirculation. Water absorption is the mostly used method for biogas upgrading. Due to the higher solubility of CO_2 than CH_4 in water, the biogas recirculation in AD reactor may also result in dissolution of CO_2 in sludge, which contains greater than 98% of water. In chapter 2, the results of carbon profiles revealed that partial CO_2 in biogas was probably dissolved in sludge liquor or adsorbed on sludge particles (Fig. 2-3). In chapter 3, different concentrations of $FeCl_3$ were added in sludge during AD. The $FeCl_3$ dosage over 600 mg-Fe/L further enhanced the CH_4 content probably due to the low K_{sp} of $FeCO_3$. According to the SI of oversaturated CO_3^{2-} related precipitates in digestates estimated using Minteq v3.1 (Fig. 3-2), both biogas recirculation and increased dosages of $FeCl_3$ promoted the CO_3^{2-} precipitation, possibly contributing to the enhanced CH_4 content (Fig. 5-1). In chapter 4, the mechanism was further clarified through microbial analysis. The enhanced CH_4 content might also be associated with the promoted H_2/CO_2 pathway via biogas recirculation. The increased relative abundance of acetoclastic Methanosarcinales and hydrogenotrophic Methanomicrobiales in R0 during phase 3 with biogas recirculation contributed to both increased CH_4 yield and CH_4 content (Fig. 4-8).

During the semi-continuous AD in chapter 4, the methane yield was around 320-370 mL/g-VS_{fed}, which was significantly higher than the methane yield (170-180 mL/g-VS_{fed}) obtained during batch AD in chapters 2 and 3, probably due to the high biodegradability of the sludge used in semi-continuous test. By comparing the methane yield based on the reduced VS content, the methane yield was around 620-640 mL/g-$VS_{reduced}$ during the batch AD, while it was 530-580 mL/g-$VS_{reduced}$ during the semi-continuous AD, indicating the similar microbial activity in both batch and semi-continuous AD tests.

5.1.2 Phosphorus conservation

Two chemicals, including $MgCl_2$ and $FeCl_3$, were attempted in this thesis to realize P conservation and sludge dewaterability simultaneously during AD. Struvite precipitation by adding Mg sources in digested liquid are usually applied for simultaneous P and N recovery. Though most previous researches reported that the optimum pH range for struvite precipitation was 8.0-10.0, some of them verified that the optimal pH for the formation of pure struvite fell in the range of 7.0-7.5 but the precipitation rate was quite low (Hao *et al.*, 2008). In chapter 2, the direct addition of $MgCl_2$ during AD at a Mg:P_{ortho} molar ratio of 1:1 reduced soluble ortho-P by 78% (digestate pH=7.2), which further increased to 86% with biogas recirculation (digestate pH=7.9). The ortho-P concentration in digestate was reduced from 440 mg-P/L to 63 mg-P/L in the reactor with biogas recirculation and $MgCl_2$ addition. However, the $MgCl_2$ addition during the semi-continuous AD did not contribute much to ortho-P reduction. Only 28% reduction of ortho-P was observed when the HRT was 15 days with biogas recirculation (digestate pH=7.3). The ortho-P concentration in the digestate was reduced from 60 mg-P/L to 43 mg-P/L. As a result, struvite precipitation during AD of sewage sludge largely depends on the pH value and original ortho-P concentration in the substrates.

On the contrary, $FeCl_3$ showed better and stable performance on P conservation during both batch and semi-continuous AD. In chapter 3, the $FeCl_3$ addition at a Fe:P_{ortho} molar ratio of 1.5:1 reduced ortho-P by 96% (digestate pH=7.3), which further increased to 97% with biogas recirculation (digestate pH=7.8). The ratio of 1.5:1 depends on the Fe:P molar ratio of vivianite ($Fe_3(PO_4)_2$), which could be easily formed under reducing environment and neutral pH condition (Rothe *et al.*, 2016). The ortho-P concentration was reduced from 400 mg-P/L to 10 mg-P/L in the reactor with biogas recirculation and $FeCl_3$ addition. During the semi-continuous AD, the ortho-P was reduced by 94%, from 60 mg-P/L to 3.4 mg-P/L with $FeCl_3$ addition and biogas recirculation. The phenomenon may be resulted from both low K_{sp} of Fe-related precipitates and their strong binding capacities.

5.1.3 Sludge dewaterability

The effect of biogas recirculation on sludge dewaterability and settleability was investigated in chapter 2. Long operation duration of biogas recirculation during AD (HRT=30 days) deteriorated the sludge dewaterability significantly, possibly due to the high shear force brought about by the biogas recirculation. The results of EPS revealed that the proteins in TB-EPS was possibly transferred into the soluble phase under the long-term biogas recirculation (Fig. 2-10). Accordingly, a shorter HRT (12-15 days) was applied during the semi-continuous AD in chapter 4, where no deterioration of sludge dewaterability was observed. In addition, the results of EPS in chapter 2 also indicated that the added Mg^{2+} might bridge with the biopolymers in TB-EPS, contributing to the enhanced sludge dewaterability. Charge neutralization is one of the main mechanisms for enhanced sludge dewaterability by adding multivalent cations due to the negative surface charge of sludge flocs. In chapter 4, the effects of divalent Mg^{2+} and trivalent Fe^{3+} on sludge dewaterability were compared at the same equivalent concentrations. The results indicated the better performance of $FeCl_3$ on sludge dewaterability enhancement was probably associated with the complex Fe chemistry especially when organics were involved. Fe could not only tightly bind with organics, such as EPS excreted by microorganisms, but also form amorphous ferric oxides. The latter may have strong adsorption or binding capacities, which might contribute to the excellent performance on P conservation and sludge conditioning (Wilfert *et al.*, 2015).

5.2 Analysis on the industrial-scale application of the novel AD system

The sewage sludge used in this study was sampled from the Shimodate WWTP in Chikusei, Ibaraki, Japan. Taking this WWTP as an example, the expected results of the industrial-scale application of this novel AD system was further discussed.

From the operation data in 2017, the WWTP had an average wastewater treatment capacity of 8190 m^3/d (Table 5-1). Averagely about 38 m^3 of concentrated sludge including 16 m^3 primary sludge and 22 m^3 excess sludge, was daily treated by AD, and its daily biogas production was 433 m^3/d with an average methane content 60.2%. If all the biogas was used for electricity generation, approximately 2.48×10^5 kWh/y could be gained according to Eq. 5-1. The generated electricity could cover 17.3% of the total electricity consumption of the WWTP (1,431,948 kWh/y in 2017).

$$E_{biogas}(\text{kWh/y}) = (V_{CH_4} \times LHV_{CH_4} \times E_{ff})/(3.6 \times 100\%) \qquad \text{5-1}$$

where E_{biogas} (kWh/y) is the amount of electricity generated using the produced biogas per year; V_{CH4} (m^3/y) is the annual methane yield produced in the WWTP (for 365 days' operation); LHV_{CH4} (35.8 MJ/m^3) is the lower heating value of methane; E_{ff} (%) is the thermal efficiency of biogas engine (given the assumption that the same spark ignition engine was used as Porpatham *et al.* (2008)); 3.6 (MJ/kWh) is the unit conversion from MJ to kWh.

In this study, the methane content in biogas was increased by 11% through biogas recirculation during both batch and semi-continuous AD. According to Porpatham *et al.* (2008), the E_{ff} of biogas engine could be improved from 26.2% to 27.1% if the CO_2 content in biogas was reduced from 41% to 30% (at a constant value of total methane amount). Thus, through applying biogas recirculation in the anaerobic digesters of the Shimodate WWTP, the E_{biogas} would increase to 2.56×10^5 kWh/y, which could cover 17.9% of the total electricity consumption of the WWTP (Table 5-2). If further considering the 13% increase of methane yield during semi-continuous AD, the E_{biogas} would increase to 2.9×10^5 kWh/y, which could cover 20.2% of total electricity consumption of the WWTP (Table 5-2). The increased amount of electricity (8,000~42,000 kWh) by using the novel AD system corresponds to $3.1\sim6.4\times10^5$ JPY (about 2820~5820 USD) according to the feed-in tariff (FIT) purchase price of electricity generated from biogas (39 JPY/kWh, about 0.35 USD/kWh). According to MLIT (2018), the annual biogas production from sewage sludge in Japan is 3.3×10^8 m^3/y, in which 37% is utilized for electricity generation. The 11% increase of CH_4 content in the biogas by applying the novel AD system, the E_{biogas} in Japan could be increased by 6.56×10^6 kWh/y, corresponding to 2.56×10^8 JPY (about 2.3 million USD).

In addition, the addition of $MgCl_2$ or $FeCl_3$ during AD could increase the phosphorus content in the solid phase of digested sludge and the sludge dewaterability. Phosphorus is an essential element for crop growth, and regarding phosphorus sources Japan totally relies on imports from other countries. Nowadays, along with the increased phosphorus price which has become a serious issue, phosphorus recovery from wastewater has been strongly prompted by the Government of Japan (MLIT, 2018). Results from this study show that the phosphorus contents in the solid phase of digested sludge was increased by 82-108%. According to the data published by MLIT in 2010, the phosphorus content in the sewage sludge disposed by composting was around 4400 t/y (MLIT, 2010b). The application of the novel AD system in this study would contribute to an annual increase of 3600-4750 t of P, which could save at least $6.6\sim8.7\times10^7$ JPY (about 0.6-0.8 million USD) for importing phosphorus rock from other countries (MLIT, 2010b). Restated, the above two value-added aspects, i.e. enhanced methane content in the biogas and phosphorus content in the solid digestate, can be greatly recognized if this novel AD system is popularized in all the WWTPs nationwide.

Furthermore, by adding $MgCl_2$ or $FeCl_3$ during AD with biogas recirculation in this study, the sludge dewaterability was enhanced by 37%~94%. The reduced cost related to sludge dewatering process may also be expected. An enhanced dewaterability of sludge can contribute to the followings: 1) reduction of electricity consumption during dewatering process due to a shorter duration for dewatering (Chu *et al.*, 2005); 2) reduction of energy consumption for thermal drying because of the increased solids content in the dewatered sludge (Wang *et al.*, 2010); and 3) decrease of the addition of chemical flocculants. Eventually, all the above aspects could contribute to the reduced operation costs of the WWTP. Currently, $FeCl_3$ and lime are used as coagulants for sludge conditioning in this WWTP. More in-depth research works are demanding in order to confirm whether they are still necessary and their optimum dosages for conditioning the digested sludge from the novel AD system in the context of the whole WWTP. When this novel AD is introduced into the WWTP, the whole AD system and digested sludge conditioning can be simplified as shown in Fig. 5-2. Further cost reduction in operation and management is expected regarding the dewatering and disposal of the digested sludge from the WWTP.

Table 5-1 Operation data of Shimodate WWTP, Chikusei, Ibaraki in 2017.

Parameters	Units	Data
Average wastewater treatment capacity	m^3/d	8,190
Influent amount of AD tank	m^3/d	38
Effluent amount of AD tank	m^3/d	32
Average biogas yield	m^3/d	433
Average methane content	%	60.2
Hydraulic retention time	Days	30-40
Amount of sludge cake	t/d	2.3
Total electricity consumption	kWh/y	1,431,948

Table 5-2 The expected results of the industrial-scale application of the novel anaerobic digestion system calculated using Eq. 5-1.

CH_4 content (%)	E_{ff} (%)	V_{CH4} (%)	E_{biogas} (kWh/y)	Coverage of total electricity consumption in WWTP (%)
60	26.2	95143	2.48×10^5	17.3
71	27.1	95143	2.56×10^5	17.9
71	27.1	107521	2.9×10^5	20.2

E_{ff}, thermal efficiency of biogas engine; V_{CH4}, annual methane yield produced in Shimodate WWTP; E_{biogas}, the amount of electricity generated using the produced biogas per year.

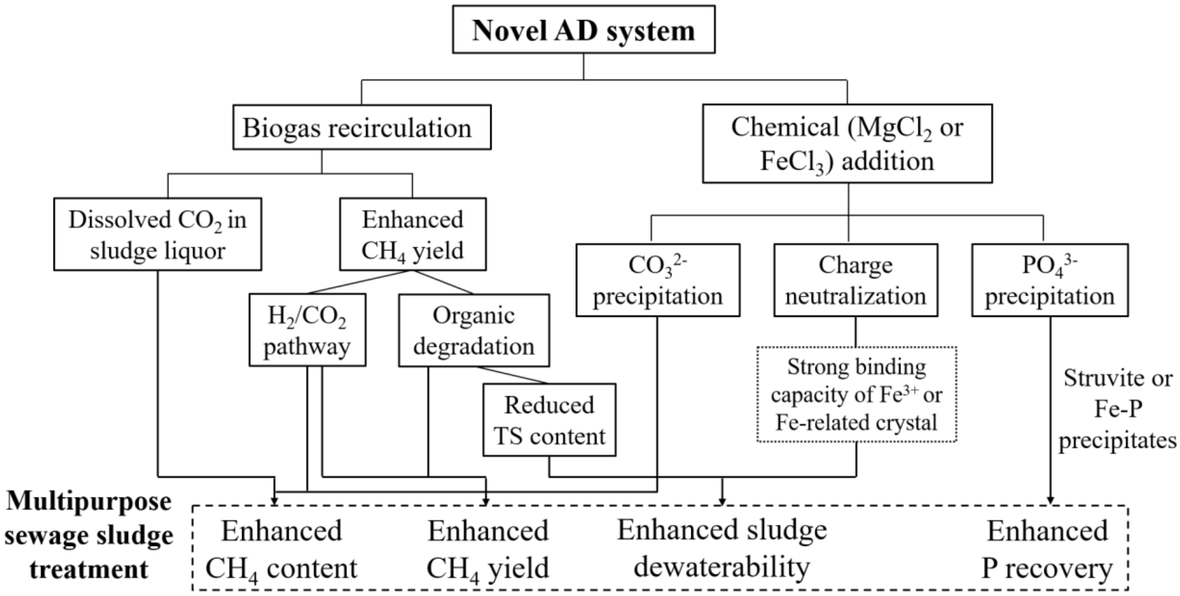


Fig. 5-1 Possible mechanisms involved in the novel anaerobic digestion (AD) system with biogas recirculation and chemical addition.

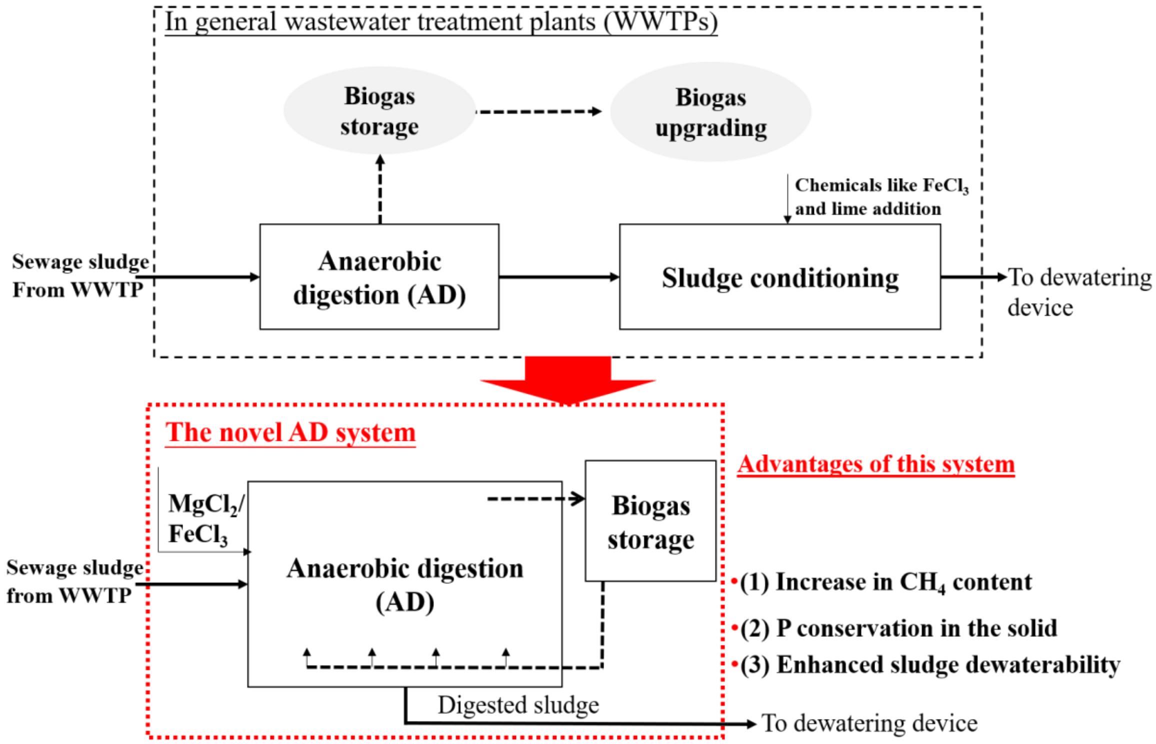


Fig. 5-2 Comparison between the sludge treatment processes in general wastewater treatment plants (WWTPs) and the novel anaerobic digestion (AD) system developed in this thesis.

Chapter 6 Conclusions and future research

6.1 Conclusions

In this thesis, a novel AD system that can realize simultaneous biogas upgrading, resources conservation and sludge conditioning was established. The main results regarding biogas production, methane content, resources conservation and sludge dewaterability during batch and semi-continuous operation of the novel AD system were summarized as follows (Table 6-1).

6.1.1 Comparison between batch mode and semi-continuous mode operation

(1) Intermittent biogas recirculation enhanced methane content in biogas by 11% during both batch and semi-continuous AD, mainly due to the dissolution of CO_2 in sludge liquor, possible formation of carbonate related precipitates and the enriched hydrogenotrophic methanogens during semi-continuous operation. Whereas, only methane yield was enhanced by 13% during the semi-continuous AD, possibly due to the longer acclimation duration of microbes to the new environment in AD with biogas recirculation.

(2) $MgCl_2$ addition during batch AD of primary sludge reduced ortho-P by 87%; however, it did not reduce ortho-P during semi-continuous AD of excess sludge, mainly due to the neutral pH (7.0) and relatively low ortho-P concentration (~60 mg/L) in excess sludge.

(3) $FeCl_3$ addition during both batch and semi-continuous AD with biogas recirculation showed better performance than $MgCl_2$ on sludge dewaterability enhancement and P conservation in solid, probably resulted from its strong flocculation and binding capacities.

Semi-continuous operation is more similar to the real application in industries compared with batch mode and is easier to be handled and more stable in laboratory scale compared with continuous mode. Semi-continuous mode is recommended for further investigation of the novel AD system.

6.1.2 Comparison between $MgCl_2$ and $FeCl_3$ addition

(1) When using primary sludge containing 900 mg/L TP and 1200 mg/L TN as the

substrate of AD, $MgCl_2$ addition at a Mg:P_{ortho} molar ratio of 1:1 conserved 93% of P and 69% of N in the solid phase of digestate. Around 120 mg/g-TS struvite was estimated to be formed in the digestate. $FeCl_3$ addition at a Fe:P_{ortho} molar ratio of 1.5:1 reserved 98.7% of P in the solid phase of digestate, which contains 59% of N.

(2) When using excess sludge containing 130 mg/L TP and 395 mg/L TN as the substrate of AD, $MgCl_2$ addition at a Mg:P_{ortho} molar ratio of 2:1 only contributed to 68% of P conservation in solid phase of digestate. $FeCl_3$ addition at a Fe:P_{ortho} molar ratio of 1.5:1 contributed to 97.4% of P conservation in solid phase in form of vivianite-like crystal. 57% of N remained in solid phase of digestate after semi-continuous AD.

(3) $MgCl_2$ addition of 23 mg-Mg/g-TS enhanced sludge dewaterability of digestate by 37% during batch AD of primary sludge with biogas recirculation; however, the sludge dewaterability became worse at longer operation time (30 days). $FeCl_3$ addition of 68 mg-Fe/g-TS enhanced sludge dewaterability of digestate by 79% during batch AD of primary sludge with biogas recirculation.

(4) During semi-continuous AD of excess sludge, $FeCl_3$ addition of 19 mg-Fe/g-TS enhanced sludge dewaterability of digestate by 94%, while $MgCl_2$ addition of 12 mg-Mg/g-TS enhanced sludge dewaterability by 75%.

$FeCl_3$ addition showed better performance on both sludge dewaterability enhancement and P conservation in solid compared with $MgCl_2$ addition during batch AD of primary sludge and semi-continuous AD of excess sludge, due to its strong binding capacity and the very low solubility product (K_{sp}) of Fe-related precipitates that possibly formed during AD.

6.1.3 Perspectives for WWTPs

From above, the application of intermittent biogas recirculation during AD of sewage sludge in WWTPs has the possibility to enhance both methane yield and methane content, and a shorter hydraulic retention time (HRT) is recommended from both aspects of sludge dewaterability and biogas upgrading. Further with $FeCl_3$ addition in the novel AD system, the digestate with improved sludge dewaterability and higher P content in solid part could be obtained. The targets of this thesis are consistent well with the sustainable development goals (SDGs), including affordable and clean energy, sustainable consumption and production patterns and climate action. Through the novel AD system, the biogas with a higher heating value can be produced, thus contributing to less energy consumption or operation cost in WWTPs if the biogas was used for heating or electricity generation (SDGs 7: Energy).

Moreover, the less CO_2 production can result in less GHGs emission (SDGs 13: Climate change). The increased P conservation in solid phase of digestate as vivianite-like crystal can increase the fertilizer potential of the solid digestate (SDGs 12: Sustainable consumption and production). Correspondingly, the lower ortho-P concentration in the liquid phase could ease its post-treatment (SDGs 6: Water and sanitation). The application of the novel AD system could simplify the facilities in WWTPs for sewage sludge treatment and largely reduce both total capital cost and operation cost in WWTPs (SDGs 8: Economic growth).

6.2 Future research

Though the developed novel AD system in this thesis realized simultaneous biogas upgrading, P conservation and sludge conditioning, further research is still needed to perfect the novel AD system and promote its application in practice.

(1) The enhancement efficiency on methane content in biogas should be further improved. The correlation between operation strategy of biogas recirculation and sludge properties (TS, viscosity, etc.) should be further clarified.

(2) The fertilizer value and safety of the dewatered sludge should be further studied from agricultural aspect through seed germination test and plant growth assay.

(3) Dewatering efficiency of the digestate should be confirmed in practice and the comparison on chemical consumption in novel AD system and conventional sewage sludge treatment should be conducted.

(4) The post-processing method for the dewatered sludge from the novel AD system with $FeCl_3$ addition should be explored to further reduce the waste volume and produce value-added materials, such as biochar or hydrochar for nutrient adsorption, soil amendment and so on.

(5) Other form of Fe-related chemicals or other materials that can enhance sludge dewaterability and P conservation should be attempted in the novel AD system in order to find out the most cost-effective one.

Table 6-1 Summary of the results about the effects of biogas recirculation and chemical addition on batch and semi-continuous anaerobic digestion (AD) in this research.

Operation mode	Chemical addition	CH_4 content	CH_4 yield	Nutrients conservation in solid	Sludge dewaterability enhancement
Batch AD	$MgCl_2$	11% enhanced		93% P, 69% N	37%
	$FeCl_3$			99% P, 59% N	79%
Semi-continuous AD	$MgCl_2$		13% enhanced	68% P, 57% N	75%
	$FeCl_3$			97% P, 57% N	94%

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