Rev Environ Sci Biotechnol (2019) 18:771–793
https://doi.org/10.1007/s11157-019-09515-y

REVIEW PAPER

# Biogas production from anaerobic co-digestion of waste activated sludge: co-substrates and influencing parameters

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Published online: 8 October 2019


**Abstract** Anaerobic digestion is a versatile biotechnology to treat waste activated sludge (WAS), the main by-products of biological wastewater treatment, because it can achieve simultaneously energy recovery (biogas) and pollutant reduction (organic matter, pathogens). However, the potential of biogas production from mono-digestion of WAS is usually limited by the imbalance carbon to nitrogen (C/N) ratio of WAS and ammonia accumulation. Anaerobic co-digestion, simultaneous digestion of two or more substrates, should be a feasible option to resolve these disadvantages. The abundant organic wastes from municipal, industrial, and agricultural field have been the ideal co-substrates because they not only can balance the substrate nutrient to obtain the optimal C/N ratio, but also can adjust pH and dilute the toxic materials to mitigate the inhibition to methanogens, consequently improving the yield of biogas, especially methane. This paper classified the main organic co-substrates according to their source and reviewed their application in anaerobic co-digestion of WAS. Then the influence of temperature, pH, organic loading rate, hydraulic retention time, C/N ratio, digester type and pretreatment method on biogas production was extensively discussed. Finally, this review brought forward

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**Electronic supplementary material** The online version of this article (https://doi.org/10.1007/s11157-019-09515-y) contains supplementary material, which is available to authorized users.

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the challenges and outlooks of anaerobic co-digestion in the future.

**Keywords** Anaerobic co-digestion · Biogas · Co-substrates · Influencing parameters · Waste activated sludge (WAS)

## 1 Introduction

The rapid development of industrialization and urbanization brings the soaring municipal wastewater production. Simultaneously, the disposal of waste activated sludge (WAS), main by-products from wastewater treatment plants (WWTPs) also grows up to be a serious environmental issue. For instance, the average annual production of WAS has been over 240 million wet tons in Europe, USA and China (Wang et al. 2017a). Especially in China, the value had reached nearly 7 million tons (dry weight) in 2014 accompanying with an annual growth rate of over 13% (Yang et al. 2015). Land-filling and incineration are the main ways to treat WAS nowadays, but they all do not conform to the concept of circular economy (Morris 2005). Energy consumption and acute air pollution control during the waste incineration require huge investments. The health risks from groundwater pollution, land shortage, greenhouse gas emission limit the application of land-filling (Zhang et al. 2012).

Anaerobic digestion is a biological process by which special microorganisms break down biodegradable material in the absence of oxygen, simultaneously, a versatile renewable fuel, biogas (methane ($CH_4$) or hydrogen ($H_2$)) is recovered (Thangamani et al. 2010; Tyagi and Lo 2013). Much environmental and economic welfare can be acquired from anaerobic digestion of WAS, such as sludge volume reduction, sludge stabilization, nutrient (nitrogen and phosphorus) recycling, and biogas energy production. As showed in Fig. 1a, traditional anaerobic digestion has been a hot topic in the treatment of WAS. However, the mono-digestion of WAS is often constrained by the imbalanced carbon to nitrogen (C/N) ratio, low biogas yield, unfavorable volatile solid (VS) removal,

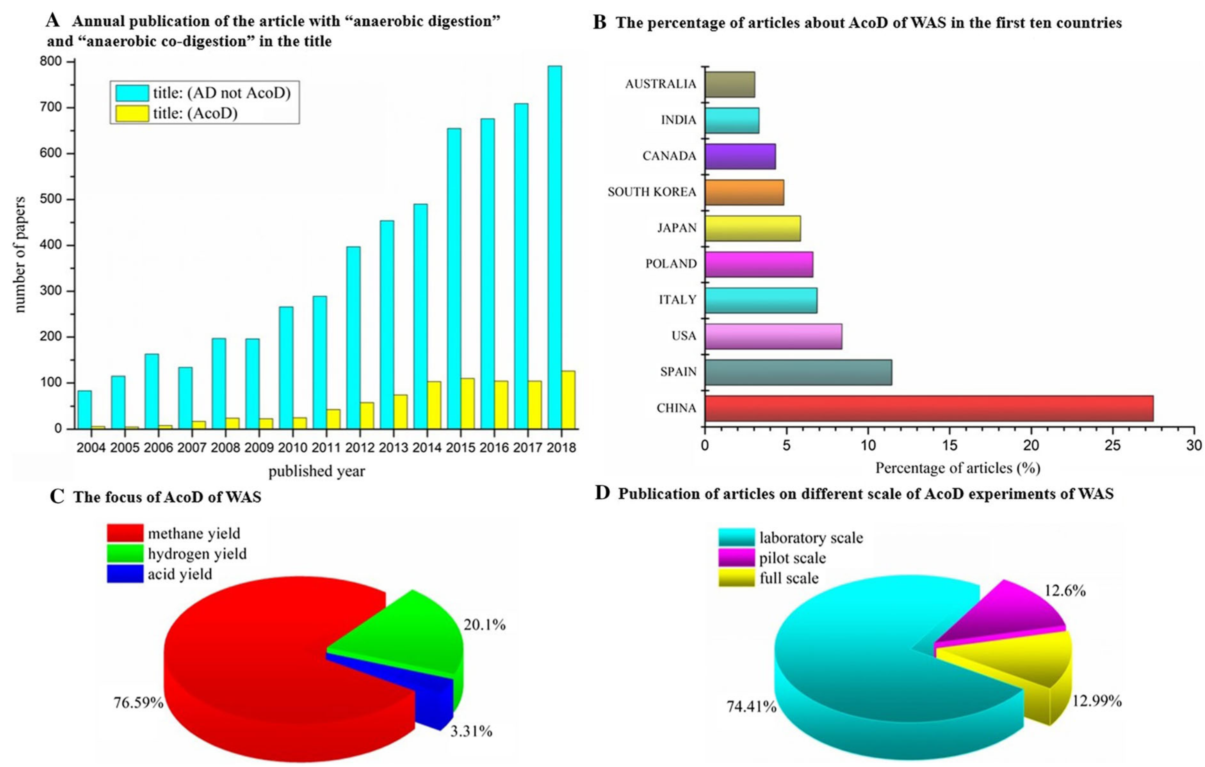


**Fig. 1** The research situation of anaerobic co-digestion. Data source: web of science, 1 July 2019; "AD" means anaerobic digestion, "AcoD" means anaerobic co-digestion

and ammonia accumulation (Creamer et al. 2010). Usually the C/N ratio of sewage WAS is around 10 because it mainly consists of microbial cells, leading to high nitrogen content, but the suitable ratio for anaerobic digestion of organic matter is around 20–30 (Parkin and Owen 1986). The low C/N results in the imbalanced diet for anaerobic organisms (Feng et al. 2009; Luo et al. 2019). Meanwhile, high organic loading rate (OLR) under low C/N ratio also causes the ammonia accumulation, which has inhibitory effects on anaerobic organisms' activity (Dai et al. 2017). A recent and notable trend in the development of anaerobic digestion is anaerobic co-digestion and the proportion of publications on co-digestion is increasing year by year (Fig. 1a, b). Anaerobic co-digestion refers to the simultaneous digestion of two or more substrates in one unit. In this process, each substrate can exhibit the respective properties (Lee et al. 2006). It is well-known that anaerobic digestion of organics usually divides into single-phase/stage and two-phase/stage. Among them, the two-phase/stage one displays better performance and explains the conversion pathway more clearly (Wang et al. 2017b). Figure 2 illustrates the conversion pathway of organic substrates in a typical two-stage anaerobic co-digestion system. Co-digestion can significantly improve biogas production and realize higher pecuniary benefits in WWTPs (Hagos et al. 2017). For example, the biogas production from co-digestion of organic fraction of municipal solid wastes (MSW) and WAS was up to 500 mL-biogas/g-$VS_{added}$, which was significantly higher than that from mono-digestion of WAS (120–150 mL-biogas/g-$VS_{added}$) (Zupancic et al. 2008; Athanasoulia et al. 2012a). Therefore, co-digestion of WAS with other organic substrates including municipal, industrial, and agricultural wastes seems to be a better solution to solve the limitation of mono-digestion. Data from web of science (Fig. 1c, d) show that the main product of WAS co-digestion are $CH_4$. Meanwhile, in some researches, $H_2$ and volatile fatty acids (VFAs) also serve as the fermentation products by inhibiting the methanogenesis. And most of anaerobic co-digestion are investigated in laboratory scale.

There are many advantages in anaerobic digestion of WAS with other organic wastes as the co-substrates, such as (1) enhancing biogas production ($CH_4$ or $H_2$) compared to the mono-digestion of WAS (Alemahdi et al. 2015; Dai et al. 2016a); (2) higher contaminant removal rates (Agdag and Sponza 2007); (3) increas-

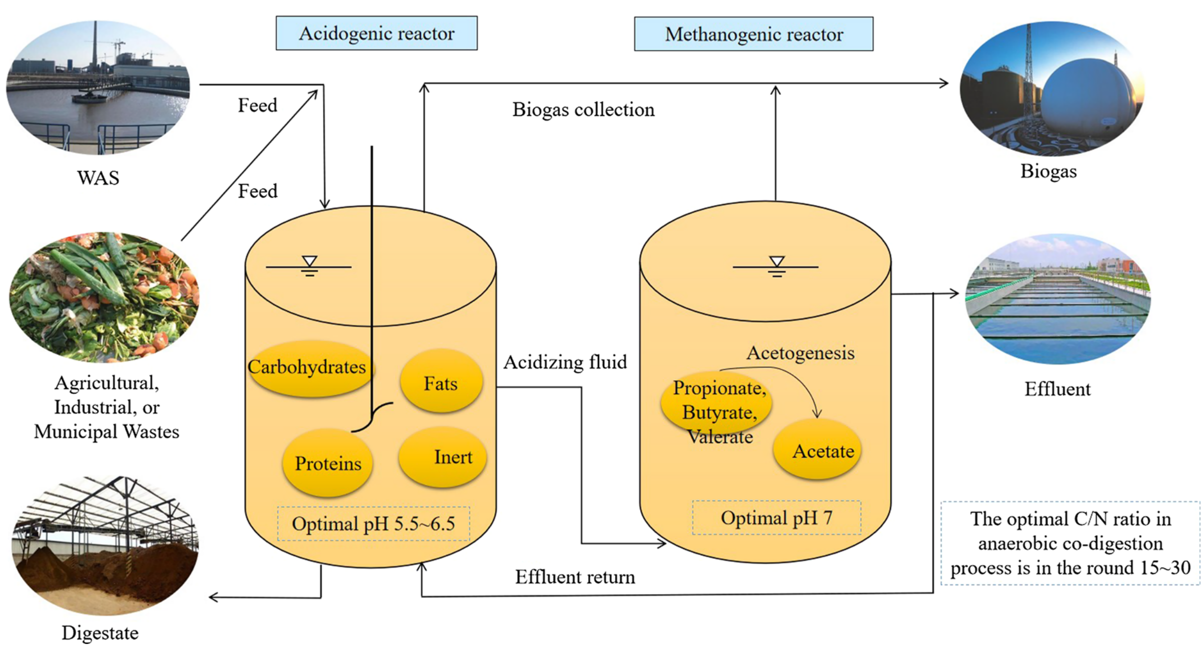


**Fig. 2** The conversion pathway of organic co-substrates in a typical two-stage anaerobic co-digestion system

ing the nitrogen and phosphorus loading rates (Caffaz et al. 2008) and improving the stability because co-substrates act as a diluting agent to attenuate the effect of toxic or inhibitory chemical compounds like ammonia or $Na^+$ (Dai et al. 2013; Sole-Bundo et al. 2017); (4) improving sludge dewaterability (Wang et al. 2013a); (5) increasing buffer capacity (Zhu et al. 2008); (6) accelerating initial biogas production time (Ji et al. 2013); (7) improving digestion product quality for agricultural use, such as lower number of pathogen, high N concentration, and not available metals state (Pecharaply et al. 2007; Sarikaya and Demirer 2013); (8) balancing C/N ratio (Wan et al. 2011; Sarikaya and Demirer 2013); (9) reducing construction and operation costs (i.e. low-cost pH control) (Zhu et al. 2008; Hosseini Koupaie et al. 2014).

Agricultural, industrial and municipal organic wastes are the most commonly used co-substrates in anaerobic digestion of WAS. Figure 3 shows the source of these main organic materials. Many cases are also applied in full-scale WWTPs successfully (Bolzonella et al. 2006; Zupancic et al. 2008). In spite of these advantages and successful experiences, anaerobic co-digestion of WAS still faces many challenges and problems. Anaerobic co-digestion is a very complicated and sensitive process involving numerous microorganisms (Hagen et al. 2014). The origins of co-substrates, environmental parameters and operational conditions have significant influences on the diversity of functional organism and performance of biogas production. Although better performances of sludge reduction and contaminant removal are maintained, toxicity led by heavy metal, ammonia or acid accumulation are regularly observed in various sludge anaerobic co-digestion cases (Agdag and Sponza 2007). Meanwhile, some specific co-substrates, for instance, the lignin-rich co-substrate from pulp and paper mill can affect the performance of anaerobic co-digestion owing to the unsuitable pH value and the nitrogen deficiency (Bayr and Rintala 2012).

This paper aims to (1) classify the main organic co-substrates based on their source (municipal, industrial and agricultural) and review their applications in anaerobic co-digestion of WAS; (2) evaluate the effects of operational parameters such as temperature, pH, organic loading rate (OLR), hydraulic retention times (HRT), C/N ratio, digester type and pretreatments on anaerobic co-digestion of WAS in terms of

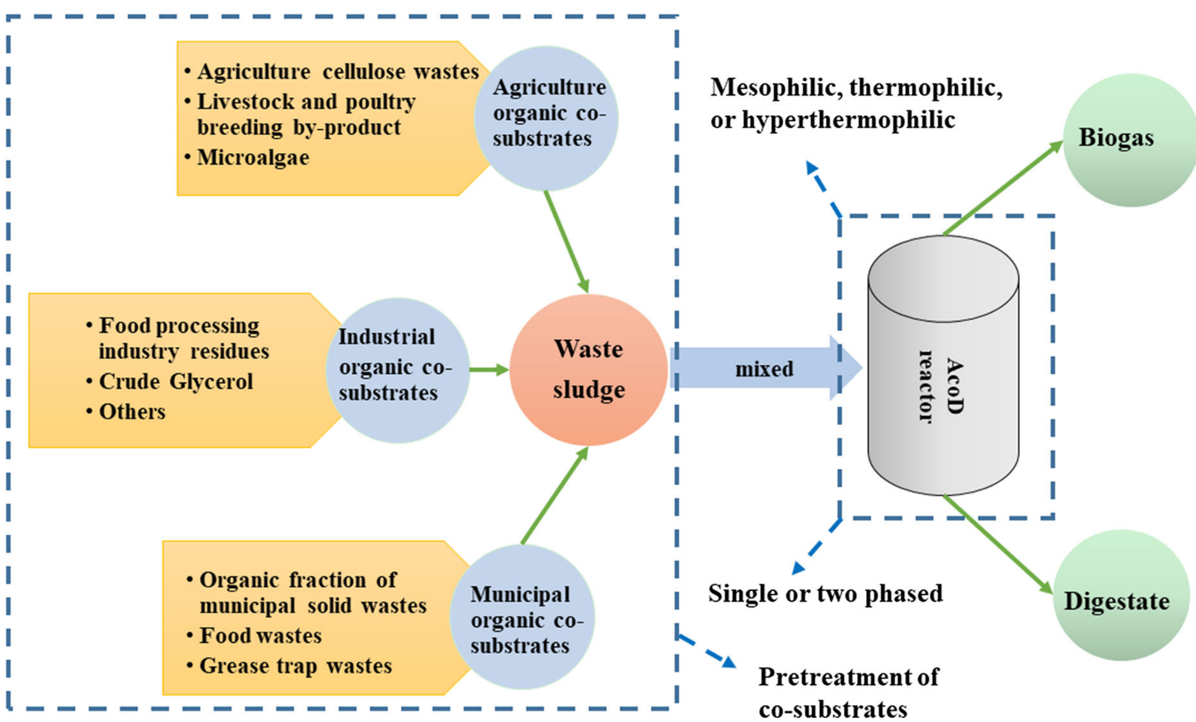


**Fig. 3** Sources of main organic materials in anaerobic co-digestion of WAS

biogas production, microbial change and system stability; (3) propose the challenging and perspective in anaerobic co-digestion of WAS.

## 2 Co-substrate in anaerobic co-digestion of WAS

### 2.1 Agricultural organic co-substrates

Organic material from agricultural sources such as cellulose crops (e.g. grass and straw), energy crops (e.g. algae), livestock manure (e.g. cattle manure, chicken manure, swine manure) are widely used as co-substrates in anaerobic digestion of WAS. Main operational parameters and biogas yield in anaerobic co-digestion of WAS with typical agricultural co-substrates are summarized in Table 1. Among them, manure and microalgae exhibit relatively better performance to achieve higher biogas yield.

#### *2.1.1 Agricultural cellulose wastes*

Agricultural cellulose residues are generated in large masses around the world, which include shredded grass, crops straw, coffee grounds and so on. Cellulose, hemi-cellulose and lignin are core components of the agricultural cellulose residues. The co-digestion of cellulose (contains relatively high carbon content) with WAS (contains relatively high nitrogen content) can overcome the deficiency of imbalanced C/N ratio in mono-digestion of WAS. In co-digestion of WAS, the addition of typical cellulose waste like grass could improve the C/N ratio, methane production, dewaterability, but did not significantly affect the ammonia concentration (Hidaka et al. 2013, 2016). That may be because WAS as a diluting agent could alleviate the change of ammonia concentration, meanwhile the cellulosic materials (i.e. grass) absorbed the ammonia in some extent (Hokkanen et al. 2016).

The insoluble and heterogeneous characteristic affects the degradation of cellulose residue. Therefore some enhanced treatments were implemented to improve the co-digestion progress. (1) A relatively higher initial pH can promote the final accumulate methane production due to the accelerated dissolution of organic matter. When WAS digested with perennial ryegrass, the proved optimum condition is C/N of 17 and initial pH 12, and the pH finally dropped to neutral level. Meanwhile, more $CH_4$ could be produced from the interspecies hydrogen/formate transfer pathway (Dai et al. 2016a). (2) The shredding or crushing of agricultural cellulose wastes can improve the biogas production from anaerobic co-digestion (Qiao et al. 2015). When adding different shredded length of grass to WAS, the biogas yield of 0–10 mm grass (330 mL-biogas/g-$VS_{added\text{-}grass}$) was slightly higher than that of 0–20 mm grass (310 mL-biogas/g-$VS_{added\text{-}grass}$) (Hidaka et al. 2013; Wang et al. 2014b). That may because that: on the one hand, the longer length grass is not suitable for fully mixing. One the other hand, length of the grass affects the particle size and the smaller particles possess a higher specific surface area. When these small grasses were used as the co-substrates with WAS digestion, an initial rapid rate of hydrolysis and higher methane production were obtained because the small grasses with higher specific area were more accessible to the bacteria (Tedesco et al. 2014). Interestingly, although bamboo was shredded into smaller length less than 10 mm, the corresponding biogas production was only 190 mL-biogas/g-$VS_{added\text{-}bamboo}$, which was far below that of grass (Hidaka et al. 2013). That suggests that the type of substrates gives more significant impact than their size. (3) Keeping suitable moisture content is important to maintain stable anaerobic co-digestion of WAS with farm cellulose residues. 70–80% moisture content in thermophilic co-digestion system of sewage sludge and rice straw led to a higher degradation efficiency of organic matter and specific biogas production (Chu et al. 2015). However, when moisture content decreased to below 65%, instability of anaerobic co-digestion process and low specific biogas yield was observed, which may attribute to the limited mass diffusion (Bollon et al. 2011). (4) pH adjustment, nutrient elements supplementation and increase in sludge proportion could make the unstable digester recovery. Qiao et al. (2015) reported that the VFA accumulation ($> 4000$ mg/L) caused the sharp decrease of biogas production, however, subsequent supplementation of $NH_4HCO_3$ and trace metal ($Mg^{2+}$, $Ca^{2+}$, $Mn^{2+}$, $Co^{2+}$, etc.) re-established a stable co-digester. In another case, Li et al. (2015b) proved that sulfate addition was an effective way to enhance methane production in thermophilic anaerobic co-digestion of coffee grounds, milk and WAS. Because the propionate-utilizing bacteria well adapted to the sulfate containing condition, the propionic acid degradation was effectively promoted and a highest OLR

**Table 1** Main operation parameters and effects in anaerobic co-digestion of WAS with typical agricultural, industrial and municipal co-substrates

| Co-substrates | Optimal mixed ratio[a] | Running condition | OLR | Biogas yield[b] | References |
|---|---|---|---|---|---|
| *Agricultural organic co-substrates* | | | | | |
| Swine manure | 30:70 (w/w) | Semi-continuous mesophilic | 1.91 g VS/L/d | 402 mL biogas/g $VS_{added}$ | Borowski et al. (2014) |
| | 50:50 (w/w, dry) | Semi-continuous thermophilic | 4.68 g VS/day/L | 470 mL $CH_4$/g $VS_{added}$ | Creamer et al. (2010) |
| Shredded grass | 1:10 (TS) | Continuous mesophilic | – | 90 mL $CH_4$/g $VS_{grass}$ | Hidaka et al. (2013) |
| | 1:4 (VS) | Continuous thermophilic | – | 343 mL $CH_4$/g $VS_{added}$ | Wang et al. (2014b) |
| Wheat straw | 19:11 (VS) | Batch mesophilic | 7.50 g VS/L | 345.50 mL $CH_4$/g $VS_{added}$ | Elsayed et al. (2016) |
| Coffee grounds | 15:85 (w/w, dry) | CSTR thermophilic | 7.54 g COD/L/d | 279 mL $CH_4$/g dry matter/d | Qiao et al. (2015) |
| Microalgae | 25:75 (VS) | Batch mesophilic | – | 442 mL $CH_4$/g $VS_{added}$ | Beltran et al. (2016) |
| *Industrial organic co-substrates* | | | | | |
| Crude glycerine | 2:98 (v/v) | Continuous mesophilic | 3.68 g VS/L/d | 483 mL $CH_4$/g $VS_{added}$ | Jensen et al. (2014) |
| | 1:99 (v/v) | Semi-CSTR mesophilic | 1.01 g VS/L/d | 1480 mL$CH_4$/g $VS_{added}$ | Rivero et al. (2014) |
| | 1.2:98.8 (v/v) | CSTR mesophilic | 1.2 g VS/L/d | 325 mL $CH_4$/ g $VS_{added}$ | Silvestre et al. (2015) |
| Cheese whey | 5:95 (v/v) | Batch mesophilic | – | 301.2 mL $CH_4$/g $VS_{added}$ | Fernández et al. (2014) |
| | 5:95 (v/v) | Batch thermophilic | – | 250.6 mL $CH_4$/g $VS_{added}$ | Fernández et al. (2014) |
| Olive mill wastewater | 5:95 (v/v) | Continuous mesophilic | 0.9 g VS/L/d | 222 mL biogas/g $VS_{added}$* | Maragkaki et al. (2017) |
| Brewery sludge | 75:25 (w/w) | CSTR mesophilic | – | 650 mL biogas/g $VS_{removed}$ | Pecharaply et al. (2007) |
| Meat processing sludge | 46:54 (VS) | CSTR mesophilic | 3.46 g VS/L/d | 463 mL $CH_4$/g $VS_{added}$ | Luostarinen et al. (2009) |
| Coffee processing waste | 7:1 (TS) | Batch mesophilic | – | 280 mL $CH_4$/g $VS_{added}$ | (Neves et al. 2006) |
| *Municipal organic co-substrates* | | | | | |
| Fat, oil and grease | 64:36 (VS) | Semi-continuous mesophilic | 2.34 g VS/L/d | 598 mL $CH_4$/g $VS_{added}$ | Wan et al. (2011) |
| | 1 g/50 mL | Semi-continuous two-stage thermophilic | 1.83 g VS/L/d | 681 mL $CH_4$/g $VS_{added}$* | Li et al. (2015a) |
| Grease trap waste | 23:77 (VS) | CSTR mesophilic | 3.0 g COD/L/d | 369 ml $CH_4$/g $VS_{added}$ | Silvestre et al. (2011) |
| | 27:73 (COD) | CSTR thermophilic | 2.8 g COD/L/d | 143 mL $CH_4$/g $COD_{added}$* | Silvestre et al. (2014) |
| Food wastewater | 75:25 (v/v) | Semi-continuous thermophilic | 6.88 g COD/L/d | 316 mL $CH_4$/g $COD_{removed}$ | Jang et al. (2015) |

**Table 1** continued

| Co-substrates | Optimal mixed ratio[a] | Running condition | OLR | Biogas yield[b] | References |
|---|---|---|---|---|---|
| | 75:25 (v/v) | Semi-continuous mesophilic | 6.88 g COD/L/d | 268 mL $CH_4$/g $COD_{removed}$ | Jang et al. (2016) |
| Food waste | 29:71 (COD) | Thermophilic | – | 849 mL $CH_4$/g $COD_{added}$* | Kim et al. (2007) |
| | 50:50 (VS) | Single-stage mesophilic | 2.43 g VS/L/d | 321 mL $CH_4$/g $VS_{added}$ | Heo et al. (2004) |
| Fruit waste | 21:79 (w/w) | Semi-CSTR mesophilic | 3.0 g VS/L/d | 300 mL $CH_4$/g $VS_{added}$ | Fonoll et al. (2015) |
| Orange peel waste | 30:70 (w/w, wet) | Semi-continuous mesophilic | 1 g VS/L/d | 165 mL $CH_4$/g $VS_{added}$ | Serrano et al. (2014) |

[a]The mixed ratio is co-substrate:WAS

[b]The data followed by an asterisk (*) are those that were calculated by the origin data of the literature

15.2 g-COD/L/d was achieved after sulfate addition at 300 mg/L. The corresponding methanogenic activity also reached to the maximum of 0.405 g-$CH_4$-COD/g-VS/d.

### 2.1.2 *Livestock manure*

There are many reports on the anaerobic co-digestion of WAS with livestock manure (Murto et al. 2004; Borowski et al. 2014; Zhang et al. 2014). According to the difference of animal species, the content of livestock manure is totally different in crude protein, carbohydrates, macromolecular substances (e.g. lignocellulose). Pig and chicken manure is rich in protein content. Contrary, cattle manure contains plenty of lignocellulose. Kafle and Kim (2013) pointed that the degradation rate of crude protein is lower than that of carbohydrates. Therefore, several peaks rather than only one of methane production were observed in anaerobic co-digestion of WAS with livestock and poultry breeding by-product (Zhang et al. 2014). In semi-continuous mesophilic reactor for co-digestion of swine waste and WAS, the optimal addition ratio of swine waste was 30% of total mass, which got a 402 mL-biogas/g-$VS_{added}$ methane yield (40% more than mono-digestion of WAS) (Borowski et al. 2014). However, when the operational model changed to thermophilic digestion, the percentage of swine manure could increase to 50% and the corresponding OLR increased from 1.91 to 4.68 g-VS/L/d. The methane production in thermophilic reactor also rose to 470 mL/g-$VS_{added}$ (Creamer et al. 2010; Borowski et al. 2014). When WAS mixed with cattle manure at a VS ratio of 3:7 in semi-continuous mesophilic reactor, methane generation was only 120 mL-$CH_4$/g-TS (total solid) (Dai et al. 2016b). That maybe attribute to the high lignocellulose content of cattle manure. The above results suggested that thermophilic condition could endure higher OLR and achieve higher biogas yield than mesophilic. Additionally, the pathogen inactivation of manure during mesophilic co-digestion is inefficient, so the pre- or post-treatment are necessary to achieve the utilization of digestate for agricultural purposes (Borowski et al. 2014).

Manure, for instance cow dung and sheep manure, usually contains abundant rumen microbes, which make the anaerobic co-digestion process become faster and more productive (Zhang et al. 2014). However, livestock and poultry manure contains relatively high nitrogen content, which may bring positive or negative influence on the anaerobic digestion. On the one hand, appropriate ammonia concentrations contributed to the buffering capacities due to the neutralization of $NH_3{\cdot}H_2O$, which could hold a stable status of the system to some extent. On the other hand, excess ammonia had significantly inhibition to methanogens, further resulting in the VFA accumulation (Murto et al. 2004). When addition ratio of poultry manure was beyond 10% in co-digestion system of WAS and poultry, the significant inhibition of methanogenesis by free ammonia could be observed (Borowski et al. 2014).

#### 2.1.3 Microalgae

Microalgae is becoming the most promising sources of biodiesel to replace the petroleum-based fuel due to their characteristics of nonfood source, higher growth speed and adaptability (Mandal and Mallick 2009). Compared with cellulose wastes, algae contains lower lignin (0–2%) and more easily hydrolyzed sugars (10–30%) and proteins (40–70%), which make it easy to be decomposed in anaerobic digestion. Besides, nutrient elements like C, N and P and some micronutrient elements (e.g. Fe, Co, Zn) in microalgae are also found to be beneficial for methanogenesis (Ververis et al. 2007; Ajeej et al. 2015)

Compared with WAS (primary and secondary sludge), microalgae had the lower C/N or higher $NH_3$ content converted from high protein, so it yielded less methane than WAS (Ajeej et al. 2015). However, microalgae is a suitable co-substrate for anaerobic digestion of WAS. Olsson et al. (2014) observed that co-digestion of microalgae and sewage sludge from municipal waste water treatment in VS ratio of 37: 63 promoted biogas production in mesophilic condition and methane yield reached to 408 ± 16 mL-$CH_4$/g-$VS_{added}$, which was 23% higher than that of sewage sludge alone. They ascribed the improvement to the addition of micronutrients introduced by the microalgae (Karlsson et al. 2012). In the study of Wang et al. 2013a, although co-digestion of microalgae *Chlorella* and WAS only yielded similar gas production from mono-digestion of WAS, the gas phase could reach more quickly. Furthermore, the co-digestion of microalgae and WAS not only obtains the co-digestion products with better dewaterability, but also recycles the released nutrients. Compared with 100% WAS, co-digestion with 11% algae made capillary suction times (CST) value more than half less, simultaneously maintained similar nutrients release ($NH_4^+$-N and $PO_4^{3-}$-P) (Wang et al. 2013a). Because the content level of protein and lipid in the cell walls was different for various microalgae species, the divergence of methane yields in anaerobic co-digestion of WAS with various microalgae species was great (Ajeej et al. 2015; Olsson et al. 2014).

It's worth noting that protein-rich algae biomass is easier to produce a toxic environment in higher temperature (thermophilic conditions) due to the releasing of extreme high concentration ammonia. Therefore, the biogas production was relatively low in thermophilic conditions compared to mesophilic conditions. Olsson et al. (2014) reported that 37% of microalgae (wet slurry) could achieve the highest biogas production (408 ± 16 mL-$CH_4$/g-$VS_{added}$) in the mesophilic co-digestion with WAS. However, in thermophilic condition (55 °C), the same mixture ratio could only bring a 113 mL-$CH_4$/g-$VS_{added}$ biogas production, which was even lower than that of WAS alone (159 mL-$CH_4$/g-$VS_{added}$). The non-adaption of inoculum to thermophilic condition and higher ammonium release from protein degradation in higher temperature jointly caused the results. Furthermore, the cultivation style of microalgae also influences the performance of digestion. Hidaka et al. (2017) soundly cultivated the native wild type *Chlorella* in the dewatering filtrate of anaerobically digested sludge without controlling microalgae species. Nearly 46% higher heating value of *Chlorella* mixture (22 MJ/kg) was recovered as methane during anaerobic co-digestion of *Chlorella* and WAS. Olsson et al. (2018) employed the microalgae growing in municipal wastewater as the co-substrate in mesophilic digestion of sewage sludge. The adding microalgae improved the dewaterability of digested sludge instead of the methane production. Contrarily, the methane yield decreased from 200 ± 25 mL-$CH_4$/g-$VS_{added}$ to 168 ± 22 mL-$CH_4$/g-$VS_{added}$. The flue gas containing high content heavy metals was used as a $CO_2$ source during the microalgae cultivation, so the heavy metals content in the microalgae was much high and inhibited the methanogen, resulting in the lower methane yield (Olsson et al. 2018).

An appropriate proportion (around 10–50% based on VS) of microalgae is crucial for higher biogas production in co-digestion with WAS (Wang et al. 2013a; Olsson et al. 2014; Beltran et al. 2016). Too high proportion of algae would limit the biogas production because of tight cell wall of algae and high ammonia toxicity. Thus carbon-rich wastes such as waste paper and corn straw could be added to the co-digestion reactor of WAS and algae to achieve optimal C/N ratio (20–25), high methane production rate, and cellulose activity as well as to eliminate the ammonia toxicity (Ajeej et al. 2015). Yen and Brune (2007) found that when 50% waste paper was added to co-digest with the algae (*Scendesmus* and *Chlorella*), the C/N ratio increased from 6.7 to 18, and the corresponding methane yield also raised from 143 to 293 mL/g-$VS_{added}$.

## 2.2 Industrial organic co-substrates

A large variety of organic wastes are produced from food processing industry, biodiesel industry and other industrial production annually. The common industrial co-substrates in anaerobic co-digestion of WAS are listed in the Table 1. Crude glycerin is the most investigated industrial co-substrate in anaerobic co-digestion with WAS.

### *2.2.1 Food processing industry residues*

Food processing industry produced solid and liquid organic wastes. Solid residues derived from food processing whose sources are extensive, quantities are huge and compositions are intricate. So when they co-digest with WAS, there are totally different effects on the performance of anaerobic digestion. Some common industrial residues such as wine lees, molasses and fruit-juice industrial wastes have been introduced in anaerobic digestion of WAS as the co-substrates. Wine lees, the solid wastes from wine processing industry, are typically characterized by exceptionally high levels of soluble chemical oxygen demand (COD) and high biodegradability. Anaerobic co-digestion of wine lees and WAS (in COD ratio of 80: 20, OLR of 3.2 kg COD/$m^3$/d) achieved a biogas yield of 386 mL/g-$COD_{added}$ in mesophilic (37 °C) condition, however, the co-digestion process failed in thermophilic (55 °C) condition (Da Ros et al. 2014). The addition of trace elements (Fe, Co and Ni) made the unstable thermophilic reactor recover, and the biogas production returned to 450 mL/g-$COD_{added}$. The mesophilic process was more stable than thermophilic according to the variation of pH, alkalinity, ammonium concentration and VFAs (Da Ros et al. 2014). If the digestion product was considered to reuse in agriculture, the thermophilic digestion was more favorable because the inactivation of pathogens *Escherichia coli* was more effective at higher temperature. Molasses, a by-product from sugar factory, could be as a carbon source to adjust the digestion of WAS. Kalemba and Barbusinski (2017) found that 0.5 wt % of molasses to co-digest with WAS gained 95.69 mL/g-$VS_{added}$ biogas yield with 73% methane content, however, adverse effect on dewatering properties of sludge was indicated by the CST. When fruit-juice industrial wastes and WAS (1:3, w/w) was co-digested at food to microorganism (F/M) ratio around 1.65 g/g, the synergetic effect on the performance of anaerobic co-digestion was not significant based on the kinetic coefficients and ultimate methane yields (Hosseini Koupaie et al. 2014). But the co-digestion could significantly decrease the unit cost (about 37%) due to sharing of both facility and operation.

Liquid organic wastes from food processing were also widely used in the anaerobic co-digestion of WAS. Athanasoulia et al. (2012b) investigated the anaerobic co-digestion of WAS with olive mill wastewater (OMW) in mesophilic condition. Although the highly toxic phenols in OMW maybe inhibit the biodegradation and methanogenesis in anaerobic process (Boari et al. 1993), the biogas production in co-digestion process increased 157% compared to mono-digestion of WAS and reached to 77 L-biogas/d. The increase was estimated at 326.1% in terms of added soluble COD (0.98 L-biogas/g-SCOD) (Athanasoulia et al. 2012b). Compared to the solid residues, the co-digestion of WAS with industrial wastewater is easier to failure due to high water content and complex composition. Rao and Baral (2011) reported that the addition of fruit juice wastewater (FJW) in major proportion to sewage sludge resulted in the significant dropping of pH, which inhibited the activity of methanogens, the key organisms to convert the acids into methane. The overload risk often happened in the co-digestion of oily wastewater (OWW) with WAS for a high amount of fat, oil, and grease (FOG) in OWW, thus the critical feeding rate of OMW should be below 30% (v/v) (Athanasoulia et al. 2012b, 2014).

### *2.2.2 Crude glycerol/glycerine*

Crude glycerol/glycerine is a mixture of glycerol, free fatty acids, salts, un-reacted triglycerides and water, which is mainly from glycerol refining treatment and biodiesel production industry and is about 10% by weight of the starting materials (Nartker et al. 2014). Low N (TN $<$ 400 mg/L) content and extreme pH (pH $>$ 9–10 or pH $<$ 4–5) limit the mono-anaerobic digestion of crude glycerol, but it is an ideal co-substrate for WAS due to its easily biodegradable character and high biogas production potential (Siles

López et al. 2009; Fountoulakis et al. 2010; Hu et al. 2012). According to Buswell's equation, 1 mol $C_3H_5(OH)_3$ could produce 1.75 mol $CH_4$ through anaerobic digestion, thus the theoretical methane potential of glycerol is estimated as 244 mL/g-glycerol (Buswell and Neave 1930). During anaerobic co-digestion of WAS, the methane production in stable condition was around 1100 mL-$CH_4$/d without glycerol addition, however, it could reach to nearly 2400 mL-$CH_4$/d after the addition of glycerol (1% v/v in the mixture) (Fountoulakis et al. 2010).

A small amount of glycerol (1–5 vol%) could promote a relatively higher methane production (325–1480 mL-$CH_4$/g-$VS_{added}$) (Table 1). The optimum glycerol loading was from 25% to 60% OLR in the anaerobic co-digestion with WAS, in which the specific gas production improved 82–280% (Nartker et al. 2014). However, a strict control strategy for the glycerol loading is required to avoid the risk of organic overloading (Astals et al. 2011). Glycerol addition could stimulate propionate specific activity of the biomass (Silvestre et al. 2015). Razaviarani and Buchanan (2015) found that except that VFA concentrations (especially propionate) increased, alkalinity, pH, biogas production and methane content all declined with the increasing addition of biodiesel waste glycerin. Many alkaline substances in crude glycerol can serve as catalysts for esterification and promote the conversion of crude glycerol to organic acids (Kurahashi et al. 2017). Interestingly, when the additive amount of crude glycerol was few (0.126 g/L), only methane was produced. Increasing the crude glycerol concentration had a positive effect on hydrogen production, followed by methane production. 5.04 g/L crude glycerol caused the pH of fermentation liquid increasing from 6.49 to 8.85, simultaneously, more organic acids and the hydrogen precursor 1,3-propanediol (1,3-PDO) were measured in the liquid phase (Kurahashi et al. 2017). Adding glycerine to the co-digestion of sewage sludge increased the C/N ratio, which is benefit to the extracellular polymeric substances (EPS) production, resulting in the worsened digestate dewaterability (Silvestre et al. 2015). However, in another case, the digestate dewaterability was not distinctly affected by glycerol addition in a continuous bench-scale co-digester (Jensen et al. 2014).

*Methanosaeta* (acetoclastic) and *Methanomicrobium* (hydrogenotrophic) are the dominant methanogenic genera in stable co-digester of waste glycerin and WAS (Razaviarani and Buchanan 2015). Jensen et al. (2014) found that there was no gross change in microbial community structure and only minimal changes in diversity when the crude glycerol was added into the anaerobic digestion of sewage sludge, but members of the Phylum *Thermotogae* emerged in the co-digester. *Thermotoga sp.* are regarded as the functional bacteria to ferment glycerol to acetate and hydrogen (Tien and Sim 2012).

## 2.3 Municipal organic co-substrates

### *2.3.1 Municipal food wastes*

Because of the various compositions of municipal food waste (FW), the recommended mixing ratio of food waste and sludge differs in the literatures (Table 1). Simultaneously, plenty of additives in FW also have significant influence on the performance of anaerobic co-digestion. For example, salt (i.e. NaCl) as the prevailing food flavoring usually maintains a high level in FW. Zhao et al. (2016) found that low level NaCl (8 g/L) improved the short-chain fatty acid (SCFA) production in FW-WAS co-digestion reactor and SCFA yield increased from 367.6 (no NaCl addition) to 638.5 mg COD/g of VSS. Appropriate amount of NaCl not only accelerated the release of soluble substance from food waste and disruption of both EPS and cell envelops, but also promoted the conversion of protein released from the co-fermentation system. However, excess NaCl (16 g/L) caused severe inhibition to the acidification and methanogenesis process, resulting in the maximal SCFA yield was only 168.9 mg-COD/g-VSS. Generally, FW have higher C/N ratio than the WAS (Table S1). When the mixed ratio of FW and WAS was adjusted to an optimal C/N value (20–25), the production of soluble proteins, carbohydrates and SCFAs were enhanced consequently (Liu et al. 2015). And the dewaterability of anaerobic digestate was improved (Wang et al. 2018). However, synergetic effects in terms of methane yield and VS removal rate were not found in the co-digestion of WAS with food wastewater and livestock wastewater (Lee 2012). Similarly, Liu et al. (2016) did not observe a synergetic phenomenon at the low-solids co-digestion system (total solid = 4.8%), but found the best synergetic effect in the high-solids co-digestion of low-organic WAS and FW (FW

50 vol%, TS 14%, pH 7.5–8.5). Fruit/vegetable waste is the most attractive food waste for co-digestion with WAS due to its larger VS content and higher biodegradability than WAS, wide source and large output (Gomez et al. 2006; Anhuradha et al. 2007; Rizk et al. 2007). The proportion of VS in vegetable waste and WAS was 87.14% and 62.92% of TS, respectively. So the specific gas yield of vegetable wastes (750 mL-biogas/g-$VS_{added}$) was higher than that of WAS (430 mL-biogas/g-$VS_{added}$) (Anhuradha et al. 2007).

The recommended mixed proportion of fruit/vegetable waste for the co-digestion with WAS was 20–30% (based on wet mass). Under this ratio condition, higher VS reduction (65–88%) and biogas production (165–400 mL-$CH_4$/g-$VS_{added}$) were achieved compared to mono-digestion of WAS (60–250 mL-$CH_4$/g-$VS_{added}$) (Habiba et al. 2009; Serrano et al. 2014; Fonoll et al. 2015). Fonoll et al. (2015) investigated the co-digestion of sludge with different fruit waste (peach, banana and apple waste) in the semi-continuous reactor at mesophilic condition (37 °C). When the type of co-substrate was changed and the OLR kept constant, there was no significant difference in the amount of VFAs. Due to different biodegradability, the specific methane production (SMP) with different fruit waste as the co-substrates was 230–270 mL/g-$VS_{added}$, which was 110–180% that of mono-digestion of WAS. Moreover, the digester alkalinity changed slightly, which should attributed that the alkalinity of co-digester mainly was controlled by the properties of WAS instead of FW (Fonoll et al. 2015). During anaerobic co-digestion of FW-WAS, high amount of VFA from the easily biodegradable organic in FW maybe led to the process instability. Di Maria et al. (2014) found that the total VFAs concentration in co-digestion of waste mixed sludge (WMS) and fruit vegetable waste (FVW) increased from 100 to 250 mg/L with OLR increasing from 1.50 to 2.75 kg-VS/$m^3$/d, accordingly, the specific methane productions was decreased from the peak value of 450 to 250 mL/g-$VS_{added}$. Obviously, the increased amount of rapidly degradable compounds introduced by the FVW caused the results.

For hydrogen generation, anaerobic co-digestion of FW and WAS could also get a better performance than mono-anaerobic digestion. The specific hydrogen production potential of FW was higher than that of WAS (121.6 vs 32.6 mL-$H_2$/g-$COD_{carbohydrate}$) (Kim et al. 2004b), which should be the result of that the organic content in FW was higher than WAS. The mixture of WAS and FW also provided pH buffering capacity for hydrogen fermentation (pH 5.5–6.0). When mixed FW and the blend of sludges (primary sludge and waste activated sludge) with a volume ratio of 1:1, the maximum hydrogen yield reached to 250 mL/g-$VS_{added}$, which was substantially higher than that mono-anaerobic digestion of FW (90 mL/g-$VS_{added}$) (Zhu et al. 2008). Kim et al. (2004b) reported the maximum hydrogen production of 123 mL/g carbohydrate-COD in anaerobic co-digestion of FW and WAS at VS ratio of 87:13 and the VS concentration of 3.0%.

### 2.3.2 *Lipid-rich wastes*

The theoretical biological methane potential (BMP) of lipids (1014 mL-$CH_4$/g-VS) is greater than that of carbohydrates (415 mL-$CH_4$/g-VS) and proteins (496 mL-$CH_4$/g-VS) (Angelidaki and Sanders 2004). In the co-digestion of WAS, the specific methane yield of the lipid-rich organics (> 60% lipid content) as the co-substrate was between 688 and 1040 mL-$CH_4$/g-VS, while the values of carbohydrate- and protein-rich organics as the substrate were recorded 486 and 669 mL-$CH_4$/g-VS, respectively (Ohemeng-Ntiamoah and Datta 2018). Grease trap wastes (GTW) and fat, oil, grease (FOG) are the main co-substrates from city to co-digest with WAS (Table 1). Researches authenticated that the inhibition effects of anaerobic biocenosis, accumulating and forming hardened deposits and other operational problems in mono-digester of lipid-rich wastes could be overcome by co-digestion with WAS (Pereira et al. 2004). In recent years, lipid-rich wastes have been considered as the attractive co-substrates with WAS.

Anaerobic co-digestion could strengthen the specific microbial activity (Yang et al. 2016a). In the co-digestion of FOG with WAS, 40% increase in the release of EPS enhanced the co-digestion system (Yang et al. 2016a). Additional EPS may provide more adsorbing surface for microbe colonization, which is in favour of the biomass degradation. The appropriate addition ratio of grease waste is 20–65% of total VS, which could increase 1.5–4 times methane yield than that of mono-digestion of WAS (Table 1.), and methane content was around 60–70% (Silvestre et al. 2011; Noutsopoulos et al. 2013; Wang et al. 2013b).

70% (based on VS) proportion of grease waste is identified the digestion limitation, inhibition phenomenon would happen when overdose grease waste is added (Wan et al. 2011; Yalcinkaya and Malina 2015). The inhibitory problems of lipids are mainly related to long-chain fatty acids (LCFAs), which have low solubility (Silvestre et al. 2014). LCFAs could be adsorbed onto the cell wall of microorganisms (Cirne et al. 2007) and take an acute toxicity to anaerobic biological communities (Angelidaki and Ahring 1992; Noutsopoulos et al. 2013). Slowly increasing the addition of lipid-rich materials may be a sensible way to strengthen the LCFAs tolerance (Silvestre et al. 2014).

*Methanomicrobium* and *Methanosaeta* were the dominating acetoclastic and hydrogenotrophic methanogen genera in anaerobic digestion, respectively (Razaviarani and Buchanan 2014). The lipid-rich wastes could enhance the acetoclastic microorganism activity but worsen the activity of hydrogenotrophic activity (Silvestre et al. 2014). A higher biomass of slow growing *Syntrophomonas,* which is known to perform syntrophic degradation of complex organics to simple VFAs, was associated with longer anaerobic digester solids retention time (SRT) (Ziels et al. 2016). Lee et al. (2011) noticed the significant shifts in bacterial population when SRT decreased from 20 to 4 days. Dominant Bacteria *Chloroflexi* declined from 28 to 4.5%. *Syntrophomonas* also decreased from 9% to 0%. However, *Bacteroidetes* (12.5 to 20%) and two acetogenic genera belonging to the *phyla Firmicutes* and *Spirochaetales* (6.3 to 12%) all increased.

## 3 Factors of influence on anaerobic co-digestion of WAS

### 3.1 C/N ratio and pH

The C/N ratio and pH value is dependent on the mixed ratio of organic co-substrate and WAS. Among most of the co-substrates, the optimal C/N ratio of anaerobic co-digestion is located between 15 and 30, where a higher biodegradability of the blends could be gotten (Heo et al. 2004). Different C/N or feed ratio would make a significant influence on microorganism in co-digestion system (Jang et al. 2015, 2016; Liu et al. 2015). Moreover, a suitable pH value (pH 5.5–6.5 and pH 7 for acidogenic and methanogenic phase, respectively) is very important to keep a stable digestion process to avoid the negative influence on the activity of methanogenic microorganism. The composition of various co-substrates is different, which lead to the different optimal mixture ratio of co-substrate and WAS to achieve the most suitable co-digestion C/N (15–30) and pH (around 7 for methane production). And C/N ratio and pH of various co-substrates for AcoD with WAS were showed in Table S1 (E-supplementary data).

pH value significantly influences the dissolubility and hydrolysis of organics. High influent pH of anaerobic reactor could promote the anaerobic microbes and protozoa suffer decay because of the alkaline solubilization, the decay products were converted into soluble substrates like VFA, and the anaerobic or anoxic biomass were produced again from that substrates by the growth process (Wang et al. 2007). In the co-digestion of printing and dyeing wastewater and WAS, high influent pH could encourage the reduction of excess sludge production because of the alkaline hydrolysis and self-digestion of some micro-organisms (Wang et al. 2007). When unstable phenomenon happened, pH adjustment promotes good recovery of the digester (Qiao et al. 2015; Montanes et al. 2014).

The stable operation of co-digestion depends on whether it is a high buffered system in some extent (Murto et al. 2004). The buffering capacity of co-digestion with WAS mainly is determined by the bicarbonate/carbon dioxide buffer and other ions buffering system such as ammonium from proteins degraded (Gallert et al. 1998). Anaerobic co-digestion could provide a relative high pH buffering capacity for stable and higher biogas production (Zhu et al. 2008). The feed mixture ratio of anaerobic co-digestion significantly affects the buffer capacity of the digestion process (Heo et al. 2004). However, the own buffering capability of co-digestion reactor often is limited (Murto et al. 2004). Therefore, the ways to enhance the buffered capacity of anaerobic system should be further studied.

### 3.2 Temperature

Temperature exerts a relatively important influence on biogas yield and digestion product. Mesophilic, thermophilic, and even hyperthermophilic conditions have been applied to co-digestion process (Wang et al.

2014a, b). Because of better stability, easy-control and low-cost of operation, mesophilic digestion was widely used in the anaerobic treatment of WAS. It is well-known that the hydrolysis of organics is the rate-limiting step in anaerobic digestion. Considering a fast and balanced conversion of substrate to methane throughout all reaction pathways, thermophilic adaptation of anaerobic microorganisms is a promising technology to make anaerobic metabolism robust for a wide variety of OLR (Kim et al. 2011). Generally, thermophilic condition significantly increases the biogas production and the endurable OLR value compared to mesophilic condition (Gou et al. 2014; Li et al. 2017). The increases of biogas production and OLR could be resulted from: (1) Higher growth rates of thermophilic bacteria and faster biochemical reactions rates than mesophilic system were obtained (Zábranská et al. 2000), although the bacterial diversity in thermophilic co-digestion reactor is lower than mesophilic reactor (Jang et al. 2016). (2) At higher temperature, co-substrates are easy to decompose and the solubilization of substrates to produce sCOD is improved, which strengthen the biodegradability of mixture (Heo et al. 2003). (3) Higher hydrolysis rate can be achieved under the thermophilic condition (Li et al. 2017). Moreover, high temperature decreases the pathogen amount of effluents (Kim et al. 2011).

However, thermophilic anaerobic co-digestion also has some disadvantages, such as low stability, sensitive to inhibitors, great energy requirements, high VFA residue in the effluent, and poor dewaterability (Silvestre et al. 2014; Zábranská et al. 2000). Higher concentration nutrients were released at the thermophilic conditions and subsequent higher VFA or $NH_3$ concentration may lead to the inhibition to methane producing bacteria (Kabouris et al. 2009; Montanes et al. 2015). Great instability occurred at thermophilic condition even at low doses of glycerine in co-digestion of WAS (Silvestre et al. 2015; Silva et al. 2018). Intense inhibitory of LCFAs to thermophilic anaerobe was more than that to mesophilic anaerobes because of the different anaerobes cell wall structure and composition (Hwu and Lettinga 1997; Creamer et al. 2010). Trace metal elements addition (4.3 mg-$FeCl_3$/L, 0.46 mg-$NiCl_2 \cdot 6H_2O$/L and 0.51 mg-$CoCl_2 \cdot 6H_2O$/L in the feed) improved process stability and biogas yield of thermophilic reactor (55 °C), which should attributed that trace metal elements decreased the toxicity of $H_2S$ by facilitating the precipitation of insoluble metal sulfides, simultaneously increased the activity of microorganisms and fundamental enzymes (Takashima et al. 2011; Da Ros et al. 2017). However, the high cost of metals addition and worse dewaterability is the limitation (Da Ros et al. 2017).

### 3.3 OLR and HRT (or SRT)

With the increasing of HRT (or SRT) in a certain co-digestion system, the OLR would decrease. OLR and HRT (or SRT) demonstrate respectively the organic and hydraulic load ability of the anaerobic co-digestion reactor. The stability of co-digestion is significantly affected by the HRT (Angeriz-Campoy et al. 2017). Generally, when HRT decreases to an extreme low level, the methane content in biogas greatly decreases and the alkalinity firstly reaches a peak value then declines, because of the VFA accumulation and the decrease of conversion ratio from protein to ammonium inside the methane tank (Liu et al. 2012; Ratanatamskul et al. 2015; Li et al. 2017). Moreover, the biogas production is slower at higher organic load (Sosnowski et al. 2003). Excessive hydraulic load and organic load maybe cause a failure of the reactor due to the wash-out of microorganisms and THE inhibition to the microorganisms from the VFA accumulation (Murto et al. 2004).

In the two-phased system, the acidogenic HRT and methanogenic HRT usually are 1–5 days and 10–20 days, respectively, and the change of HRT can make different response in different stage. In the acidogenic phase, enough HRT ensures high VFA production. Wang et al. (2014c) found that the propionate percentage became less in acidogenic phase of FW and WAS co-digestion. And in the methanogenic stage, adequate HRT makes a low VFA content to prevent the souring (Dinsdale et al. 2000). Although the increase of HRT (or SRT) to some extent brings higher methane production, pollutant removal efficiency and lower acidification risk for anaerobic co-digestion process, longer HRT (or SRT) decreases the handling capacity of the unit, thus increasing the operating costs (Wang et al. 2014c; Boonnorat et al. 2019). Therefore, a modest HRT (or SRT) is necessary for the maximized profits.

### 3.4 Digester type

Different types of WAS co-digestion devices have been reported in published papers. The construction and operation cost, temperature control, digestion by stages, and running condition are the most commonly considered parameters for better biogas production and pollutants removal.

Two-phase systems (including acidogenic/methanogenic stage reactor and temperature-phased reactor) have been widely applied in anaerobic co-digestion of WAS. And two-phase system showed better performance than single-phase/stage co-digestion system at same HRT because it established the optimum conditions for acid-producing and methane-producing bacteria, respectively. A higher methane yield of 314 mL/g-$VS_{added}$ and average VS reduction of 61% were achieved in the two-stage operation (HRT 15 days). However, a comparative methane yield of 325 mL/g-$VS_{added}$ and VS reduction of 57% required a longer HRT (25 days) in the single-stage reactor (Wang et al. 2017b). Furthermore, in a dual-stage hyper-thermophilic (70 °C)/thermophilic (55 °C) anaerobic co-digestion system, the maximum methane production reached to 576.5 mL/g-$VS_{added}$ at HRT 15 days with a 70% (based on VS) FOG proportion in the mixture of FOG and thickened WAS, while the corresponding value was only 440.4 mL/g-$VS_{added}$ at the same HRT in single-stage thermophilic co-digestor (Alqaralleh et al. 2018). The bacterial structures in the single-phase and two-phase reactor have significant difference. In the co-digestion of FW and WAS, the phylum *Proteobacteria* was predominant in the single-stage anaerobic digestion system in term of operational taxonomic units (OTU), while the phylum *Firmicutes* is more common in the methanogenic phase of two-stage anaerobic digestion reactor (Wang et al. 2017b). Schmit and Ellis (2001) compared the performance of two-stage and temperature-phased system in anaerobic co-digestion of primary sludge and MSW, at the same MSW/PS ratio. The highest methane yield of temperature-phased system (418 mL/g-$VS_{added}$) is higher than that of two-stage system (332 mL/g-$VS_{added}$). This may because the first stage of the temperature-phased system had greater specific hydrolysis rates than the first stage of the two-phase system. However, the higher energy consumption and operational requirement become the limitation of temperature-phased system.

In previous papers, anaerobic co-digestion reactor was optimized by many methods. (1) Adding a pretreatment unit to the traditional digestion system. For example, alkaline pretreatment unit was proved to be a useful way to enhance the methane generation and reduce the VFAs (Dai et al. 2016b). Mo et al. (2017) added a thermo-alkali solubilization unit before the anaerobic digestion elutriated phased treatment (ADEPT). The biogas yield, COD and VS removal efficiencies were significantly boosted due to the increase of dissolved organics, simultaneously, the activity of key enzymes and microorganisms also were improved. (2) Modifying the internal structure of the reactor. For example, the two-stage anaerobic co-digestion of WAS and fruit/vegetable waste (75:25 based on VS), consisting of an acidogenic completely stirred tank reactor (CSTR) and a methanogenic inclined tubular digester (OLR 5.7 kg-VS/$m^3$/d, acidogenic HRT 3 days and methanogenic HRT 10 days) achieved a steady higher biogas production (370 mL-biogas/g-$VS_{added}$) (Dinsdale et al. 2000). Suitable mixing condition in anaerobic co-digester can accelerate the stability of C/N ratio, prevent local acidification because of the good contact between the substrate and the microorganisms, but too strong mixing force also may disrupt the spatial juxtaposition of syntrophic bacteria and their methanogenic partners (Gomez et al. 2006; Rizk et al. 2007). (3) Changing the operational mode. The temperature-phased anaerobic sequencing-batch reactor (TPASBR) system operated under thermophilic (55 °C) and mesophilic (35 °C) conditions at first and second sequencing-batch reaction unit respectively, which could enhance the performance of co-digestion owing to the synergy effect of sequencing-batch, co-digestion, and temperature-phasing. In the case of co-digestion WAS and FW (60:40 based on VS), the maximum methane yield in TPASBR reached to 280 mL/g-$VS_{added}$, while it was only 190 mL/g-$VS_{added}$ in the mesophilic two-stage ASBR (Kim et al. 2004a, 2011).

Generally, quasi-continuous or continuous feeding co-digestion device exhibits better methane production performance than batch reactor (Sosnowski et al. 2003). In some studies, researcher tried to adjust the feeding frequency from once to several times daily so as to regularize a suitable OLR and HRT without irrationally diluting the co-substrates (Dai et al. 2013; Li et al. 2017). High feeding frequency applied in a co-digester of FW and WAS could perform well at a rather high OLR and the lower feeding shock in that system prevented the massive accumulation of VFAs (Dai et al. 2013; Li et al. 2017). Compared the batch reactor and CSTR for co-digestion of greasy sludge and WAS, it could be found that inhibitions happened at 20–30% greasy waste ratio (based on the feed COD) in batch reactor, but at 80% in CSTR. And this may indicate that CSTR could endure higher organic shock loading than batch reactor. Thus Girault et al. (2012) pointed that batch experiments should not be used in determining the maximal ratio of co-substrate.

### 3.5 Pretreatment

Mechanical, chemical and even biological pretreatment were widely applied to enhance the performance of WAS co-digestion. Due to the diverse composition of substrates, each pretreatment gave distinctive effects on different co-digestion biomass (Naran et al. 2016).

Mechanical treatments such as crushing, sieving, compression, drying, evaporation are easy to operate and usually as the first step in WAS treatment. Crushing is necessary for the pretreatment of wastes with longer length or larger volume. The importance of crushing and sieving was also evidenced by De la Rubia et al. (2018). The methane production of MSW after crushing and sieving (20 mm diameter) was 1.6 times that of the MSW without pretreatment. In another example, mechanical biological treatment (MBT) is an effective technology for solid waste treatment, which contains two processing units: mechanical processing such as crushing and air classification, and bio-conversion unit, such as composting or anaerobic digestion (Velis et al. 2009). Over the last 25 years, MBT technologies were popular in Europe and attractive in developing countries. The biggest advantage of MBT is that the quality of pretreated material (suitable physical composition, acceptable content of heavy metals and other contaminant) meets the requirements of processing, and the substrate after MBT is more advantageous for co-digestion due to the great reduction in biomass and size (Pahl et al. 2008). In the co-digestion of primary sludge and MSW after MBT, the biogas production and methane content were 130 mL/g-$VS_{added}$ and 43% (25% MBT products), 240 mL/g-$VS_{added}$ and 47% (12.5% MBT products), while the values were 290 mL/g-$VS_{added}$ and 35% in control (100% primary sludge). Though synergistic effect on biogas yield was not observed in the co-digestion, the methane content in biogas increased with the addition of MBT products (Pahl et al. 2008).

Microwave and ultrasonic treatments could destroy the cell wall of the microorganism, which results in the discharging of intracellular matter and fluid, further promoting the substrates biodegradation (Baier and Schmidheiny 1997). The level of soluble COD, protein or carbohydrate all increased after microwave or ultrasonic pretreatment (Guo et al. 2008). In anaerobic co-digestion of olive pomace and pretreated WAS by ultrasonic (at frequency of 20 kHz, amplitude of 70% and power of 200 W for 30 min) and microwave (at 175 °C and 2000 kPa for 30 min), the pretreated WAS by ultrasonic and microwave resulted in 124% and 152% increase in the methane production respectively, compared to co-digestion of un-pretreated WAS (Alagoz et al. 2015). Quiroga et al. (2014) evidenced that ultrasound pretreatment of cattle manure and sewage sludge allowed shorter HRT and higher energy yields in the co-digester of cattle manure, FW and WAS. The methane recovery increased 67% and 31% at 55 °C and 37 °C, respectively. Furthermore, the pretreatments affect the evolution of microorganism community. Zhang et al. (2016) pretreated the FW and WAS by microwave (MW) at 600 W and used them as the co-digestion substrates respectively. At the active methane production phase, *Methanosphaera* dominated in co-digestion of MW-FW and WAS while *Methanosarcina* was predominant in co-digestion of FW and MW-WAS.

Thermal pretreatment is widely applied in contamination removal due to the dissolution promotion, reaction acceleration, pathogen removal. For example: Steam-treatment (200 °C and 1.0–2.0 MPa for 15 min) of *Quercus serrata* chips reduced the inhibit components, increased the methane conversion ratio and decreased the acid-soluble lignin content in the chips (Wang et al. 2014a). Hygienization (70 °C for 60 min) improved methane production from the co-digestion of meat-processing by-products and WAS, but the impact on the quality of the digestate was little (Luste and Luostarinen 2010). In the co-digestion of thermal hydrolysis FW with WAS, the optimal thermal hydrolysis temperature for FW solubilization was 150 °C within the range of 100–175 °C (Liu et al. 2015). However, in the co-digestion of microalgae-WAS, thermal pretreatment (75 °C for 10 h) to microalgae accelerated the release of inhibitory compounds (i.e. $NH_4^+$-N, VFA) and increased the potential phytotoxicity, but these disadvantages were weakened by co-digestion due to the dilution effect of WAS. Meanwhile, the fermentation stabilization and co-digestate hygenisation were achieved through co-digestion (Sole-Bundo et al. 2017).

Alkaline pretreatment is a proved way to enhance the dissolution of certain refractory materials. Alkaline pretreatment could applied on WAS or co-substrates (e.g. MSW, microalgae). In the co-digestion of WAS and FW, alkaline pretreatment of WAS enhanced the solubilization of sludge particle, and the maximum SCOD solubilization after 4 h improved from 27.7 to 38.3% with the increase of reacted temperature from 25 to 55 °C. And when the mixed ratio of FW/WAS was in a VS ratio of 10: 90, the ultimate methane production from the mixtuture of FW and pretreated WAS was 63% higher than that from the mixtrue of FW and un-pretreated WAS (Heo et al. 2003). In another study, alkaline pretreatment of WAS has been established that alkaline circumstance (especially pH 10) in favor of SCFA production, but inhibit the activities of methanogens significantly in the mono-digestion of WAS (Zhao et al. 2015). NaOH alkali pretreatment of MSW not only promoted the swelling of solids which provided more accessible habitats for anaerobic microorganisms, but also broke down the complex structure of lignin and hemicelluloses. Therefore, higher biomethane yield of 337 mL/g-$VS_{added}$ was observed in co-digestion of WAS and alkali pretreated MSW while the methane production was around 250 mL/g-$VS_{added}$ in co-digestion of WAS and MSW (Ahmadi-Pirlou et al. 2017).

### 3.6 Supplement of additives

The addition of flocculants, adsorbing materials, surfactants, metal elements, enzymes and other matters may have influences on biogas yield and co-digestate character, and their individual and combined effect were investigated (Yang et al. 2010, 2017; Luo et al. 2011, 2013; Wang et al. 2017c). In the previous study, some deleterious effects on WAS from self-carrying toxicity factors (e.g. enrichment of nickel) could be overcome by the supplement the additives (e.g. EDTA or citrate) (Yang et al. 2017). A low dosage of sodium dodecyl benzene sulfonate (SDBS) (0.02 g-SDBS/g-dry sludge) could improve the activities of amylase and protease, thereby the WAS digestion process was accelerated (Yang et al. 2016b). It suggested that suitable additives can be applied to overcome the negative effects from co-substrates in co-digestion of WAS.

Polymer such as polyacrylamide (PAM), poly-glycoside surfactants, enzymes are widely used in the anaerobic co-digestion of WAS. Cationic PAM can increase the density of anaerobic active bacteria and mass transfer resistance. In the co-digestion of flocculated WAS and wine distillery wastewater (3:1, v/v), the highest total biogas production at thermophilic (55 °C) and mesophilic (35 °C) condition was 11.7 and 6.7 L/$L_{mixture-added}$, respectively when PAM addition was 5 g-PAM/kg-TS. Contrarily, the maximum biogas production without PAM addition was 11.1 and 8.2 L/$L_{mixture-added}$ in thermophilic and mesophilic reactor, respectively (Tai et al. 2009). This indicated that PAM supplement may inhibit the methane production in co-digestion of WAS, but with the increase of digestion temperature (from 35 to 55 °C), this inhibition was weakened. By co-digesting WAS with winery waste, a good dewatering property of digestate was obtained when enough dose of cationic polymer (Hidrofloc C 675-Hydrodepur, more than 25 g/kg-dry sludge) was added to the reactor (Da Ros et al. 2014). However, in the pig slurry-WAS co-digestion system, PAM dose beyond 12 g/kg-TS resulted in the inhibition to hydrolysis due to the strong colloidal aggregation and relatively high ammonia nitrogen content (Campos et al. 2008). In the mesophilic anaerobic co-digestion of FW, WAS and green wastes, 5 mg/g alkyl poly-glycoside (APG, non-ionic surfactants) showed a positive effect on anaerobic co-digestion, but a harmful effect appeared at 15 mg/g, which resulted in the change of microbial community structure in reactor (Sun et al. 2019). It also has been proposed that 0.06 g/g-dry sludge additional mixed enzymes (protease/α-amylase, 1:3 w/w) significantly affect the level of dissolved organic matter and EPS in digestion system (Luo et al. 2013; Yang et al. 2010). Therefore, the polymer dose should be controlled at a reasonable level so that suitable particle aggregation and microbial community structure could be obtained in anaerobic co-digestion system.

Inorganic additives, especially metal or metal ions were also implemented to promote the anaerobic co-digestion of WAS. For instance, 5 g/L zero-valence iron ($Fe^0$) significantly enhanced the reduction of tetracycline resistance genes (except *tetw*) and class 1 integrons in co-digestion of WAS and kitchen waste, but extra amount of $Fe^0$ could not affect the amount of *tet* genes and *intI1* considerably (Gao et al. 2017). $Ca^{2+}$ in concentration range of $1.8 \pm 0.1$ to $6.3 \pm 0.5$ g/L had positive influence on the $H_2$ yield from anaerobic co-digestion of OFMSW with paperboard mill sludge and gelatin solid waste (Elsamadony and Tawfik 2015). It was found that Mg, K and S elements related to the performance of anaerobic co-digestion of fibre sludge and WAS, and a stable process was reached when the Ca: Mg ratio below 16:1 (Ekstrand et al. 2016).

Besides, adding nano-particles and biochars provide a feasible method to overcome the poor digestion performance owing to their excellent material properties. Li et al. (2018) added the sawdust-derived biochar into the co-digestion reactor of WAS and FW. The biochar showed excellent capacity to promote the co-digestion performance through enhancing the system buffer capacity, stimulating the syntrophy of anaerobic microorganisms and enriching the communities of *Methanosaeta* and *Methanosarcina.*

## 4 Conclusion and perspectives

This review introduced the progress of anaerobic co-digestion of WAS with different organic feedstocks such as municipal, industrial, agricultural waste. Because of the different composition for each kind of substance, different effects are observed on their co-digestion with WAS. Hence, appropriate mixing strategies are necessary for different kind of organic co-substrate. Simultaneously, the main operational factors including temperature, pH, organic loading rate (OLR), C/N ratio, digester type, pretreatments and additives show significant influence on enhanced biogas production of co-digestion. These important criteria could be used to select the co-substrates and design the anaerobic reactor for WAS co-digestion.

For better understanding of co-digestion with WAS, some research gaps are listed as follow: (1) Reported industrialize co-digestion cases of WAS in the literature are scarce, although this does not indicate the cases reported in the literatures are the only ones in practice. To industrialize the biogas plant, the public administration bodies need to improve the solid and liquid waste sorted collection and management. Additionally, the large-scale comprehensive utilization of co-digestate is still a challenge. (2) Enhanced ways such as hyperthermophilic pretreatment, supplement of new accelerator, and high organic loading need to be further studied and implemented at the site-scale. Reducing the energy consumption and attenuating the influences of enhanced co-digestion system on environment are still major concerns for WWTP managers. And more detailed life cycle assessment of enhanced anaerobic co-digestion in term of global carbon balance is needed. (3) To date, some accelerators also have been used in WAS co-digestion process to improve the biogas production, such as biochars, activated carbon, microelement, and biological agent (i.e. enzyme). However, the deep mechanism investigation of the accelerators is needed. (4) The combination of mixing three or more individual wastes needs more heuristic researches. And the simultaneous biodegradation of emerging co-contaminants, such as antibiotics, pharmaceutical and personal care products should also be taken seriously. (5) Co-digestion of WAS and other organic wastes increases the nutrient load of reactor, the extra N and P may result an instability of co-digestion system. Therefore the ways to solve the nutrient problem are necessary. Adding nano-particles and biochars may provide a feasible method to overcome this problem owing to the excellent material properties. (6) Today, though the anaerobic digestion model No.1 (ADM1) has been established already and tested by lots of researches, few papers of WAS co-digestion address the modeling aspects. Further development of the model of WAS co-digestion need to consider the intrinsic characteristics of WAS, and introduce the conversion and corresponding inhibitory effect of common pollutant in WAS such as trace heavy metal and antibiotic substance.

**Acknowledgements** This research was financially supported by National Natural Science Foundation of China (NSFC) (Nos. 51779088, 51709104), Hunan University Innovation Foundation for Postgraduate (CX2017B097), Postdoctoral Innovation Support Program (BX20180290) and Postdoctoral Science Foundation (Grant No. 2018M640595).

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