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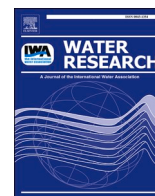
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# Impact of primary treatment methods on sludge characteristics and digestibility, and wastewater treatment plant-wide economics

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## ABSTRACT

Biogas production from anaerobic sludge digestion plays a central role for wastewater treatment plants to become more energy-efficient or even energy-neutral. Dedicated configurations have been developed to maximize the diversion of soluble and suspended organic matter to sludge streams for energy production through anaerobic digestion, such as A-stage treatment or chemically enhanced primary treatment (CEPT) instead of primary clarifiers. Still, it remains to be investigated to what extent these different treatment steps affect the sludge characteristics and digestibility, which may also impact the economic feasibility of the integrated systems. In this study, a detailed characterization has been performed for sludge obtained from primary clarification (primary sludge), A-stage treatment (A-sludge) and CEPT. The characteristics of all sludges differed significantly from each other. The organic compounds in primary sludge consisted mainly of 40% of carbohydrates, 23% of lipids, and 21% of proteins. A-sludge was characterized by a high amount of proteins (40%) and a moderate amount of carbohydrates (23%), and lipids (16%), while in CEPT sludge, organic compounds were mainly 26% of proteins, 18% of carbohydrates, 18% of lignin, and 12% of lipids. The highest methane yield was obtained from anaerobic digestion of primary sludge ( $347 \pm 16$  mL CH<sub>4</sub>/g VS) and A-sludge ( $333 \pm 6$  mL CH<sub>4</sub>/g VS), while it was lower for CEPT sludge ( $245 \pm 5$  mL CH<sub>4</sub>/g VS). Furthermore, an economic evaluation has been carried out for the three systems, considering energy consumption and recovery, as well as effluent quality and chemical costs. Energy consumption of A-stage was the highest among the three configurations due to aeration energy demand, while CEPT had the highest operational costs due to chemical use. Energy surplus was the highest by the use of CEPT, resulting from the highest fraction of recovered organic matter. By considering the effluent quality of the three systems, CEPT had the highest benefits, followed by A-stage. Integration of CEPT or A-stage, instead of primary clarification in existing wastewater treatment plants, would potentially improve the effluent quality and energy recovery.

## 1. Introduction

Wastewater treatment is essential for the protection of public health and ecosystems. Wastewater treatment plants (WWTPs) are in the first place designed to meet the required effluent criteria in terms of organics (expressed as biological oxygen demand (BOD) or chemical oxygen

demand (COD)) and nutrients. Besides, energy efficiency has become more and more important. Over the last decade, WWTPs have even been rebranded as water resource recovery facilities to recover the energy and other resources included in the wastewater (Coats and Wilson, 2017). Municipal wastewater contains chemical energy (1.5–1.9 kWh/m<sup>3</sup> of wastewater), which is enclosed in the chemical bonds of organic

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molecules (Scherson and Criddle, 2014; Hao et al., 2019). Furthermore, wastewater itself can be considered a thermal source of energy (4.6–7.0 kWh/m<sup>3</sup> of wastewater), which can be recovered by heat pumps (McCarty et al., 2011; Hao et al., 2019). Thus, recovering this energy has the potential to cover more than the energy consumption (0.3–2.1 kWh/m<sup>3</sup> of wastewater) in the WWTPs (Siegrist et al., 2008; Gandiglio et al., 2017).

The anaerobic digestion process is widely applied to convert the organics in sludge into biogas (methane), which can be utilized in combined heat and power units for energy recovery. This recovered energy is used to offset the energy consumed in the WWTP (Appels et al., 2008; Abdelrahman et al., 2021). Generally, the biomethane potential (BMP) test is performed to measure the digestibility of any substrate including sludge. In this test, inoculum obtained from a well-functioning digester is mixed with the sludge in bottles. These bottles are incubated under a specific temperature and mixing intensity, and the generated methane amount is then counted (Holliger et al., 2016). BMP represents the maximum methane amount, which can be produced from sludge during anaerobic digestion per mass of volatile solids (VS) or COD. BMP can be used to design full-scale digesters in terms of digester size, organic loading, and potential biogas production. It also can be used to investigate treatment options prior and after anaerobic digestion (Filer et al., 2019).

To recover the energy, primary clarifiers are constructed to redirect part of the organics from the wastewater into the sludge line for anaerobic digestion. These clarifiers capture up to 40% of the organics present in wastewater, while the rest of the organics are transferred to a downstream biological system (Wan et al., 2016; Ersahin, 2018). Increasing the capture of organic matter will improve the energy recovery in WWTPs (Ozgun, 2019). Novel process configurations have been developed to concentrate soluble and suspended organic matter in sludge, e.g., based on A-stage and chemically enhanced primary treatment (CEPT) (Wan et al., 2016). A-stage, the first stage of the Adsorption-Bio-oxidation (A-B) process, is a high-rate activated sludge (HRAS) system which is operated at a high sludge loading rate (> 2 g BOD/g VSS/d) followed by an intermediate clarifier. The activated sludge system is operated at short hydraulic retention times (HRTs) (<1 h) and sludge retention times (SRTs) (<2 days), and low dissolved oxygen (DO) concentration (<1 mg/L). In this process, extracellular polymeric substances (EPS) produced by the bacteria play an important role in capturing the organic carbon via adsorption and bioflocculation mechanisms (Rahman et al., 2017). The removal of COD can reach more than 70% with the help of this physical entrapment (Kinyua et al., 2017; Guven et al., 2019a). CEPT is another configuration, in which chemicals such as ferric chloride (FeCl<sub>3</sub>) or poly-aluminum chloride are added to the wastewater to enhance the coagulation and flocculation of organics. Then, the organics are collected through the underflow of a clarifier, which can be constructed on half of the area of a conventional primary clarifier. High COD removal efficiency was reported by the CEPT process, in which 80% of COD can be redirected to sludge (Guven et al., 2019a).

Several studies have investigated the digestibility of CEPT sludge and A-stage sludge (A-sludge). Kooijman et al. (2017) compared the digestibility of primary sludge and CEPT sludge obtained with different flocculant concentrations (2.5–10 g/kg). A decrease in BMP was reported for sludge samples with flocculant concentrations of more than 5 g/kg. This observation was explained by the fact that flocculants partially irreversibly bound organics and made them unavailable for digestion. Ge et al. (2013) investigated the digestibility of A-sludge from an A-stage system operated under different SRTs (2–4 days) and found that specific methane production was lower at SRT of 4 days (306 mL CH<sub>4</sub>/g VS) compared to 2 days (352 mL CH<sub>4</sub>/g VS), which was explained by higher COD oxidation in A-stage at higher SRT. Meerburg et al. (2015) reported that the specific methane yield of A-sludge was more than two-fold of that of waste activated sludge. Taboada-Santos et al. (2020) compared the digestibility of sludge originated from CEPT and

A-stage via the BMP test. It was found that CEPT sludge yielded similar methane as A-sludge, which was around 300 mL CH<sub>4</sub>/g VS. Different methane production of sludge reported in the literature could be related to the different physicochemical composition of each sludge (Bernat et al., 2017).

The wide range of reported values for BMP of the sludge originating from the previously mentioned systems is sometimes conflicting, which requires further research to investigate to what extent these different treatment steps affect the sludge characteristics and digestibility. Thus, characterization of A-sludge and CEPT sludge in comparison with primary sludge is important to understand the effect of use of these systems on sludge stream and energy recovery potential. Therefore, the aim of this study was to investigate the characteristics of primary sludge, A-sludge and CEPT sludge in detail. The physicochemical characteristics and digestibility of each sludge were investigated. Degradation of each organic fraction and changes in sludge characteristics during anaerobic digestion were determined. Additionally, techno-economic analysis was carried out to investigate the impact of sludge digestibility on the economic feasibility of the integrated systems. The techno-economic analysis, including plant-wide mass balance for COD, nitrogen and phosphorous, energy balance and operational costs, was performed for each configuration.

## 2. Materials and methods

### 2.1. BMP tests

#### 2.1.1. Inoculum characteristics

Inoculum to perform the BMP test was obtained from a full-scale (8000 m<sup>3</sup>) anaerobic digester of a WWTP with a daily treatment capacity of 250,000 m<sup>3</sup> in Istanbul. The ratio of volatile solids to total solids (VS/TS) in the inoculum was around 47.6%. The characteristics of the inoculum are illustrated in Table 1, which fulfilled the recommended values in the studies of Holliger et al. (2016) and Angelidaki et al. (2009).

#### 2.1.2. Substrates

Sludge samples from primary clarification, A-stage and CEPT were used as substrates in this study. All sludge samples were screened through a 10 mm sieve to remove coarse particles and kept at 4 °C. The primary sludge sample was taken from a primary clarifier of a full-scale municipal WWTP with a daily treatment capacity of 600,000 m<sup>3</sup>. The WWTP contained two circular primary clarifiers with a diameter of 50.5 m and a depth of 3.2 m, which are operated at an HRT of 30 min. The A-sludge sample was obtained from a pilot-scale A-stage system. This pilot system was fed with municipal wastewater after grit removal units in a preliminary WWTP. The average DO and MLSS concentrations in the biological tank were 0.50 and 2600 mg/L, respectively, and the average of SRT was 0.5 day. The HRT of the A-reactor and clarifier were 75 and

**Table 1**  
Inoculum characteristics.

Parameters	Mean value ± standard deviation	Recommendation
TS (g/L)	24.64 ± 0.11	–
VS (g/L)	11.73 ± 0.12	–
COD (g/L)	16.85 ± 0.01	15–20 <sup>a</sup>
Alkalinity (g CaCO <sub>3</sub> /L)	6.50 ± 0.05	> 3 <sup>a</sup>
pH	7.87 ± 0.01	7.0 - 8.5 <sup>a</sup>
Median particle size (d <sub>50</sub> )	38.43 ± 0.39	–
Ammonium-nitrogen (NH <sub>4</sub> -N) (g/L)	1.05 ± 0.03	< 2.5 <sup>a</sup>
Volatile fatty acids (VFA) (g CH <sub>3</sub> COOH/L)	0.16 ± 0.01	< 1.0 <sup>a</sup>
Specific methanogenic activity (SMA) (g CH <sub>4</sub> -COD/gVS-d)	0.124 ± 0.004	> 0.1 <sup>b</sup>

<sup>a</sup> Recommended by Holliger et al. (2016).

<sup>b</sup> Recommended by Angelidaki et al. (2009).

30 min, respectively. Lamella plate settlers are installed in the clarifier. In order to obtain CEPT sludge, 60 L of raw wastewater was supplied with 100 mg FeCl<sub>3</sub>/L without pH adjustment, using jar-test set-up (Fig. S1). The wastewater was processed as follows: rapid mixing at 150 rpm for 1 min, slow mixing at 50 rpm for 15 min, followed by settling for 30 min. The supernatant was withdrawn, and the settled sludge was collected and considered as CEPT sludge.

### 2.1.3. BMP experimental set-up

The BMP test was conducted by using an automated methane potential test system (AMPTS II) (Bioprocess Control, Sweden). The test was conducted in triplicate. The mixture ratio was 2:1 between inoculum and substrate based on added VS. The working volume of the mixture of inoculum and substrates was 400 mL. Oxygen-free deionized water was added to the mixtures to compensate for the missing volume. Phosphate buffer, macronutrients and trace elements were prepared according to Zhang et al. (2014). Cellulose microcrystalline (Sigma Aldrich, USA) was used as a positive control of the BMP test.

### 2.1.4. BMP modeling and kinetics

The modified Gompertz model was used to simulate the digestion process (Fig. S2), as shown in Eq. (1):

$$B(t) = B_0 \cdot \exp \left\{ - \exp \left[ \frac{R_m \cdot \exp(1)}{B_0} (\lambda - t) + 1 \right] \right\} \quad (1)$$

where  $B(t)$  is the simulated cumulative methane yield (mL CH<sub>4</sub>/g VS),  $B_0$  is the simulated highest cumulative methane yield (mL CH<sub>4</sub>/g VS),  $R_m$

represents maximum methane production rate (mL CH<sub>4</sub>/g VS-d),  $\lambda$  refers to the lag phase (d), and  $t$  refers to the SRT in the digester (d). The lag phase ( $\lambda$ ) describes the minimum time for biogas production or acclimation of the bacteria to the environment. The maximum methane production rate ( $R_m$ ) represents the maximum catabolic methane production rate of methanogenic archaea.

## 2.2. Techno-economic assessment

### 2.2.1. Primary and sludge treatment configurations under study

Three different process flow diagrams were designed (Fig. 1), including:

- Scenario 1: a primary clarifier was applied, and the sludge was sent to the anaerobic digester.
- Scenario 2: A-stage was applied as a primary treatment unit and the sludge was thickened by a gravitational thickener then sent to the anaerobic digester.
- Scenario 3: CEPT, including flash and slow chemical mixing tanks, and clarification were carried out and the sludge was sent to the anaerobic digester.

The digestate was assumed to be dumped into landfills in all scenarios.

### 2.2.2. Mass balances

Mass balances were set up for each scenario, describing the fate of COD, total nitrogen (TN) and total phosphorous (TP) in the primary

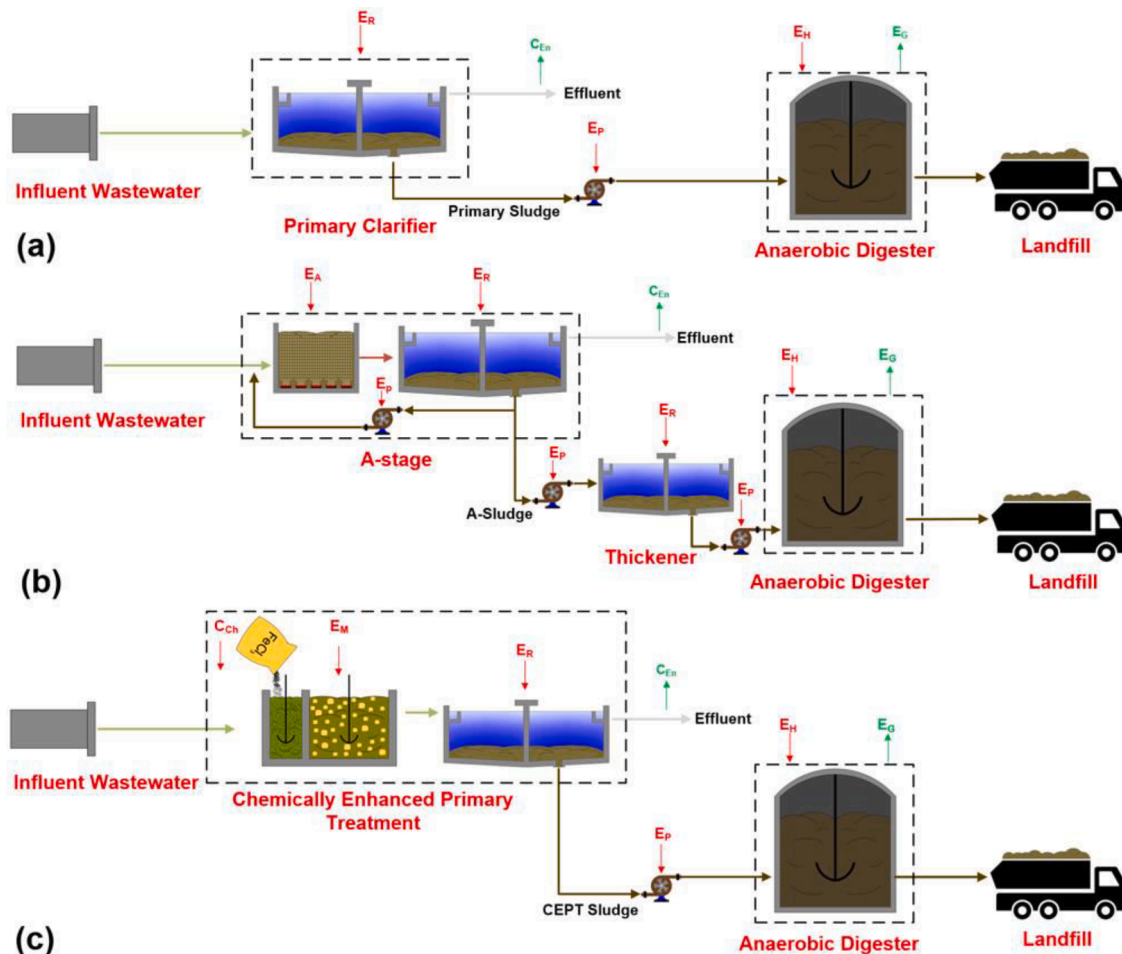


Fig. 1. Techno-economic evaluation boundary for each scenario: (a) primary clarification, (b) A-stage, (c) CEPT. The dashed lines indicate units included in the mass balance calculations. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

treatment units (primary settler, A-stage and CEPT) and in the anaerobic digester. For this purpose, samples of influent, effluent and sludge were taken from each primary treatment configuration. Furthermore, experimental data obtained from the BMP test was used to represent the data needed to conduct the mass balance over the anaerobic digesters. The mass balance did not include the thickener in the A-stage scenario since the A-sludge was not thickened before conducting BMP test. For primary clarification and CEPT configurations, the COD load in the influent ( $COD_{Inf}$ ) (g/d) was considered as the sum of COD load in the effluent ( $COD_{Eff}$ ) (g/d) and in the sludge ( $COD_{Sludge}$ ) (g/d). For the A-stage configuration, the mineralized fraction of COD ( $COD_{Min}$ ) (g/d), which is lost in oxidation for bacterial growth, was used to close the COD mass balance as shown in Eq. (2):

$$COD_{Min} = COD_{Inf} - COD_{Eff} - COD_{Sludge} \quad (2)$$

For the anaerobic digesters, the COD load in the influent sludge ( $COD_{Sludge}$ ) (g/d) was considered as the sum of COD which is converted into methane gas ( $COD_{Methane}$ ) (g/d) and the COD that remained in the digestate ( $COD_{Digestate}$ ) (g/d).  $COD_{Methane}$  was calculated based on the experimental results from BMP test as shown in Eq. (3):

$$COD_{Methane} = \frac{Q_{Methane}}{0.35} \cdot COD_{Sludge} \cdot \left( \frac{VS}{COD} \right)_{Sludge} \quad (3)$$

where  $Q_{methane}$  (L/g VS) is the methane amount produced per g VS of influent sludge; 0.35 is the theoretical methane production per g COD at standard temperature of 273 K and 1 atmosphere pressure (in L methane/g COD);  $(VS/COD)_{Sludge}$  is the VS to COD ratio in the influent sludge.

Like COD, mass balances for TN and TP were conducted for primary treatment configurations and digesters. The influent loads of TN and TP (g/d) were considered as the sum of TN and TP loads in both the effluent and the sludge.

### 2.2.3. Economic analysis

Energy balance and operational cost analysis were estimated for a hypothetical WWTP with a daily influent flow of 500,000 m<sup>3</sup> and COD concentration of 500 mg/L. Net energy recovery ( $E_N$ ) (in Wh/m<sup>3</sup>) was calculated as the difference between energy recovery from methane in the biogas ( $E_G$ ) (Wh/m<sup>3</sup>) and energy consumption (Wh/m<sup>3</sup>) for arm rotation in clarifier and thickener ( $E_R$ ), sludge pumping ( $E_P$ ), chemical tanks mixing ( $E_M$ ), aeration ( $E_A$ ) and/or digester heating ( $E_H$ ), as shown in Eq. (4):

$$E_N = E_G - E_R - E_P - E_A - E_M - E_H \quad (4)$$

Energy recovery was calculated based on the BMP results obtained in this study. Heat and electric energy production from methane in biogas ( $E_G$ ) (Wh/m<sup>3</sup>) was estimated based on Eq. (5):

$$E_G = \frac{Q_m \cdot CV_m \cdot E \cdot 1000}{Q_{inf}} \quad (5)$$

where  $Q_m$  is methane daily production (m<sup>3</sup>/d),  $CV_m$  is calorific energy of methane (kWh/m<sup>3</sup>),  $E$  is heat and electricity conversion efficiency (-),  $Q_{inf}$  is the theoretical influent wastewater flow (m<sup>3</sup>/d), 1000 corrects for kWh to the Wh.

Energy consumed by rotation arm ( $E_R$ ) (Wh/m<sup>3</sup>) in clarifiers and thickeners were estimated based on Eq. (6):

$$E_R = \frac{W \cdot r \cdot v \cdot 24}{e \cdot Q_{inf}} \quad (6)$$

where  $W$  is arm loading factor (N/m),  $r$  is radius of tank (m),  $v$  is tip velocity (m/s), 24 refers to h/d and  $e$  is efficiency (-).

Energy consumption of sludge pumping ( $E_P$ ) (Wh/m<sup>3</sup>) were calculated based on Eq. (7):

$$E_P = \frac{Q_s \cdot H \cdot \rho \cdot g}{e \cdot Q_{inf} \cdot 3600} \quad (7)$$

where  $Q_s$  is sludge flow rate (m<sup>3</sup>/d),  $H$  is pressure head (m),  $\rho$  is sludge density (kg/m<sup>3</sup>),  $g$  is gravity (m/s<sup>2</sup>), and 3600 refers to s/h.

Aeration energy consumption ( $E_A$ ) (Wh/m<sup>3</sup>) in A-stage configuration was calculated based on Eq. (8):

$$E_A = \frac{COD_{Min} \cdot DO_{Sat} \cdot 1000}{A.E. \cdot (DO_{Sat} - DO_{Dis}) \cdot Q_{inf}} \quad (8)$$

where  $COD_{Min}$  is the concentration of mineralized or oxidized COD in the aeration process (kg COD/d),  $DO_{sat}$  is saturation concentration of dissolved oxygen (kg/m<sup>3</sup>),  $DO_{Dis}$  is dissolved oxygen concentration (kg/m<sup>3</sup>), A.E. is aeration efficiency (kg O<sub>2</sub>/kWh), 1000 corrects for kWh to Wh.

Mixing energy in chemical tanks ( $E_M$ ) (Wh/m<sup>3</sup>) in CEPT configuration was calculated based on Eq. (9):

$$E_M = \frac{G^2 \cdot \mu \cdot V \cdot 24}{Q_{inf}} \quad (9)$$

where  $G$  is gradient velocity (s<sup>-1</sup>),  $\mu$  is dynamic viscosity (N·s/m<sup>2</sup>),  $V$  is tank volume (m<sup>3</sup>), and 24 refers to h/d.

Energy consumed to heat the digester ( $E_H$ ) (Wh/m<sup>3</sup>) is the sum of influent sludge heating and heat loss.  $E_H$  was calculated based on Eq. (10):

$$E_H = \left[ \frac{Q_s \cdot (T_{AD} - T_{inf}) \cdot \rho \cdot C \cdot (1 - \Phi)}{3600 \cdot Q_{inf}} \right] + \left[ \frac{A \cdot (T_{AD} - T_{sur}) \cdot U \cdot 24}{Q_{inf}} \right] \quad (10)$$

where  $T_{AD}$  is the temperature of the anaerobic digester (°C),  $T_{inf}$  is the temperature of influent sludge (°C),  $C$  is specific heating capacity of sludge (J/kg·°C),  $\Phi$  is heat recovery efficiency by heat exchanger (-),  $A$  is digester surface area (m<sup>2</sup>),  $T_{sur}$  is surrounding temperature (°C),  $U$  is heat coefficient of heat transfer (W/m<sup>2</sup>·°C), 3600 refers to s/h, and 24 refers to h/d.

The operational costs included the electricity costs for operation of the plant units and the costs of the chemicals in the CEPT scenario. The cost of FeCl<sub>3</sub> ( $C_{ch}$ ) and electricity were considered 220 €/ton in 2008 (De Feo et al., 2008) and 0.1445 €/kWh in 2021 (Eurostat, 2022), respectively. The economic analysis included environmental benefits ( $C_{En}$ ), which represent the costs avoided for removal of undesirable outputs (COD, TN, TP, etc.) during wastewater treatment if these pollutants are released into the marine environment based on the prices reported in the study of Hernández-Sancho et al. (2010). The  $C_{En}$  (€/m<sup>3</sup>) was calculated as shown in Eq. (11):

$$C_{En} = C_{TSS} \cdot TSS + C_{COD} \cdot COD + C_N \cdot TN + C_P \cdot TP \quad (11)$$

where  $C_{TSS}$ ,  $C_{COD}$ ,  $C_N$ , and  $C_P$  (€/kg) are the environmental benefits of the removal of TSS, COD, TN, and TP, respectively. TSS, COD, TN, and TP (kg/m<sup>3</sup>) are the concentrations of TSS, COD, TN, and TP, respectively, in the effluent of the primary units. All design parameters used in this economic evaluation are illustrated in Table S2. Chemical and electricity costs, and environmental benefits were corrected for the time value of money, based on Eq. (12):

$$C_F = C_P \cdot (1 + i)^n \quad (12)$$

where  $C_F$  is future value (€),  $C_P$  is present value (€),  $i$  is the interest rate (%), and  $n$  is number of years (yr).

### 2.3. Experimental analyses

The TS and VS content as well as concentrations of COD, soluble COD (sCOD), TN, total Kjeldahl nitrogen (TKN), NH<sub>4</sub>-N, TP, alkalinity, and

pH of all samples were measured based on standard methods for the examination of water and wastewater (APHA, 2017). VFAs were measured for all sludge samples by a gas chromatograph equipped with a flame ionization detector (Shimadzu, Japan). Protein content of the sludge was estimated based on multiplying the difference between TKN and  $\text{NH}_4\text{-N}$  by 6.25 (FAO, 2002). Soluble carbohydrate (sCarbohydrates) of all sludge samples was measured based on the phenol-sulfuric acid method (Dubois et al., 1956). Lipids content in all sludge samples was measured by using the chloroform-methanol extraction method (Bligh and Dyer, 1959). Cellulose, hemi-cellulose and lignin were measured based on the Van Soest Method (Van Soest, 1963). The particle size distribution of sludge samples was measured by a Mastersizer 2000 (Malvern Instruments, Hydro 2000 MU, UK). Sludge samples were imaged by a scanning electron microscope (Thermo Fisher Scientific Inc., FEI Quanta FEG 250 ESEM, UK) coupled with an energy dispersive x-ray spectrometer (Ametek GmbH, AMETEK EDAX Apollo X, Germany). Principal component analysis (PCA) was performed to investigate the correlation between sludge characteristics (organic fractions) and digestibility of sludge by using Origin 2019b (OriginLab Corporation, USA). The SMA was measured for inoculum and sludge mixtures after BMP to evaluate the activity of methanogenic bacteria after BMP. The SMA was measured by using AMPTS II (Bioprocess Control, Sweden) based on the method described by Abdelrahman et al. (2022). Briefly, the anaerobic sludge samples were analyzed as triplicates. Sodium acetate was used as a substrate. SMA tests were conducted in bottles with effective volumes of 400 mL at 37 °C. The mixing ratio of anaerobic sludge VS concentration to substrate COD concentration was set as 2:1. Phosphate buffer, macronutrients and trace elements were prepared according to Zhang et al. (2014).

### 3. Results and discussion

#### 3.1. Physicochemical characteristics of each sludge

The implementation of a primary clarifier, A-stage or CEPT for wastewater treatment had different impacts on the physicochemical characteristics of the obtained sludge (Table 2). The TS concentration of primary sludge was the highest, which is in the range of the reported values in the literature (20–60 g/L) (Tchobanoglous et al., 2014).

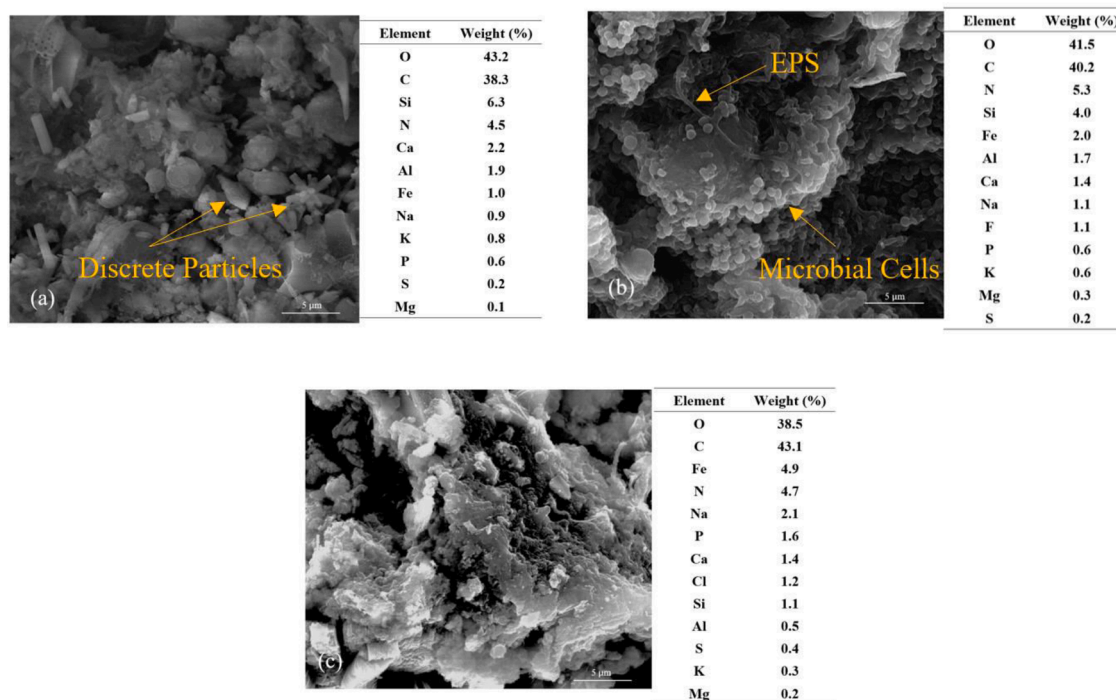
**Table 2**  
Sludge characteristics.

Characteristics	Primary Sludge	A-sludge	CEPT Sludge
<b>Physicochemical Parameters</b>			
TS (g/L)	53.73 ± 0.55	11.14 ± 0.21	27.46 ± 0.16
VS (g/L)	23.35 ± 0.09	5.73 ± 0.06	14.61 ± 0.08
VS/TS (%)	43.5 ± 0.4	51.4 ± 0.4	53.2 ± 0.2
COD (g/L)	36.27 ± 0.07	10.20 ± 0.02	21.27 ± 0.41
TN (%TS)	2.06 ± 0.05	5.54 ± 0.21	3.20 ± 0.05
$\text{NH}_4\text{-N}$ (%TS)	0.54 ± 0.02	2.04 ± 0.05	0.75 ± 0.02
TP (%TS)	0.48 ± 0.01	0.69 ± 0.01	1.00 ± 0.06
pH	5.82 ± 0.02	7.31 ± 0.01	7.25 ± 0.01
<b>Organic fractions</b>			
VFA (mg/g VS)	42.6 ± 1.4	17.3 ± 1.1	9.2 ± 0.1
Proteins (mg/g VS)	208.2 ± 7.8	403.4 ± 28.5	261.9 ± 15.9
Lipids (mg/g VS)	234.2 ± 10.0	158.6 ± 12.0	123.2 ± 11.5
Soluble carbohydrates (mg/g VS)	9.0 ± 0.3	8.4 ± 0.0	11.7 ± 0.8
Cellulose (mg/g VS)	195.5 ± 31.6	98.8 ± 17.0	41.7 ± 11.1
Hemi-cellulose (mg/g VS)	194.5 ± 14.9	121.6 ± 18.8	130.3 ± 25.5
Lignin (mg/g VS)	87.1 ± 6.5	50.0 ± 7.7	180.9 ± 19.1

A-sludge had the lowest solids concentration, which is a typical concentration for activated sludge (8–12 g/L), and requires thickening to 20–30 g/L prior to anaerobic digestion (Tchobanoglous et al., 2014). The VS/TS ratio was relatively low in all sludge types compared to the typical value of primary sludge (60–80%) (Tyagi and Lo, 2013). This low value could be due to inappropriate operation of the grit removal unit, in which some inorganics (e.g., sands) may pass to the primary treatment unit. Therefore, these inorganic particles settle in the primary treatment unit, causing a decrease in the VS/TS ratio in the sludge. TN content was the highest in the A-sludge, which was close to TN content reported for waste activated sludge (2.4–5%) (Tyagi and Lo, 2013). Rahman et al. (2019) reported that assimilation for biomass growth was the main mechanism for nutrients capture in A-stage. CEPT sludge contained a high content of TP, which was double that of primary sludge. This high content was related to the addition of  $\text{FeCl}_3$ , which binds orthophosphate in the wastewater (Wilfert et al., 2015).

Each sludge type had a unique organic fraction composition, including VFA, proteins, lipids, sCarbohydrates, cellulose, hemi-cellulose, and lignin (Table 2). The VFA concentration was the highest in primary sludge, which could explain its relatively low pH. The protein content in A-sludge was the highest, reaching two-fold the protein content in primary sludge. The high protein content could be explained by originating from bacterial cells, exo-enzymes and microbial metabolic products (Guo et al., 2020). Lipids content was the highest in primary sludge contributing to around 23% of VS, which was in the range reported in the literature (7–35%TS) (Tyagi and Lo, 2013). Liu et al. (2020) reported a higher lipids content in primary sludge than in sludge obtained from HRAS systems operated at different SRTs, in which lipids concentration was lower for those operated at higher SRTs. Free fatty acids can be used as substrates for assimilation by microorganisms and released in the wastewater as by-products (Chipasa and Mdrzycka, 2008). The content of carbohydrates, including sCarbohydrates, cellulose, and hemi-cellulose, was around 40% of VS in primary sludge, which was almost double of the carbohydrates content in CEPT and A-sludge. Bernat et al. (2017) reported that organic matter in primary sludge mainly consists of lipids and carbohydrate fibrous material. Cellulose accounts for 25–30% of the suspended solids in the wastewater due to the discharge of toilet papers directly into sewer. It was reported that primary clarifiers can capture around 80% of cellulose, while it is partially degraded in activated sludge systems (Ahmed et al., 2019). Therefore, lower cellulose content could be expected in A-sludge compared to primary sludge. Lignin was around 18% of VS in CEPT sludge, which was higher than those in primary (8.7%) and A-sludge (5%). Ma et al. (2022) reported that the phenolic functional group on lignin-like compounds can provide binding sites for  $\text{Fe}^{3+}$  and form complexes, which facilitate the formation of flocs. Thus, lignin content in CEPT sludge could be relatively high. The remaining uncharacterized organic matter in A-sludge (~14% of VS) and CEPT sludge (~24% of VS) may be other organic compounds such as humic, fulvic, and nucleic acid compounds (Jimenez et al., 2013), and/or organic compounds that were trapped inside the flocs and were not extracted by the used extraction techniques (Zhu et al., 2018).

The morphology of the different types of sludge was imaged by scanning electron microscopy (Fig. 2). Primary sludge contained small discrete particles, which facilitate its gravitational settling, possibly preceded by flocculation with other particles. Bacteria cells conglomerated forming large flocs in A-sludge. In the A-stage process, the bacteria play the main role in the removal of organics, in which particulate and colloidal organics are adsorbed by EPS. In the A-stage, soluble organics are converted to biomass, which will jointly settle with the particulate and colloidal matters in the intermediate clarifier (Kartal et al., 2010). In CEPT sludge, the presence of  $\text{FeCl}_3$  enhanced coagulation and flocculation of particles in the wastewater, forming large flocs (Fig. 2c). The main removal mechanisms of organics in CEPT are charge neutralization and electrostatic bridging (Zhu et al., 2011). Energy dispersive x-ray spectrometer was used to determine the inorganic



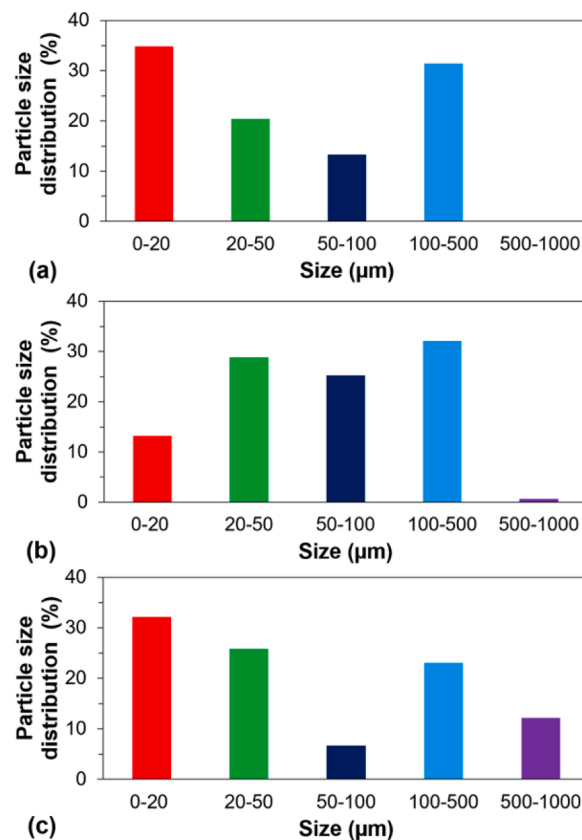
**Fig. 2.** Scanning electron microscope images: (a) primary sludge, (b) A-sludge, (c) CEPT sludge. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

compositions for each sludge. Elements such as O, C and N were the main components in all types of sludge, forming more than 86% of the inorganic elements on the surface of the sludge. Fe and P in CEPT sludge were more than double that of primary sludge and A-sludge because of the addition of  $\text{FeCl}_3$ , which highly removes the phosphorous from the wastewater. This finding was consistent with the measured TP content in each sludge (Table 2).

The particle size distribution was distinctly different in each sludge (Fig. 3). All particle sizes in primary sludge were less than 500  $\mu\text{m}$ , in which the majority with particle sizes less than 20  $\mu\text{m}$  (35%) and between 100 and 500  $\mu\text{m}$  (31%). On the other hand, a small amount of particles with sizes less than 20  $\mu\text{m}$  (only 13%) could be found in A-sludge. The particle sizes were distributed mainly among 20–50, 50–100 and 100–500  $\mu\text{m}$  in A-sludge. On contrary to A-sludge and primary sludge, CEPT sludge was also composed of large particle sizes (500–1000  $\mu\text{m}$ ), which contributed to around 12% of the total mass. Guo et al. (2020) found a small fraction of particles in a size between 500 and 2000  $\mu\text{m}$  in primary sludge, which is consistent with this study. Compared to primary sludge, the presence of bigger particles in A- and CEPT sludge was due to biosorption, bioflocculation, and coagulant binding effect, respectively (Güven et al., 2019a). The average median particle size ( $d_{50}$ ) of primary sludge, A-sludge and CEPT sludge was  $36.8 \pm 0.7$ ,  $60.7 \pm 0.2$  and  $34.0 \pm 0.5$   $\mu\text{m}$ , respectively.

### 3.2. BMP batch test results and modeling

The digestibility of the sludges was assessed by conducting the BMP test (Fig. 4). The average BMP of the positive control (PC) was  $356.3 \pm 17.5$  mL  $\text{CH}_4/\text{g VS}$ . Holliger et al. (2021) reported that BMP values of PC should be between 340 and 395 mL  $\text{CH}_4/\text{g VS}$ , which was fulfilled in this study. The highest BMP value was for primary sludge, which was  $346.6 \pm 16.3$  mL  $\text{CH}_4/\text{g VS}$  on average. This high BMP value might be related to the relatively high lipids content (Table 2), which yields higher methane than protein and carbohydrates (Hu et al., 2020). The BMP value of A-sludge was slightly lower than primary sludge, reaching  $333.3 \pm 5.7$  mL  $\text{CH}_4/\text{g VS}$ . This value was higher than the values



**Fig. 3.** Particle size distribution: (a) primary sludge, (b) A-sludge, (c) CEPT sludge. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)



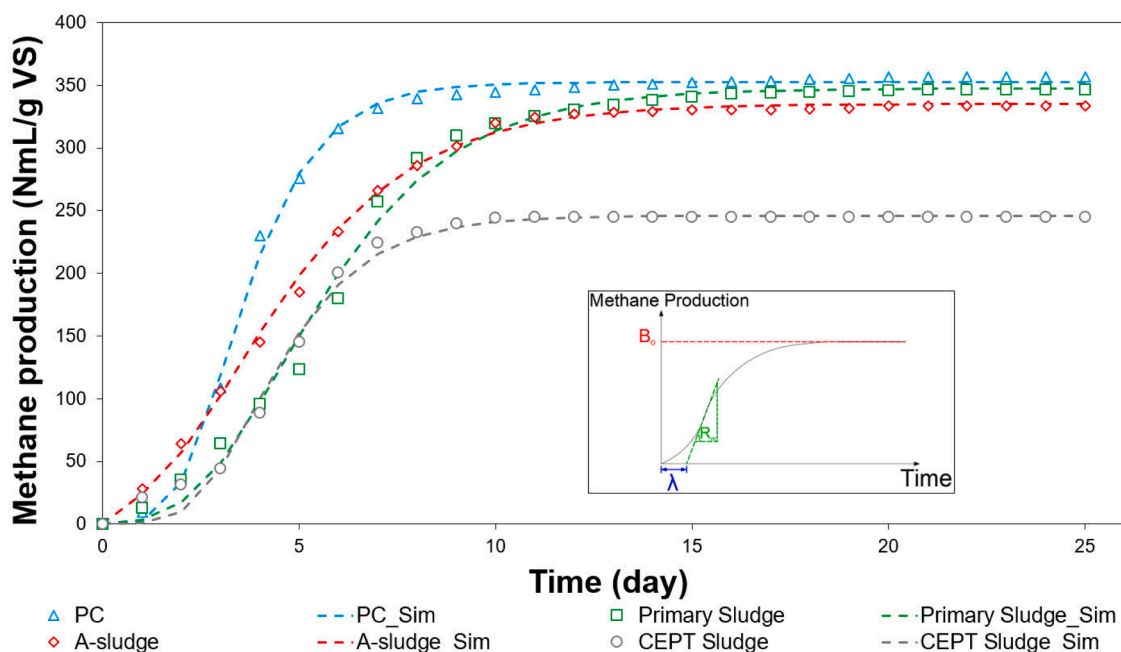


Fig. 4. Experimental and simulated BMP results of each sludge. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

reported by Taboada-Santos et al. (2020), in which A-sludge yielded around 295 mL CH<sub>4</sub>/g VS. The lower BMP value obtained in the latter study could be related to the higher SRT (2.5–3 days) at which their A-stage was operated. The digestibility of the sludge indeed shows an inverse correlation with the operational SRT in activated sludge systems (Guven et al., 2019a). Interestingly, CEPT sludge had the lowest BMP value (244.5 ± 4.5 mL CH<sub>4</sub>/g VS). Lin et al. (2017) and Kooijman et al. (2017) reported that the aggregate flocs due to the addition of coagulant would create a “cage” effect that would restrict the accessibility of bacteria and enzymes to organic compounds trapped inside the flocs, resulting in a relatively low degradability of CEPT sludge.

Different methane production rates and lag phases were observed for each sludge (Fig. 4). Therefore, the modified Gompertz model was used to determine the methane production instead of a simplified first-order rate model (Kafle and Chen, 2016). The methane production curves were well-fitted with the modified Gompertz model ( $R^2 > 0.95$  for all curves) (Fig. 4). The average  $B_0$  values for primary, A- and CEPT sludge were 347.3 ± 16.9, 335.0 ± 5.2 and 245.9 ± 5.5 mL CH<sub>4</sub>/g VS, respectively. The CEPT sludge showed the highest  $R_m$  (57.7 ± 0.6 mL CH<sub>4</sub>/g VS·day) and  $\lambda$  (2.3 ± 0.1 day) among the other sludges (Fig. 5). The kinetics of primary sludge was close to CEPT sludge with an average  $R_m$  and  $\lambda$  of 54.0 ± 2.0 mL CH<sub>4</sub>/g VS·day and 2.2 ± 0.1 day, respectively. The shortest lag phase was during A-sludge digestion (1.0 ± 0.0 day), which could be related to its relatively high protein content (Table 2). Astals et al. (2014) reported that proteins yielded methane with shorter lag phase than carbohydrates and lipids. A-sludge had slightly lower  $R_m$  (49.0 ± 0.3 mL CH<sub>4</sub>/g VS·day) in comparison with other sludges.

The organic composition of sludge affects its digestibility (Mottet et al., 2010). Therefore, PCA was applied for each sludge to understand the correlation between the organic fractions and the observed BMP (Fig. 6). The first component (PC1) (65.2%) and second component (PC2) (30.6%) together represent more than 95% of the total dataset variability. Different score combinations for PC1 and PC2 were obtained for the three sludge types, which confirms that they had distinct organic fraction characteristics and BMP. The angle between the vectors in the PCA graph represents the correlation between variables. Small angles (< 90°) represent positive correlation and angles of 90 and 180 correspond

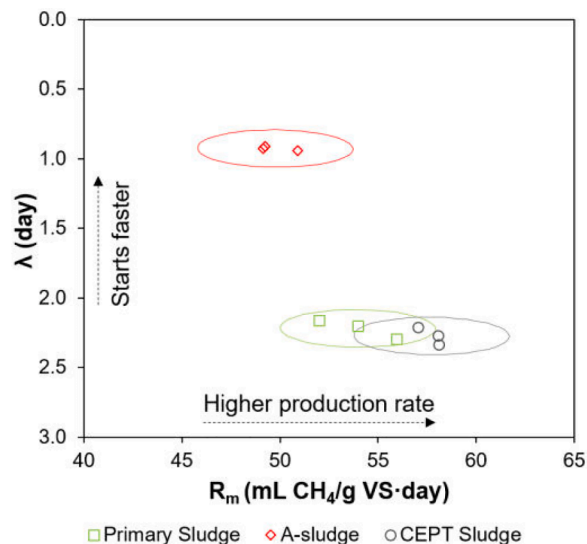


Fig. 5. Correlation between maximum methane production rate ( $R_m$ ) and lag phase ( $\lambda$ ) for each sludge. The three markers for each sludge type indicate triplicate samples. The circle represents 95% confidence level. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

to not correlated and negative correlations, respectively. Strong positive correlations were found between BMP and fractions of cellulose, lipids and VFA. Lignin exhibited a strong negative correlation with BMP. Only a weak positive correlation between BMP and hemicellulose was observed, and a very weak positive correlation between BMP and proteins.

### 3.3. Changes in sludge characteristics during digestion

The effect of sludge type and sludge characteristics on anaerobic digestibility was investigated by determining the sludge characteristics before and after digestion (Table 3). COD removal was consistent with

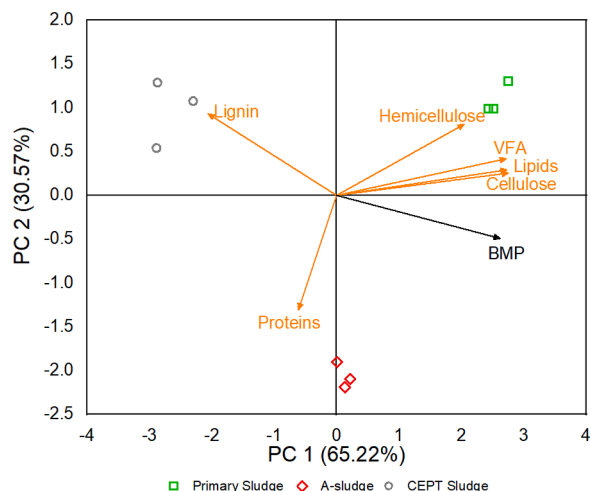


Fig. 6. Loading plot of PCA. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Table 3

Characteristics of sludges before and after BMP.

Parameter	Sludge characteristics before BMP			Sludge characteristics after BMP		
	Primary Sludge	A-sludge	CEPT Sludge	Primary Sludge	A-sludge	CEPT Sludge
COD (g/L)	12.78 ± 0.08	12.29 ± 0.11	13.33 ± 0.08	7.58 ± 0.10	7.16 ± 0.04	9.90 ± 0.27
NH <sub>4</sub> -N (mg/L)	734 ± 5	807 ± 3	711 ± 1	787 ± 3	19	772 ± 4
pH	7.51 ± 0.01	7.42 ± 0.01	7.50 ± 0.01	8.00 ± 0.02	8.09 ± 0.01	7.91 ± 0.01
d <sub>50</sub> (µm)	36.64 ± 1.02	40.64 ± 0.75	30.73 ± 0.27	34.53 ± 0.76	34.33 ± 0.24	30.10 ± 0.76
SMA (gCH <sub>4</sub> -COD/gVS-d)	-	-	-	0.135 ± 0.004	0.156 ± 0.003	0.133 ± 0.003

BMP results, in which COD removal with primary sludge digestion was the highest (40.7%), followed by A-sludge digestion (38.8%). The COD removal during digestion of CEPT sludge was the lowest (25.7%), which resulted in a high COD concentration in the digested sludge. The residual organics in digested sludge can be converted to energy through thermal processes such as pyrolysis (Cao and Pawlowski, 2012). NH<sub>4</sub>-N concentration and pH increased after digestion of all sludges due to the breakdown of protein compounds (Yenigün and Demirel, 2013). Digestion of A-sludge resulted in a relatively high NH<sub>4</sub>-N production and an increase in pH since A-sludge contained the highest protein content. The SMA was measured for the different sludges after digestion to investigate whether there was any improvement or inhibition in methanogenesis by considering SMA of the inoculum as a reference. The results showed that digestion of A-sludge improved the methanogenesis more than those in primary and CEPT sludge. These results confirmed the BMP results, in which digestion of A-sludge had the lowest  $\lambda$ , indicating the highest bacterial acclimation.

The organic fractions were measured for sludge mixtures before and after BMP to reveal the differences in anaerobic biodegradation of each sludge (Fig. S3). Lipids represented the major fraction of the degraded organic compounds for all sludges, around 57.2, 52.2 and 58.4% during digestion of primary sludge, A-sludge and CEPT sludge, respectively (Fig. 7). Proteins were around 20 and 16% of degraded organic compounds during digestion of A-sludge and CEPT sludge, respectively. No VFA was detected in the sludge after BMP, in which VFA was around 19, 10.3 and 14% of the degraded organic compounds during digestion of

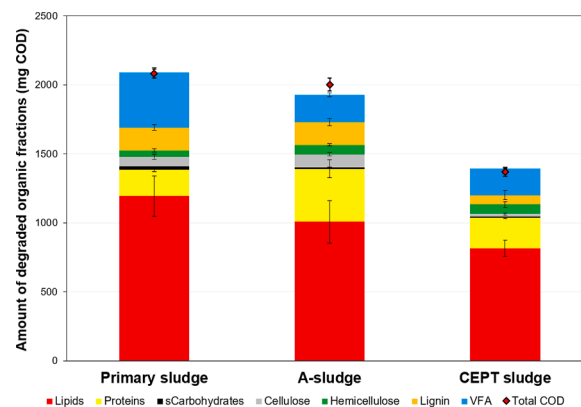


Fig. 7. Organics degraded during digestion. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

primary sludge, A-sludge and CEPT sludge, respectively. Degradation of carbohydrates including cellulose, hemicellulose and sCarbohydrates were responsible for around 6.6, 9.0 and 6.8% of COD decrease during digestion of primary sludge, A-sludge and CEPT sludge, respectively. The other remaining organic compounds such as lignin did not significantly affect the COD decrease (< 9%).

### 3.4. Techno-economic assessment

#### 3.4.1. Mass balances

Mass balances were set up to analyze the fate of influent COD, TN and TP in each scenario (Fig. 8). Primary clarification had the lowest removal efficiency of all parameters, whereas, CEPT had the highest removal efficiency of COD and TP. A-stage had a removal efficiency of COD (64.4%) and TN (22.8%) close to CEPT, with moderate TP removal (32.2%). Rahman et al. (2019) reported that A-stage could capture 19–27% of TN and 30–36% of TP in the influent into sludge. A-stage redirected less COD to sludge for anaerobic digestion, since 13% of COD was lost via oxidation for bacterial growth, which was converted to CO<sub>2</sub>. This value was consistent with the oxidation values reported by Ge et al. (2017), in which COD loss via oxidation was less than 25% at different operational SRTs (0.5–3 days). Integration of an A-stage or CEPT instead of a primary clarifier is expected to affect the side stream as well. Based on the COD mass balance, in comparison with primary clarification, integration of A-stage and CEPT could recover more COD from the wastewater, i.e., 37 and 67%, respectively, for subsequent conversion into methane gas. Partial nitrification-Anammox technology with low aeration requirements can be used for the treatment of effluent of the A-stage and CEPT since COD/TN ratio was low (around 3) in the effluent, which is favorable for Anammox bacteria (typically 2–3) (Zhang et al., 2019).

#### 3.4.2. Energy and cost considerations

Overall energy balance and operational costs were estimated for each scenario (Fig. 9). In the case of primary clarification, nearly all the energy consumed (27.2 Wh/m<sup>3</sup>) was required for operation of the anaerobic digester. Energy consumption was the highest in A-stage scenario (73.9 Wh/m<sup>3</sup>), which was mainly due to aeration in the A-stage system. Aeration contributed to around 54.5% of the total energy consumption in the A-stage scenario, while the rest was mainly for heat requirement of anaerobic digestion (41.4%) and sludge pumping (4.1%), especially for sludge recirculation in the A-stage system. In the CEPT scenario, total energy consumption was around 56 Wh/m<sup>3</sup>, which was mainly because of heating requirement for anaerobic digestion (83.6%) and mixing of chemicals dosing tanks (16.3%). Among all scenarios, heating requirement for anaerobic digestion was the highest in CEPT scenario due to the high sludge production in CEPT

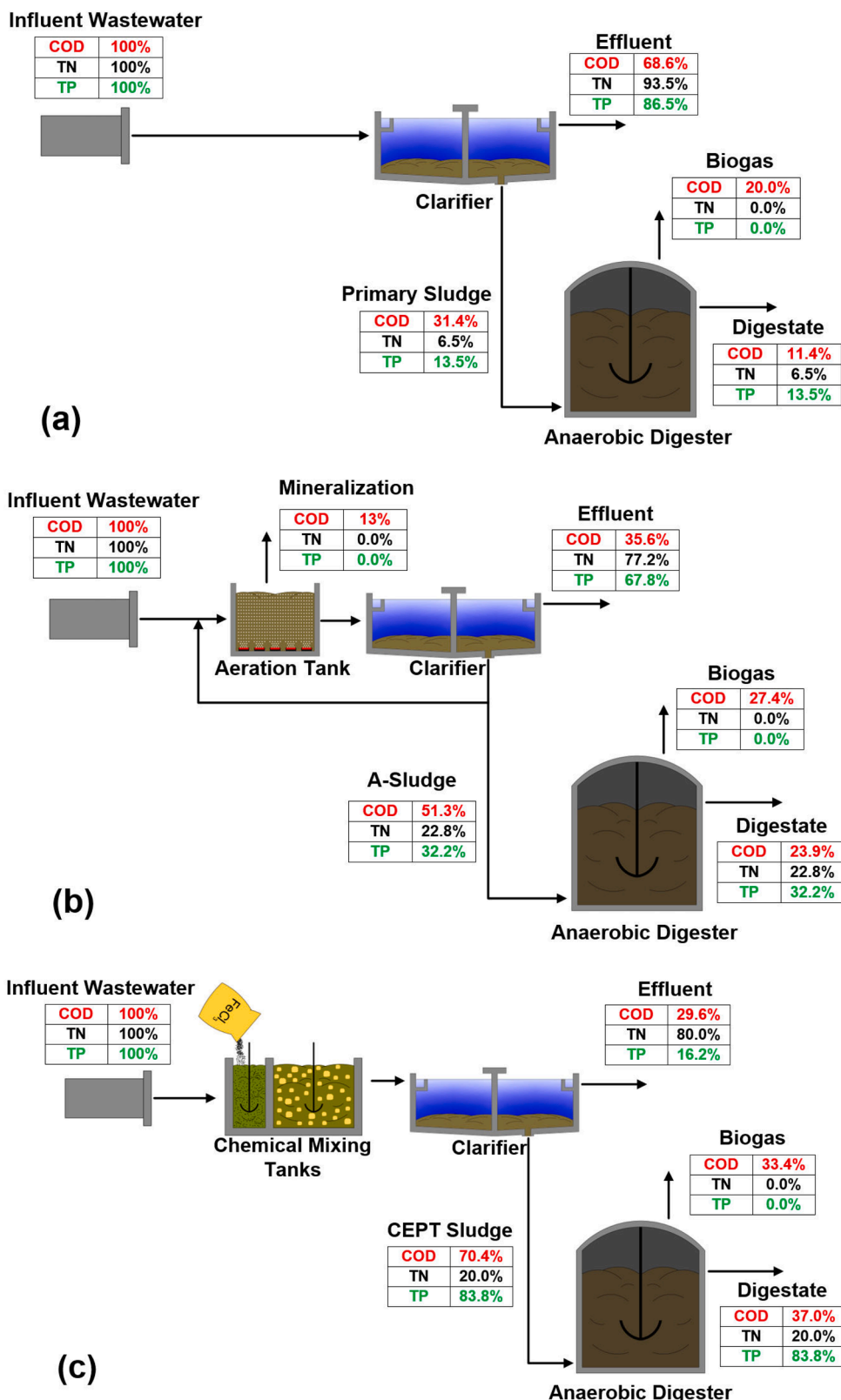


Fig. 8. COD, N and P mass balance: (a) primary clarification, (b) A-stage, (c) CEPT. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

configuration, which needed to be heated (Guyen et al., 2019a). Energy recovery from methane production was the highest in CEPT scenario (449 Wh/m<sup>3</sup>), followed by energy recovery in A-stage (393 Wh/m<sup>3</sup>) and primary clarification (304 Wh/m<sup>3</sup>) scenarios (Fig. 9a). Guven et al.

(2019b) reported that the energy recovery in the form of electricity was 180 Wh/m<sup>3</sup> for a designed WWTP with a capacity of 100,000 m<sup>3</sup>/d and influent COD of 509 mg/L. Based on the energy balance, CEPT showed to be the most energy-efficient system among the other systems with a

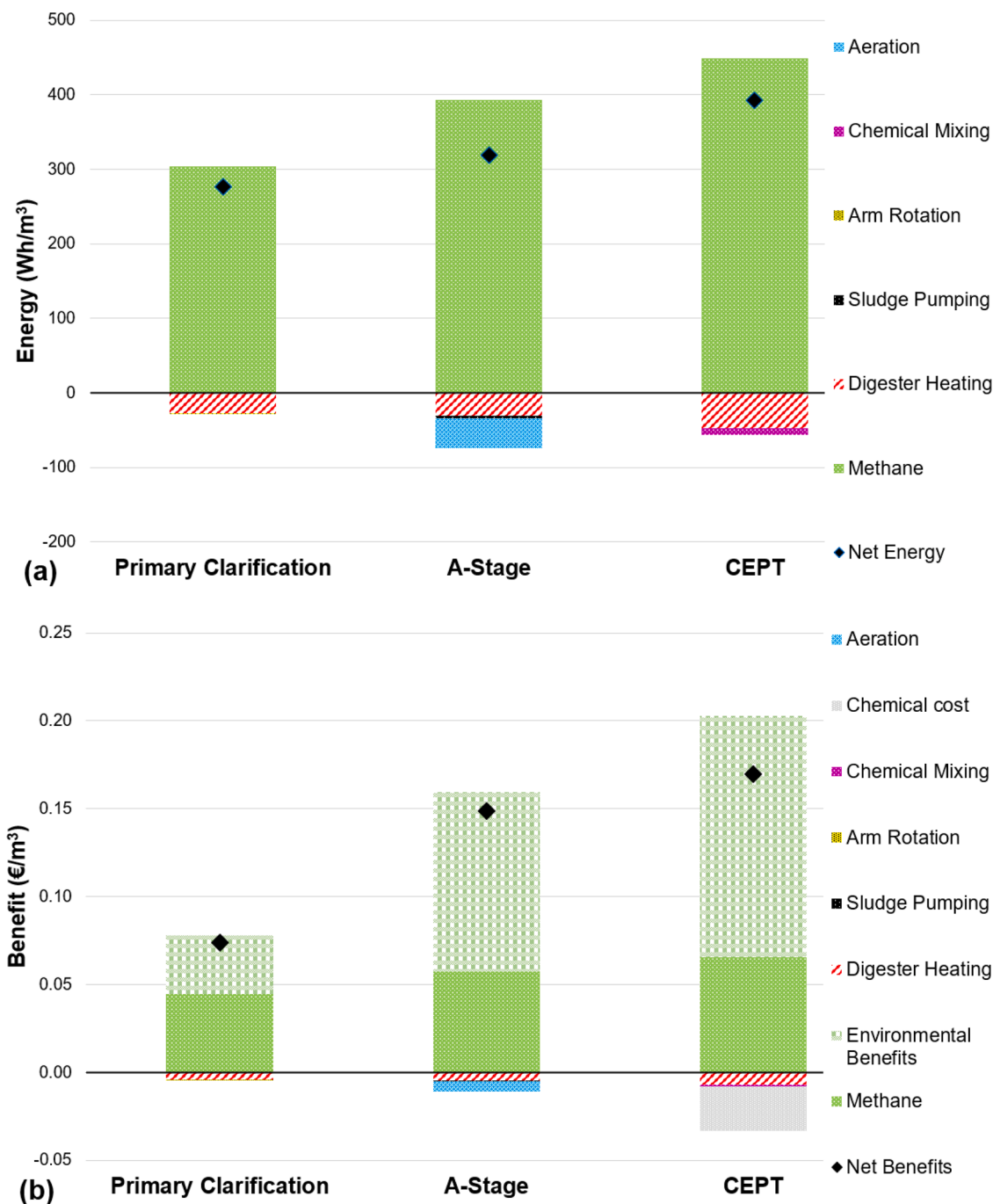


Fig. 9. Economic evaluation: (a) energy balance, (b) cost analysis. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

net positive energy recovery of around 393 Wh/m<sup>3</sup> (Fig. 9a). It should be noted that the highest amount of COD was recovered accompanied by CEPT with a concomitant low energy consumption for mixing of chemicals. The net energy gained via A-stage (~319 Wh/m<sup>3</sup>) was close to that via primary clarifier (~277 Wh/m<sup>3</sup>).

The operational costs in the CEPT scenario (~0.04 €/m<sup>3</sup>) were the highest among all scenarios, mainly due to the use of chemicals, which accounted for around 75.6% of the total operational costs (Fig. 9b). The benefits gained via heat and electricity generation from methane were higher than the operational costs in all the scenarios, which were in the range of 0.04 - 0.07 €/m<sup>3</sup>. Since these systems vary in the removal performance of different pollutants, which could affect the following treatment processes, environmental benefits were integrated into the

economic analysis. CEPT scenario had the highest environmental benefits (~0.14 €/m<sup>3</sup>), followed by A-stage (~0.10 €/m<sup>3</sup>) and primary clarification (~0.03 €/m<sup>3</sup>) scenarios. All configurations had positive net benefits accounting for around 0.17 and 0.15 €/m<sup>3</sup> by A-stage and CEPT, respectively, as benefits, which were significantly higher than benefits gained with primary clarification (~0.07 €/m<sup>3</sup>).

#### 4. Conclusions

- This study investigated the impact of primary treatment methods (primary clarifier, A-stage or CEPT) on sludge characteristics and digestibility, and plant-wide economics. The treatment process/

technology affected the sludge characteristics and digestibility distinctly.

- Primary sludge contained the highest amount of lipids, cellulose and hemicellulose, while A-sludge had the highest amounts of proteins and CEPT sludge had the highest amounts of lignin.
- Digestion of primary sludge yielded the highest amount of methane, followed by A-sludge. CEPT sludge digestion yielded the lowest amount of methane, which was 30% lower than that of primary sludge.
- Based on plant-wide mass balances, the amount of organic matter in wastewater converted into methane gas was around 20, 27.4 and 33.4% with the implementation of primary clarifier, A-stage and CEPT, respectively.
- Energy consumption of A-stage was the highest among the three configurations due to aeration energy demand, while CEPT had the highest operational costs due to chemical use.
- By considering the effluent quality, CEPT and A-stage had more than two-fold net benefit compared to primary clarification.
- Integration of CEPT or A-stage instead of primary clarifier in WWTPs can improve energy recovery and effluent quality.

### Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

### Data availability

No data was used for the research described in the article.

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### Supplementary materials

Supplementary material associated with this article can be found, in the online version, at [doi:10.1016/j.watres.2023.119920](https://doi.org/10.1016/j.watres.2023.119920).

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