

Article

Resilience of Organic Matter Recovery to Seasonal Temperature Changes in a Wastewater Treatment System Adopting the High-Rate Contact Stabilization Process

Kensuke Sakurai *  and Chika Abe 

Innovative Materials and Resources Research Center, Public Works Research Institute, 1-6, Minamihara, Tsukuba 305-8516, Ibaraki, Japan

* Correspondence: recycle-imarrc21@pwri.go.jp

Abstract

The high-rate contact stabilization–activated sludge (HiCS–AS) process, which integrates an upstream high-rate contact stabilization process with a downstream activated sludge process, is a promising approach for enhancing energy recovery from municipal wastewater. This 16-week continuous study evaluated the organic matter recovery capacity and process stability of a HiCS–AS process, configured as a sequencing batch reactor, under realistic conditions where water temperatures decreased from 29 °C to 14 °C. The upstream HiCS process, operating at a solids retention time of 0.60 days, exhibited stable sludge settling behavior and achieved a stable carbon recovery rate of 0.23 ± 0.04 g-COD/g-COD. The overall COD removal rate for the system was $86\% \pm 3\%$, and the HiCS–AS process demonstrated a statistically significant increase in organic matter recovery compared to a parallel single activated sludge process (paired Student's *t*-test, $p < 0.05$). These findings indicate that the HiCS–AS process provides a consistent and effective approach to organic matter recovery compared to conventional processes.

Keywords: carbon recovery rate; sequencing batch reactor; bioflocculation; short solids retention time; biomethane

1. Introduction

Collectively, wastewater drainage and treatment account for approximately 1% of global electricity consumption, and this demand is expected to rise as the number of sewage system users increases globally [1]. Reducing electricity consumption in the wastewater sector has therefore become an important issue in response to the energy crisis and climate change [2,3]. To offset the substantial energy consumption requirements of wastewater treatment plants (WWTPs), it is critical to recover as much organic matter from wastewater as possible, convert it into biomethane, and utilize it as a renewable energy source [4,5].

The high-rate contact stabilization (HiCS) process is a high-rate process capable of minimizing solids retention time (SRT) and achieving significant suppression of oxidation; consequently, it has emerged as a key method for the efficient recovery of organic matter from sewage [6]. Unlike the representative high-rate activated sludge (HRAS) process, the HiCS process features two separate phases: a contact phase and a stabilization phase [7]. During the contact phase, incoming wastewater is mixed and comes into contact with returned sludge. Dissolved organic matter in the influent is absorbed by microorganisms, whereas colloidal and suspended organic matter undergo bioflocculation [8–10]. Sludge



Academic Editor: Christos S. Akratos

Received: 2 June 2026

Revised: 3 July 2026

Accepted: 7 July 2026

Published: 8 July 2026

Copyright: © 2026 by the authors.

Licensee MDPI, Basel, Switzerland.

This article is an open access article distributed under the terms and conditions of the [Creative Commons Attribution \(CC BY\)](https://creativecommons.org/licenses/by/4.0/) license.

settles and is partially withdrawn during the sedimentation phase, whereas the remainder is carried over into the next cycle. The remaining sludge is then aerated through the stabilization phase. During this process, the sludge is subjected to a famine–feast cycle, promoting the production of extracellular polymeric substances (EPS) and enhancing bioflocculation [7,11]. Furthermore, sludge produced in the HiCS process has been shown to generate greater quantities of biomethane than sludge from the conventional activated sludge (CAS) process [12,13].

Combining the primary settling tank (PST) for the initial removal of settleable organics with the subsequent HiCS process is an effective strategy that has the potential to substantially enhance energy recovery efficiency from wastewater. PSTs typically have the disadvantage of considerable space requirements; however, they are energy-efficient devices allowing organic matter to be removed as sludge without being lost through oxidation [14]. The subsequent HiCS process can efficiently recover the low-strength organic matter that passes through the PST via enhanced bioflocculation [11,15]. Primary effluent differs from untreated water by PST in terms of the amount of settleable solids, and it is expected to greatly influence the concentration of sludge in the reactor and the settling efficiency during the settling phase. Although the HRAS process for untreated water by PST is well characterized [10], current understanding of the HiCS process for primary effluent remains insufficient.

To compensate for the relatively weak solid–liquid separation during the HiCS process, it may be beneficial to adopt a sequencing batch reactor (SBR) configuration. Relative to continuous-flow systems, the SBR configuration generally offers improved sedimentation performance because there is no inflow or outflow of water during the sedimentation phase [16,17]. The high-rate operation of the SBR configuration has also been shown to outperform that of continuous systems; the SBR-based HiCS process has thus emerged as a promising approach [18].

To further enhance the overall organic recovery of the system, one practical solution is to install an activated sludge (AS) stage downstream of the HiCS process to capture residual organic matter in the HiCS effluent and recover it as excess sludge. This two-stage SBR combining the HiCS and AS processes was termed the HiCS–AS process. From an energy balance perspective, the advantages of the HiCS–AS process have been demonstrated even when accounting for the additional energy required for aeration and other operations [6]. The HiCS–AS process shares configurational similarities with the A–B process, which has historically been introduced to WWTPs [19]. However, the HiCS–AS process in this study differs in several aspects: it requires a PST, assumes low levels of settleable organic matter in the influent, incorporates contact and stabilization phases, and involves differences between the continuous and SBR configurations. According to Sakurai et al. [13], the carbon recovery rate (CRR)—i.e., the proportion of organic matter recovered as excess sludge relative to the COD mass of the influent—of the HiCS–AS process employing the HiCS process with an SRT of 0.44 days was 0.22 ± 0.06 g-COD/g-COD; however, investigations into optimal SRTs remain insufficient.

In temperate regions, wastewater temperatures typically fluctuate between 15 °C and 25 °C [20]. Understanding the impact of these fluctuations is essential for the full-scale implementation of new technologies, as temperature sensitivity varies significantly with a solids retention time (SRT) [13,21]. While higher temperatures can increase organic matter loss via hydrolysis in systems with an SRT longer than 3 days [21], the effects on short SRT processes remain less clear, particularly during the gradual temperature transitions encountered in real-world operations.

This study addresses these gaps by providing 16 weeks of continuous monitoring of the SBR-based HiCS–AS process under practical, seasonally shifting conditions. The

primary objective is to improve the HiCS process by employing a modified SRT of 0.60 days and to demonstrate its stability and enhanced performance in organic matter recovery relative to a parallel single activated sludge (SAS) process. By analyzing the statistical correlations between temperature changes and key operational parameters, this research clarifies the seasonal resilience of the HiCS–AS configuration as a promising approach for energy recovery in temperate climates. Although nutrient removal can be achieved by employing advanced treatment during the AS stage, the present study did not target nutrient removal because it was designed for application at WWTPs where nutrient removal is not required by regional regulatory guidelines.

2. Materials and Methods

2.1. Operation of the Test Plants

A test plant operating the HiCS–AS process, configured as a two-stage SBR, was installed within buildings at a full-scale publicly owned WWTP treating municipal wastewater at a rate of approximately 100,000 m³/d. To provide a baseline for assessing the organic matter recovery efficiency of the HiCS–AS system, the SAS process was operated in parallel as a control. Both processes were operated in parallel for 16 weeks, spanning summer to winter.

As shown in Figure 1, municipal wastewater entering the full-scale WWTP initially passed through an experimental 54 L PST with a surface overflow rate of 25 m³/(m²·d). The resulting primary effluent was supplied to the HiCS and SAS reactors, and the effluent from the HiCS reactor was directed to the AS reactor. The effective volumes of the HiCS, AS, and SAS reactors were 30, 24, and 30 L, respectively. Each reactor was equipped with influent, effluent, and sludge withdrawal pumps, as well as an air blower (APN-057R, Iwaki Co., Ltd., Tokyo, Japan); the HiCS reactor also featured an agitator. All equipment was timer-controlled at fixed intervals. The flow rates for each reactor are listed in Table 1. The volumetric exchange rate of all reactors was 50%. During each process, treated wastewater was decanted during the final 10 min of the sedimentation phase. No mechanical temperature control was installed.

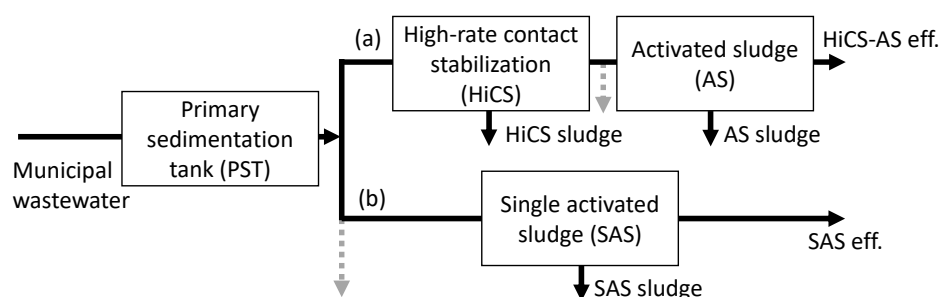


Figure 1. Process flow diagram of the (a) HiCS–AS and (b) SAS processes operated continuously for 16 weeks. Gray dashed arrows indicate escape lines.

Table 1. Flow rates of the HiCS, AS, and SAS processes.

Process	Influent (L/d)	Sludge Withdrawn (L/d)	Effluent (L/d)	HRT (h) ¹
HiCS	180	18	162	4
AS	72	1.8	70.2	8
SAS	90	2.3	87.7	8

Note: ¹ HRT (hydraulic retention time) was calculated by dividing the reactor capacity by the inflow water flow rate.

2.1.1. Operating Cycle of the HiCS Process

The operating cycle of the HiCS process lasted for 120 min, consisting of a 40 min stabilization phase, a 20 min contact phase, and a 60 min sedimentation phase. During

the stabilization phase, air was supplied at an airflow rate that achieved a dissolved oxygen (DO) concentration of approximately 5 mg/L by the end of the phase. The oxygen remaining and carried over into the contact phase was consumed by the influent, reducing DO levels below 0.5 mg/L during the first half of the contact phase; this environment is considered suitable for bioflocculation [7]. Excess sludge was withdrawn at the start of the stabilization phase. During the contact phase, no aeration was applied, and the sludge was gently stirred at 200 rpm using a propeller-type agitator to prevent accumulation at the bottom. Primary effluent was introduced during the first half of the contact phase. Sludge settling during the sedimentation phase was then carried over to the next cycle. A portion of HiCS effluent not supplied to the AS process was either used for analysis or discharged.

2.1.2. Operating Cycle of the AS and SAS Processes

The operating cycle of the AS and SAS processes consisted of a 180 min aeration phase followed by a 60 min sedimentation phase, totaling 240 min. After excess sludge was withdrawn at the start of the aeration phase, influent was introduced into the reactor, and the airflow rate was adjusted to maintain a DO level of 2 mg/L or higher to prevent oxygen deficiency.

2.2. Sampling and Analysis

Samples for analysis were collected weekly during the 16-week operational period. One sampling event was omitted because heavy rainfall substantially diluted the influent, resulting in a total of 15 sampling events. Influent and effluent were analyzed for chemical oxygen demand (COD), particulate COD (pCOD), colloidal COD (cCOD), soluble COD (sCOD), total suspended solids (TSS), biochemical oxygen demand (BOD), allylthiourea-BOD (ATU-BOD), ammonium nitrogen (NH_4^+ -N), nitrite nitrogen (NO_2^- -N), and nitrate nitrogen (NO_3^- -N). Excess sludge was collected at the beginning of the stabilization or aeration phase and analyzed for each COD fraction, TSS, volatile suspended solids (VSS), loosely bound (LB) EPS, and tightly bound (TB) EPS. sCOD was determined using the method proposed by Mamais et al. [22], involving coagulation with zinc sulfate at pH 10.5 and measuring the COD of the filtrate passed through a 0.45 μm cellulose acetate filter (Membrane Solutions, LLC, USA). cCOD was calculated by subtracting sCOD from the COD of the filtrate obtained using a glass fiber filter (934-AH, Whatman). pCOD was determined as the difference between COD and cCOD. COD, TSS, VSS, BOD, and ATU-BOD were measured according to Standard Methods [23]. LB-EPS and TB-EPS were measured according to the method proposed by Rahman et al. [24]. NH_4^+ -N, NO_2^- -N, and NO_3^- -N were analyzed using an autoanalyzer (QuAatro39, BL TEC K.K., Osaka, Japan). The pH of the influent was measured using a pH meter (HM-40P; DKK-TOA Corporation, Tokyo, Japan). Water temperature in the reactors was recorded using a temperature logger (TR-71wb; T and D Corporation, Tokyo, Japan).

2.3. Sedimentation Tests of Sludge in the HiCS and SAS Processes

Following the methods proposed by Van Winckel et al. [25] and Mancell-Egala et al. [26], the percentages of sludge with critical settling velocities (CSV) of more than 0.6 or 0.15 m/h were determined for sludge in the HiCS, AS, and SAS reactors. These were denoted as $\text{CSV}_{0.6}$ and $\text{CSV}_{0.15}$, respectively. Two liters of sludge were sampled at the end of the stabilization and aeration phase and immediately subjected to settling tests. Samples were then gently transferred into settleable solid vessels (Standard Methods 2540F) [23] to avoid shear-induced disruption. To measure target CSV values of 0.6 m/h or slower and 0.15 m/h or slower, samples were withdrawn with a pipette from a point 5 cm below the water surface after 5 min and 20 min of settling, respectively. The pCOD of samples before and after

sedimentation was measured. $CSV_{0.6}$ and $CSV_{0.15}$ denoted the percentage difference in pCOD before and after sedimentation relative to the pre-sedimentation pCOD, respectively.

2.4. Calculation

The oxidized COD mass was calculated by subtracting the COD mass discharged as excess sludge and effluent from the influent COD mass. The carbon recovery rate (CRR) represents the mass of COD recovered as excess sludge relative to the influent COD mass. For each process, the CRR was calculated using Equation (1):

$$CRR = \{(\text{COD}_{\text{was}} - \text{COD}_{\text{out}}) \times Q_{\text{was}}\} / (\text{COD}_{\text{in}} \times Q_{\text{in}}) \quad (1)$$

where COD_{was} , COD_{out} , and COD_{in} denote COD in the excess sludge, effluent, and influent, respectively, and Q_{was} and Q_{in} represent the flow rates of excess sludge and influent. As shown in Equation (1), the portion of organic matter present in the excess sludge corresponding to the organic matter present in the effluent was excluded from the CRR calculation to avoid overestimating CRR, because such organic matter is difficult to concentrate and may not be transferred to the anaerobic digestion tank. MRR is the amount of methane generated from recovered organic matter, expressed as COD, and is determined relative to the influent COD mass. MRR is the product of CRR and the methane conversion rate (MCR). The overall CRR of the HiCS–AS process ($\text{CRR}_{\text{HiCS-AS}}$) was calculated using Equation (2) from the CRRs of the HiCS and AS processes (CRR_{HiCS} and CRR_{AS}) as follows:

$$\text{CRR}_{\text{HiCS-AS}} = \text{CRR}_{\text{HiCS}} + E \times \text{CRR}_{\text{AS}} \quad (2)$$

where E is the ratio of COD discharged as HiCS effluent to COD entering the HiCS process. The overall MRR of the HiCS–AS process ($\text{MRR}_{\text{HiCS-AS}}$) was calculated using Equation (3):

$$\text{MRR}_{\text{HiCS-AS}} = \text{CRR}_{\text{HiCS}} \times \text{MCR}_{\text{HiCS}} + E \times \text{CRR}_{\text{AS}} \times \text{MCR}_{\text{AS}} \quad (3)$$

The MCR for HiCS sludge (MCR_{HiCS}) was 0.56 g-COD- CH_4 /g-COD, and the MCR for AS (MCR_{AS}) was 0.45 g-COD- CH_4 /g-COD [27]. The MCR for SAS was also taken as MCR_{AS} . The sludge concentration in the contact phase, SRT, CRR, and COD removal rates for each fraction were calculated using Equations (S1)–(S5), which are outlined in the Supplementary Information.

2.5. Statistical Analysis

To compare the performance of the HiCS–AS and SAS processes, Microsoft Excel 2016 (Microsoft Corporation, Redmond, Washington, DC, USA) was used to evaluate differences in mean CRR value using a paired Student's t -test at the 5% significance level. To assess the relationship between water temperature and operational parameters for each process, Spearman's rank correlation coefficients (r) and p -values (p) were calculated and heat maps were created using Bellcurve 4.05 for Excel (Social Survey Research Information Co., Ltd., Tokyo, Japan). All datasets used a sample size of 15.

3. Results and Discussion

3.1. Water Quality of the Influent and Effluents

Figure 2 shows temperatures of ambient air and influent wastewater during the operation period, along with water temperatures inside the HiCS and SAS reactors on sampling days. All temperatures exhibited a decreasing trend over time. The minimum and maximum influent wastewater temperatures were 20 °C and 29 °C, respectively. Detailed influent and effluent data are presented in Table 2. No seasonal variations were observed in the influent, the pH of which remained between 6.8 and 7.2.

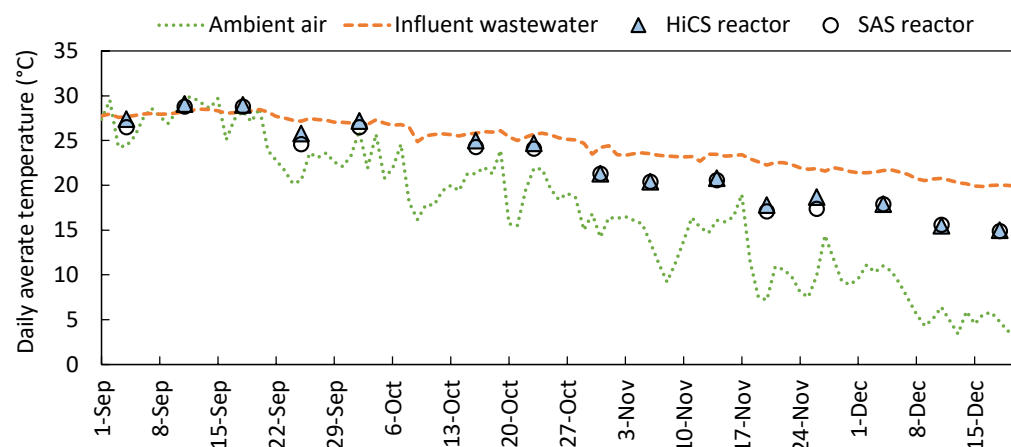


Figure 2. Temperatures of ambient air and influent wastewater during the operational period and water temperature in HiCS and SAS reactors on sampling days.

Table 2. Water quality of the influent and effluent samples collected during the operational period.

	Unit	Influent	HiCS eff.	HiCS–AS eff.	SAS eff.
tCOD	mg/L	316 ± 58	182 ± 28	43 ± 9	33 ± 7
pCOD	mg/L	147 ± 35	72 ± 11	10 ± 5	5.6 ± 4.6
cCOD	mg/L	84 ± 19	61 ± 16	14 ± 7	10 ± 5
sCOD	mg/L	86 ± 14	49 ± 12	19 ± 9	17 ± 7
TSS	mg/L	82 ± 25	35 ± 5	7.4 ± 2.6	7.0 ± 2.1
BOD	mg/L	143 ± 36	65 ± 21	9.3 ± 4.3	6.1 ± 2.3
ATU-BOD	mg/L	n.m.	n.m.	6.3 ± 2.4	4.3 ± 1.3
NH ₄ ⁺ –N	mg/L	30 ± 3	23 ± 2	1.4 ± 3.3	0.1 ± 0.1
NO ₂ [−] –N	mg/L	<0.1	<0.1	3.5 ± 2.6	0.3 ± 0.2
NO ₃ [−] –N	mg/L	<0.1	<0.1	13 ± 5	18 ± 2

Note: n.m.—not measured. Values following ± indicate standard deviation.

Effluent water quality is shown in Table 2. HiCS effluent COD and BOD concentrations were 43 ± 9 mg/L and 9.3 ± 4.3 mg/L, respectively. During 16 weeks of continuous operation, no seasonal deterioration in effluent quality was observed. The difference in sCOD between the HiCS and SAS effluents was 31 ± 13 mg/L, which corresponds to fractions classified as readily biodegradable organic matter in the AS process, as described by Mamais et al. [22]; this fraction was confirmed to remain in the HiCS effluent. According to Haider et al. [28], this behavior likely reflects changes in the microbial community caused by the shortened SRT.

NO₂[−]–N and NO₃[−]–N were not detected in the HiCS effluent. This was attributed to the short SRT, which prevented the retention of nitrifying bacteria and thereby inhibited nitrification. During the AS and SAS processes, the SRT was higher than the threshold typically required for nitrification, but the safety factor was small [20]. Although a decline in nitrification performance was anticipated with decreasing water temperature, the AS and SAS processes removed more than 90% of the ammonium nitrogen. This may be attributed to the advantages of the SBR configuration: its potential for extensive consumption of the ammonia substrate, absence of flow rate fluctuations, prevention of short-circuiting, and favorable sedimentation conditions [29].

3.2. Operating Conditions for Each Process

Table 3 summarizes the operating conditions for each process. The HiCS process had an SRT of 0.60 ± 0.05 days (hereafter, values following ± represent standard deviations). The average MLSS concentration was 316 ± 71 mg/L. In HiCS reactors treating low-strength

wastewater, the concentrations of MLSS in the contact phase are generally below 1000 mg/L due to substantial MLSS discharge from excess sludge and treated effluent [13,15,24]. The values observed in this study were consistent with these previous reports.

Table 3. Operational conditions during each process.

Process		HiCS	AS	SAS
Water temperature (°C)	Min.	15	14	15
	Max.	29	29	29
	Ave.	22	21	22
SRT (d)		0.60 ± 0.05	5.4 ± 0.4	6.2 ± 0.3
MLSS (mg/L)	Stabilization	549 ± 128	-	-
	Contact	316 ± 71	-	-
	Aeration	-	526 ± 163	1379 ± 429
LB-EPS (mg-COD/g-VSS)		81 ± 20	66 ± 21	30 ± 11
TB-EPS (mg-COD/g-VSS)		204 ± 45	236 ± 59	196 ± 36
CSV _{0.6} (%)		80% ± 6%	93% ± 4%	98% ± 1%
CSV _{0.15} (%)		94% ± 3%	99% ± 0%	99% ± 1%

Note: Values following ± indicate standard deviation. “CSV_{0.6}” and “CSV_{0.15}” mean the proportions of pCOD with CSV > 0.6 m/h and >0.15 m/h.

LB-EPS is rapidly generated in response to the feast state during the contact phase, serving to quickly adsorb and capture inflowing organic matter [7,11]. Excess LB-EPS is known to cause a deterioration in sludge settleability [10,30]. TB-EPS contributes to maintaining floc structure and retaining captured organic matter but tends to decrease under high loading conditions [31]. The average LB-EPS value in the HiCS process (81 ± 20 mg-COD/g-VSS) was higher than that observed in the SAS process (30 ± 11 mg-COD/g-VSS), confirming that the HiCS process is suitable for increasing LB-EPS. Li et al. [30] reported that, in HiCS systems operated at SRTs ranging from 0.6 to 5.0 days, LB-EPS increased with decreasing SRT, reaching 80 mg-COD/g-VSS at SRTs of 2 days or less; the present results are consistent with those findings [30]. The average LB-EPS value for the AS process (66 ± 21 mg-COD/g-VSS) was higher than that for the SAS process, suggesting the influence of residual sludge in the HiCS effluent. No substantial differences in TB-EPS were observed among the processes examined.

Settling performance is a critical factor for energy recovery in the HiCS process [32,33]. Poor settleability causes sludge to be discharged with the treated effluent, reducing the amount of excess sludge available for recovery. In the CAS process, when MLSS concentrations exceed 1000 mg/L, particle interference hinders settling [34]. Sludge from the HRAS process, which operates similarly to the HiCS process, also exhibits hindered settling at concentrations between 500 and 1000 mg/L [35]. The MLSS concentrations in the reactors used in this study were below these ranges, indicating that hindered settling was unlikely to occur. From the perspective of promoting bioflocculation, higher sludge concentrations are desirable because they increase contact efficiency with influent organic matter. The minimum solid concentration at which meaningful settling can be observed is defined as the threshold of flocculation (TOF) and characterizes the collision efficiency of sludge particles [26]. For HRAS, the TOF has been reported as 400–700 mg-MLSS/L [26]; in this study, a slightly lower MLSS concentration was observed. Approaching the TOF while suppressing sludge oxidation and decomposition may lead to improved settling performance.

CSV is used as a cutoff value to distinguish whether sludge has settled or remained. Fau et al. [36] proposed that because HRAS has a low settling efficiency, the CSV of 1.5 m/h used in CAS systems is poorly suited, and that a lower CSV value could be adopted for HRAS systems. In this study, previously proposed values of 0.6 and 0.15 m/h [25] were employed. In the HiCS reactor, CSV_{0.6} and CSV_{0.15} were $80\% \pm 6\%$ and $94\% \pm 3\%$, respectively. The settling rate obtained for the HiCS process in this study was lower than that observed in the AS and SAS processes. This difference was attributed to variations in sedimentation properties caused by differences in microbial communities associated with different SRTs, the influence of induced LB-EPS, and reduced flocculation efficiency due to sludge MLSS concentrations below the TOF. Van Winckel et al. [25] reported that, under optimal shear conditions, HRAS showed CSV distributions of 66% (>0.6 m/h) and 89% (>0.15 m/h), similar to those in the HiCS process. The values observed in this study were slightly higher than those previously reported, indicating a relatively favorable settling performance during the HiCS process.

The sludge volume index (SVI) was considered unreliable as an indicator of solid–liquid separability because it overemphasizes the importance of inhibited settling and compression in sedimentation tanks during high-rate processes [10,25]. Consequently, it was not measured in this study.

3.3. Average Removal Performance of Each COD Fraction

This section presents the average removal performance of each COD fraction over the entire 16-week experimental period. The detailed correlations between these parameters and seasonal water temperature fluctuations are discussed later in Sections 3.6 and 3.7. Figure 3 shows the average removal rates of each COD fraction. The total COD removal rate during the HiCS process was $42\% \pm 5\%$ and the removal rates of sCOD and pCOD were higher than that of cCOD. The overall COD removal rate of the HiCS–AS process was $86\% \pm 3\%$, which was close to the tCOD removal rate of the SAS process ($90\% \pm 2\%$).

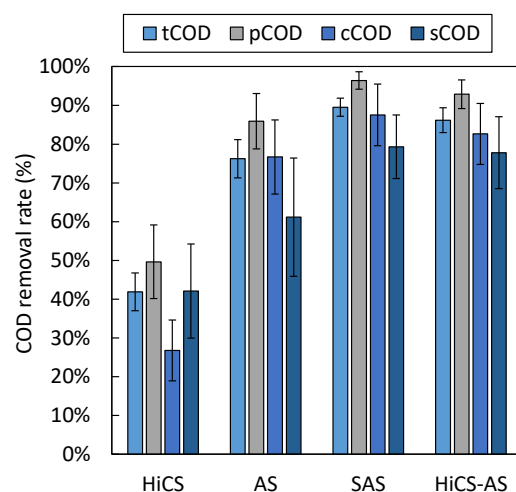


Figure 3. Removal rate of each COD fraction. HiCS–AS represents the combined HiCS and downstream AS processes. Error bars show standard deviations.

pCOD accounts for the largest proportion (47%) of organic matter in the influent as shown in Table 2. pCOD removal is influenced by the settleability of sludge in the reactor and settleable substances contained within the influent. Consequently, the pCOD removal rate of the HiCS process after PST is lower than that of the HiCS process without PST. Furthermore, the effects of hydrolysis and decay also become larger under long SRT. Extending SRT from 0.2 to 0.8 days significantly improved the pCOD removal rate during

the HRAS process from 48% to 84% [36]. The pCOD removal rate for the HiCS process in this study was $50\% \pm 10\%$, i.e., higher than the pCOD removal rate (33–40%) for the HiCS process using the same facility but with a slightly shorter SRT (0.44 days) [13].

Under short SRT conditions (less than 0.5–1 day), cCOD is not absorbed by microorganisms in the manner of sCOD, cannot settle like pCOD, and undergoes limited hydrolysis. Therefore, cCOD removal is primarily attributed to bioflocculation. The removal rate for cCOD was $27\% \pm 8\%$. This value is similar to those reported for studies with an SRT of 0.5 days (20–25%) [7,37]. Extending the SRT to 1.2 days increases the removal rate to higher values (60–80%) [8,18]; however, oxidation also increases, necessitating the exploration of optimal operating conditions for organic matter recovery.

Microorganisms utilize sCOD for cell synthesis, intracellular storage (such as PHA), and oxidation for energy production. When the SRT is less than 1 day, it selectively promotes the growth of specific heterotrophic bacteria capable of rapid growth [28]. These bacteria exhibit high rates of consuming and utilizing easily degradable sCOD [38,39]. The HRAS process with an SRT of 0.2 days has been reported to achieve sCOD removal rates of 29% [38]. Jimenez et al. [8] reported that the sCOD removal rate reaches its maximum (70–80%) under SRT of 0.5 days or longer. Furthermore, sCOD removal rates of 56% were reported for an SRT of 1 day [7,40]. The sCOD removal rate in this study was $42\% \pm 12\%$. Considering that half of the sludge retention time in the HiCS process used in this study occurs under anaerobic conditions, this result is generally consistent with existing research.

3.4. Average Organic Matter Destination and Recovery Performance

As with the COD removal rates, this section evaluates the mean carbon and methane recovery performance throughout the 16-week study. Figure 4 shows the destinations of influent organic matter in the HiCS, HiCS–AS, and SAS processes. The removal rates of influent COD mass in the HiCS, HiCS–AS, and SAS processes were 0.42 ± 0.05 , 0.86 ± 0.06 , and 0.89 ± 0.04 g-COD/g-COD, respectively. Organic matter was removed either through oxidation or by withdrawal as excess sludge. The amount of organic matter oxidized throughout the HiCS process (0.19 ± 0.07 g-COD/g-COD) was considerably lower than the corresponding values for the SAS process (0.60 ± 0.09 g-COD/g-COD), confirming that oxidation was substantially suppressed by the HiCS process.

As shown in Table 4, the CRR values for the HiCS, HiCS–AS, and SAS processes were 0.23 ± 0.04 , 0.34 ± 0.07 , and 0.29 ± 0.08 g-COD/g-COD, respectively. The average CRR of the HiCS–AS process differed significantly from that of the SAS process (paired Student's *t*-test, $p < 0.05$), confirming the effectiveness of the HiCS–AS process for organic matter recovery under practical operational conditions. The CRR of the HiCS process was substantially higher than the corresponding value (0.12 ± 0.03 g-COD/g-COD) obtained using the same facility with an SRT of 0.44 days [13]. The SBR-configured HiCS process in this study achieved a higher CRR than the continuous-configured reported previously [41].

The average MRR values for the HiCS, HiCS–AS, and SAS processes were 0.13 ± 0.02 , 0.18 ± 0.04 , and 0.13 ± 0.04 g-COD/g-COD, respectively. The difference in MRR between the HiCS–AS and SAS processes was 0.05 ± 0.02 g-COD/g-COD, which was statistically significant (paired Student's *t*-test, $p < 0.05$). Assuming a COD concentration of 260 mg/L for PST effluent [42], a methane calorific value of 13.9 kJ/g-COD-CH₄ [43], and a power generation efficiency of 35% [44], this difference corresponds to a power generation potential of 18 ± 8 Wh/m³.

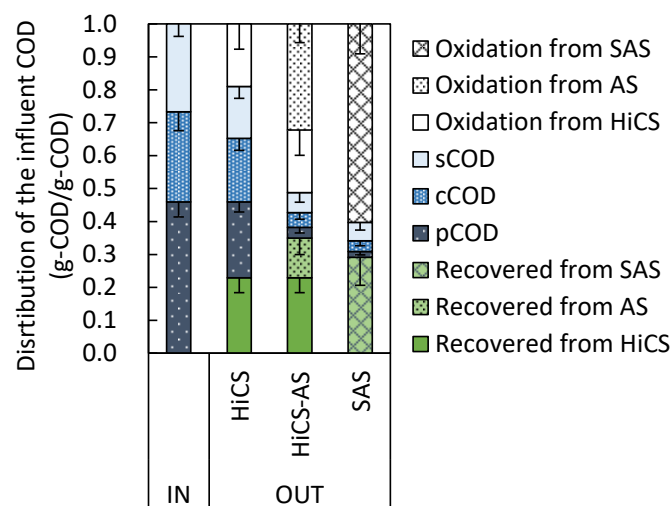


Figure 4. Redirection pathways of organic matter entering the HiCS, HiCS–AS, and SAS processes. HiCS–AS represents the combined HiCS and downstream AS processes. sCOD, cCOD, and pCOD denote the relative proportions in the influent or effluent. Error bars show standard deviations.

Table 4. Operational conditions and performance of each process treating primary effluent from municipal wastewater.

Process	WT (°C)	SRT (d)	CRR (g-COD/g-COD)	MRR (g-COD/g-COD)	Reference
HiCS	22 ± 5	0.60 ± 0.05	0.23 ± 0.04	0.13 ± 0.02	This study
HiCS–AS	22 ± 5 * 21 ± 5 **	0.60 ± 0.05 * 5.4 ± 0.4 **	0.34 ± 0.07	0.18 ± 0.04	This study
SAS	22 ± 5	6.2 ± 0.3	0.29 ± 0.08	0.13 ± 0.04	This study
HiCS	22 ± 5	0.44 ± 0.05	0.12 ± 0.03	0.09 ± 0.02	[13]
HiCS–AS	22 ± 5 * 21 ± 5 **	0.44 ± 0.05 * 5.7 ± 0.8 **	0.22 ± 0.06	0.15 ± 0.04	[13]
SAS	21 ± 4	5.8 ± 0.3	0.21 ± 0.09	0.11 ± 0.04	[13]
HiCS [†]	18 ± 1	0.98 ± 0.18	0.15 ± 0.04	0.078 ± 0.022	[41]
HiCS [†]	21 ± 0	0.49 ± 0.11	0.13 ± 0.06	0.070 ± 0.032	[41]
SAS [†]	23 ± 1	18 ± 4	0.15 ± 0.03	0.073 ± 0.014	[45]

Note: WT stands for water temperature. Asterisks (*) and double asterisks (**) represent the values of the HiCS and AS stages within the HiCS–AS process, respectively. The dagger (†) indicates that the process was operated in a continuous flow configuration, while processes without specific notation employed an SBR configuration.

3.5. Average Carbon Redirection and Harvesting Rates

This section summarizes the average carbon redirection and harvesting rates observed during the 16-week study, including a comparison with the previously reported SRT of 0.44 days to evaluate the impact of the modified SRT [13]. The carbon redirection and carbon harvesting rates are key indicators directly related to organic matter recovery [15,38]. A portion of the particulate and colloidal organic matter entering the HiCS process is adsorbed or settled and redirected to sludge. Redirected sludge increases in quantity by growth during its residence time in the system but decreases simultaneously through degradation or hydrolysis. The carbon redirection rate represents the mass of redirected sludge relative to the influent COD mass and corresponds to the sludge discharged as excess sludge or in the effluent; it is therefore also referred to as the sludge production rate [46]. The carbon harvesting rate represents the proportion of redirected sludge recovered through

sedimentation. The carbon harvesting rate is limited by the sedimentation performance of the sludge, among other factors [38].

The carbon redirection and carbon harvesting rates of the HiCS process are shown in Table 4. The carbon redirection rates of the AS and SAS processes were lower than the pCOD/COD ratio of the influent, indicating substantial degradation during retention in the reactors. In contrast, the carbon redirection rate of the HiCS process was almost identical to the pCOD/COD ratio of the influent, indicating minimal reduction. This suggests that the HiCS process minimized the degradation of particulate organic matter in the influent, and that any degraded portion was compensated for by newly formed sludge from colloidal and soluble organic matter. For carbon harvesting, the AS and SAS processes exhibited higher rates than the HiCS process, consistent with their superior settleability.

Table 5 also shows the results of a previous study [13] in which the HiCS process was operated at an SRT of 0.44 days. In the present study, the HiCS process (SRT of 0.60 days) showed an increase in MLSS concentration during the contact phase from 136 ± 24 mg/L to 316 ± 71 mg/L relative to the previous study; this can be explained by the extended SRT. This resulted in a notable improvement in CRR, with increased carbon redirection and carbon harvesting rates. The high carbon redirection rate was attributed to the slightly higher pCOD/COD ratio in the influent and the enhanced adsorption of particulate and colloidal organic matter as the MLSS concentration increased. The high carbon harvesting rate was likely due to the improved adsorption efficiency of particulate and colloidal organic matter and enhanced settleability associated with changes in the microbial community. Although both carbon redirection and carbon harvesting rates showed a correlation with water temperature during the AS process, neither showed a correlation with water temperature when using the HiCS process with SRTs of 0.60 and 0.44 days (Supplementary Figure S1).

Table 5. Key characteristics related to the carbon recovery rate (CRR) in each process.

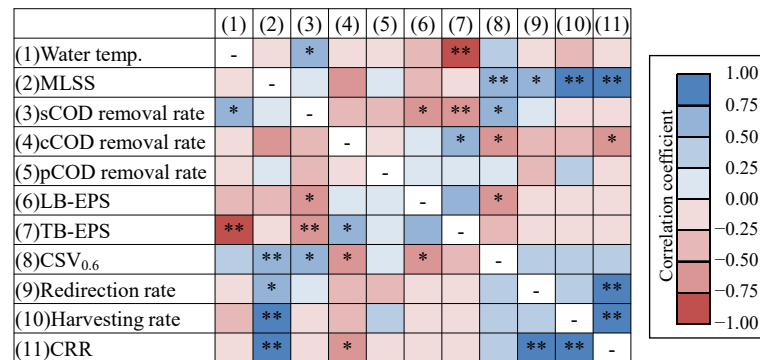
Processes	pCOD/COD in the Influent (g-COD/g-COD)	Carbon Redirection Rate (g-COD/g-COD)	Carbon Harvesting Rate (g-COD/g-COD)	CRR (g-COD/g-COD)
HiCS (0.60d, 22 °C)	0.46 ± 0.04	0.46 ± 0.05	0.50 ± 0.06	0.23 ± 0.04
AS (5.4d, 21 °C) *	0.40 ± 0.04	0.23 ± 0.09	0.79 ± 0.10	0.18 ± 0.07
SAS (6.2d, 22 °C)	0.46 ± 0.04	0.31 ± 0.08	0.94 ± 0.05	0.29 ± 0.08
HiCS (0.44d, 22 °C) **	0.40 ± 0.08	0.37 ± 0.04	0.33 ± 0.07	0.12 ± 0.03

Note: Values in parentheses denote SRT and average water temperature. * The AS process used HiCS effluent as influent. ** Values reported by Sakurai et al. [13].

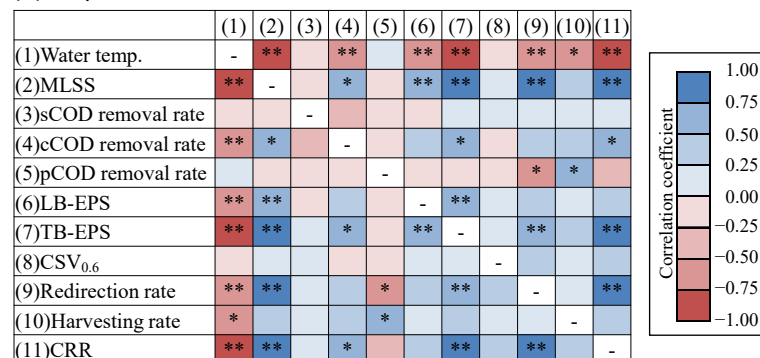
3.6. Correlation Between Water Temperature and Operational Parameters

Figure 5a shows the correlation between water temperature and operating parameters in the HiCS process. Water temperature exhibited a statistically significant positive correlation with the sCOD removal rate ($r = 0.60$, $p < 0.05$). The increasing trend in sCOD removal rate with rising water temperature aligned with previous findings [38] and was thought to result from enhanced microbial metabolic activity. Although reports have indicated that LB-EPS, which has a rapid production rate, increased with rising sCOD removal [11,40], no correlation between water temperature and LB-EPS was observed in this study (see Supplementary Figure S2). This is likely due to the balance between LB-EPS production and degradation in response to water temperature changes.

(a) HiCS process



(b) AS process



(c) SAS process

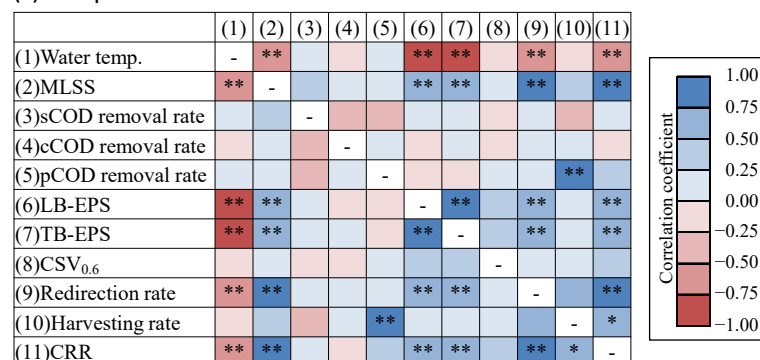


Figure 5. Heat maps showing Spearman's rank correlation coefficient between water temperature and operational parameters in the (a) HiCS, (b) AS, and (c) SAS processes. "CSV_{0.6}" indicates the proportion of pCOD in the contact or aeration phase where the critical settling velocity exceeds 0.6 m/h. (* $p < 0.05$, ** $p < 0.01$).

TB-EPS in the HiCS process showed a statistically significant negative correlation with water temperature ($r = -0.76$, $p < 0.01$); this trend is consistent with previous reports using the CAS process [47]. Since TB-EPS exhibits a slow production rate [11,31], its decomposition may have been strongly influenced by high temperatures. TB-EPS showed a positive correlation with cCOD removal rate ($r = 0.53$, $p < 0.05$). Previous studies have suggested that TB-EPS may promote the bioflocculation and sedimentation of colloidal substances [10], consistent with these results. The cCOD removal rate showed a negative correlation with CSV_{0.6} ($r = -0.60$, $p < 0.05$), which may indicate either that cCOD was adsorbed during bioflocculation or that EPS behavior negatively affected sedimentation rates. LB-EPS and CSV_{0.6} showed a negative correlation ($r = -0.61$, $p < 0.05$) consistent with previous reports indicating that excessive LB-EPS production causes deterioration in settleability [10,30,48]. No significant correlation was observed between pCOD removal

and any parameter. This may reflect the influence of various factors, such as settleable solids, in the influent and floc formation conditions.

The carbon redirection rate, carbon harvesting rate, and CRR in the HiCS process showed no significant correlation with water temperature, suggesting that changes in water temperature have no direct relationship with either sludge redirection or settling, and indicating the potential for stable carbon recovery. In summary, during the HiCS process, although water temperature changes were associated with microbial activity, no direct relationship with LB-EPS was observed. Additionally, no decrease in settleability caused by excessive LB-EPS was observed, and a stable CRR was achieved.

As shown in Figure 5b,c, water temperature exhibited a negative correlation with many parameters, including CRR, in both the AS and SAS processes. Similar to previous results [46], it was considered that a longer SRT promoted organic matter decomposition in sludge under high temperatures, leading to decreases in mixed liquor suspended solids (MLSS) and carbon redirection rate, resulting in a reduction in CRR.

3.7. Relationship Between Carbon Recovery Rate and Water Temperature in the HiCS–AS System

Figure 6 shows the relationship between water temperature and CRR. In the HiCS–AS and SAS processes, a tendency for CRR to decrease at higher temperatures was observed. Spearman's rank correlation coefficient for the HiCS process was -0.26 and was not found to be statistically significant at a p -value of 0.34 , suggesting that it is less susceptible to the influence of water temperature. In contrast, the HiCS–AS and SAS processes had Spearman's rank correlation coefficients of -0.65 and -0.66 , respectively, and both were statistically significant at p -values of <0.01 .

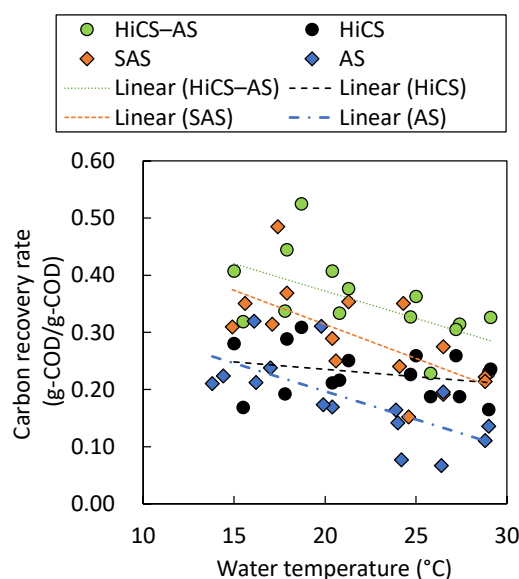


Figure 6. Relationships between water temperature and carbon recovery rate. HiCS–AS represents the combined HiCS and downstream AS processes. Trend lines are drawn for indicative purposes only.

The CRR of the HiCS process showed little influence from temperature, consistent with previous findings [13]. Additionally, no notable impact was observed during the transition period of water temperature changes caused by continuous operation. The stable recovery of HiCS sludge with high methane conversion rates contributes to the stable energy recovery efficiency in the HiCS–AS process. Similar temperature-dependent effects in these processes are expected in temperate regions.

4. Conclusions

To advance the practical application of the HiCS–AS process for enhanced energy recovery, this study evaluated a bench-scale SBR system during 16 weeks of continuous operation. By adopting an SRT of 0.60 days for the upstream HiCS stage, a stable CRR of 0.23 ± 0.04 g-COD/g-COD was achieved. This value is higher than the CRR reported in previous studies using an SRT of 0.44 days. A key finding of this research is the seasonal resilience of the HiCS stage; statistical analysis confirmed that its CRR was not significantly influenced by water temperature changes between 14 °C and 29 °C. While both the HiCS–AS and parallel SAS processes exhibited temperature-dependent trends, the average CRR of the HiCS–AS process (0.34 ± 0.07 g-COD/g-COD) was significantly higher than that of the SAS process (0.29 ± 0.08 g-COD/g-COD). Furthermore, the SBR configuration ensured consistent effluent quality, with an overall COD removal rate of $86\% \pm 3\%$, and maintained stable sludge sedimentation throughout the study. These results indicate that the HiCS–AS process demonstrated consistent performance and enhanced organic matter recovery relative to conventional processes under the conditions tested.

Supplementary Materials: The following supporting information can be downloaded at <https://www.mdpi.com/article/10.3390/w18141660/s1>: Figure S1: Relationships between (a) carbon redirection rate and (b) carbon harvesting rate and water temperature; Figure S2: Relationship between (a) LB-EPS and (b) TB-EPS and water temperature in the reaction tank for each process.

Author Contributions: K.S. and C.A. contributed to the study conception and design. Material preparation, data collection, analysis, and writing the first draft of the manuscript were performed by K.S. C.A. commented on the manuscript. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: The original contributions presented in this study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding author.

Acknowledgments: The authors express their sincere gratitude to the staff of the wastewater treatment plant for their valuable assistance with wastewater sampling.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

AS	Activated sludge
ATU-BOD	Allylthiourea-biochemical oxygen demand
BOD	Biochemical oxygen demand
CAS	Conventional activated sludge
cCOD	Colloidal chemical oxygen demand
COD	Chemical oxygen demand
CRR	Carbon recovery rate
CSV	Critical settling velocity
DO	Dissolved oxygen
EPS	Extracellular polymeric substances
HiCS	High-rate contact stabilization
HiCS–AS	High-rate contact stabilization–activated sludge
HRAS	High-rate activated sludge
HRT	Hydraulic retention time
LB-EPS	Loosely bound extracellular polymeric substances
MCR	Methane conversion rate

MLSS	Mixed liquor suspended solids
MRR	Methane recovery rate
NH_4^+-N	Ammonium nitrogen
NO_2^--N	Nitrite nitrogen
NO_3^--N	Nitrate nitrogen
pCOD	Particulate chemical oxygen demand
PST	Primary settling tank
SAS	Single activated sludge
SBR	Sequencing batch reactor
sCOD	Soluble chemical oxygen demand
SRT	Solids retention time
SVI	Sludge volume index
TB-EPS	Tightly bound extracellular polymeric substances
tCOD	Total chemical oxygen demand
TOF	Threshold of flocculation
TSS	Total suspended solids
VSS	Volatile suspended solids
WT	Water temperature
WWTP	Wastewater treatment plant

References

1. International Energy Agency. Water Energy Nexus (Excerpt from the World Energy Outlook). Available online: <https://www.iea.org/reports/water-energy-nexus> (accessed on 20 May 2026).
2. Maktabifard, M.; Zaborowska, E.; Makinia, J. Achieving energy neutrality in wastewater treatment plants through energy savings and enhancing renewable energy production. *Rev. Environ. Sci. Bio/Technol.* **2018**, *17*, 655–689. [\[CrossRef\]](#)
3. Liu, Z.; Xu, Z.; Zhu, X.; Yin, L.; Yin, Z.; Li, X.; Zheng, W. Calculation of carbon emissions in wastewater treatment and its neutralization measures: A review. *Sci. Total Environ.* **2024**, *912*, 169356. [\[CrossRef\]](#) [\[PubMed\]](#)
4. Zhang, X.; Hao, T.; Zhang, T.; Hu, Y.; Lu, R.; Li, D.; Pan, Y.; Li, Y.-Y.; Kong, Z. Towards energy conservation and carbon reduction for wastewater treatment processes: A review of carbon-neutral anaerobic biotechnologies. *J. Water Process Eng.* **2024**, *59*, 105026. [\[CrossRef\]](#)
5. Lima, D.; Li, L.; Appleby, G. A Review of renewable energy technologies in municipal wastewater treatment plants (WWTPs). *Energies* **2024**, *17*, 6084. [\[CrossRef\]](#)
6. Rahman, A.; De Clippeleir, H.; Thomas, W.; Jimenez, J.A.; Wett, B.; Al-Omari, A.; Murthy, S.; Riffat, R.; Bott, C. A-stage and high-rate contact-stabilization performance comparison for carbon and nutrient redirection from high-strength municipal wastewater. *Chem. Eng. J.* **2019**, *357*, 737–749. [\[CrossRef\]](#)
7. Meerburg, F.A.; Boon, N.; Van Winckel, T.; Vercamer, J.A.R.; Nopens, I.; Vlaeminck, S.E. Toward energy-neutral wastewater treatment: A high-rate contact stabilization process to maximally recover sewage organics. *Bioresour. Technol.* **2015**, *179*, 373–381. [\[CrossRef\]](#) [\[PubMed\]](#)
8. Jimenez, J.; Miller, M.; Bott, C.; Murthy, S.; De Clippeleir, H.; Wett, B. High-rate activated sludge system for carbon management—Evaluation of crucial process mechanisms and design parameters. *Water Res.* **2015**, *87*, 476–482. [\[CrossRef\]](#) [\[PubMed\]](#)
9. Jimenez, J.A.; La Motta, E.J.; Parker, D.S. Kinetics of removal of particulate chemical oxygen demand in the activated-sludge process. *Water Environ. Res.* **2005**, *77*, 437–446. [\[CrossRef\]](#) [\[PubMed\]](#)
10. Guthi, R.S.; Tondera, K.; Gillot, S.; Buffiere, P.; Boillot, M.; Chazarenc, F. A-Stage process—Challenges and drawbacks from lab to full scale studies: A review. *Water Res.* **2022**, *226*, 119044. [\[CrossRef\]](#) [\[PubMed\]](#)
11. Rahman, A.; Hasan, M.; Meerburg, F.; Jimenez, J.A.; Miller, M.W.; Bott, C.B.; Al-Omari, A.; Murthy, S.; Shaw, A.; De Clippeleir, H.; et al. Moving forward with A-stage and high-rate contact-stabilization for energy efficient water resource recovery facility: Mechanisms, factors, practical approach, and guidelines. *J. Water Process Eng.* **2020**, *36*, 101329. [\[CrossRef\]](#)
12. Dai, W.; Xu, X.; Yang, F. High-rate contact stabilization process-coupled membrane bioreactor for maximal recovery of organics from municipal wastewater. *Water* **2018**, *10*, 878. [\[CrossRef\]](#)
13. Sakurai, K.; Okayasu, Y.; Abe, C. Energy recovery from organic matter in municipal wastewater using a two-stage system with high-rate contact stabilization and activated sludge processes under seasonal water temperature variations. *Environ. Sci. Water Res. Technol.* **2025**, *11*, 1016–1025. [\[CrossRef\]](#)
14. Gori, R.; Giaccherini, F.; Jiang, L.M.; Sobhani, R.; Rosso, D. Role of primary sedimentation on plant-wide energy recovery and carbon footprint. *Water Sci. Technol.* **2013**, *68*, 870–878. [\[CrossRef\]](#) [\[PubMed\]](#)

15. Rahman, A.; Meerburg, F.A.; Ravadagundhi, S.; Wett, B.; Jimenez, J.; Bott, C.; Al-Omari, A.; Riffat, R.; Murthy, S.; De Clippeleir, H. Biofloculation management through high-rate contact-stabilization: A promising technology to recover organic carbon from low-strength wastewater. *Water Res.* **2016**, *104*, 485–496. [CrossRef] [PubMed]
16. United States Environmental Protection Agency. Wastewater Technology Fact Sheet Sequencing Batch Reactors. Available online: <https://www.epa.gov/system/files/documents/2022-10/sequencing-batch-reactors-factsheet.pdf> (accessed on 20 May 2026).
17. Gerardi, M.H. SBR Phases. In *Troubleshooting the Sequencing Batch Reactor*; John Wiley & Sons, Inc.: Hoboken, NJ, USA, 2010; pp. 17–28.
18. Rey-Martínez, N.; Barreiro-López, A.; Guisasola, A.; Baeza, J.A. Comparing continuous and batch operation for high-rate treatment of urban wastewater. *Biomass Bioenergy* **2021**, *149*, 106077. [CrossRef]
19. Versprille, A.I.; Zuurveen, B.; Stein, T. The A–B Process: A novel two stage wastewater treatment system. *Water Sci. Technol.* **1985**, *17*, 235–246. [CrossRef]
20. Metcalf & Eddy Inc. *Wastewater Engineering: Treatment and Resource Recovery*, 5th ed.; McGraw-Hill Education: New York, NY, USA, 2014.
21. Shao, Y.; Wang, Y.; Wang, H.; Liu, G.; Qi, L.; Xu, X.; Zhang, J.; Liu, S.; Sun, W. Effect of operating temperature on the efficiency of ultra-short-sludge retention time activated sludge systems. *Environ. Sci. Pollut. Res.* **2021**, *28*, 39257–39267. [CrossRef] [PubMed]
22. Mamais, D.; Jenkins, D.; Pitt, P. A rapid physical-chemical method for the determination of readily biodegradable soluble COD in municipal wastewater. *Water Res.* **1993**, *27*, 195–197. [CrossRef]
23. American Public Health Association; American Water Works Association; Water Environment Federation. *Standard Methods for the Examination of Water and Wastewater*, 23rd ed.; American Public Health Association: Washington, WA, USA, 2017.
24. Rahman, A.; Mosquera, M.; Thomas, W.; Jimenez, J.A.; Bott, C.; Wett, B.; Al-Omari, A.; Murthy, S.; Riffat, R.; De Clippeleir, H. Impact of aerobic famine and feast condition on extracellular polymeric substance production in high-rate contact stabilization systems. *Chem. Eng. J.* **2017**, *328*, 74–86. [CrossRef]
25. Van Winckel, T.; Ngo, N.; Sturm, B.; Al-Omari, A.; Wett, B.; Bott, C.; Vlaeminck, S.E.; De Clippeleir, H. Enhancing biofloculation in high-rate activated sludge improves effluent quality yet increases sensitivity to surface overflow rate. *Chemosphere* **2022**, *308*, 136294. [CrossRef] [PubMed]
26. Mancell-Egala, W.; De Clippeleir, H.; Su, C.; Takacs, I.; Novak, J.T.; Murthy, S.N. Novel stokesian metrics that quantify collision efficiency, floc strength, and discrete settling behavior. *Water Environ. Res.* **2017**, *89*, 586–597. [CrossRef] [PubMed]
27. Sakurai, K.; Okayasu, Y. Improvement of organic carbon recovery from wastewater by changing water temperature in high-rate contact stabilization process. *Int. J. Environ. Sci. Technol.* **2025**, *22*, 2127–2136. [CrossRef]
28. Haider, S.; Svardal, K.; Vanrolleghem, P.A.; Kroiss, H. The effect of low sludge age on wastewater fractionation (SS, SI). *Water Sci. Technol.* **2003**, *47*, 203–209. [CrossRef]
29. Sánchez, J.B.; Vuono, M.; Dionisi, D. Model-based comparison of sequencing batch reactors and continuous-flow activated sludge processes for biological wastewater treatment. *Comput. Chem. Eng.* **2021**, *144*, 107127. [CrossRef]
30. Li, D.; Gu, R.; Peng, F.; Yan, Z.; Song, Y.; Liu, T.; Liang, D.; He, W.; Feng, Y. Trade-offs between bio-floculation and oxidation: Carbon capture pathway governed by SRT in high-rate contact stabilization process. *Chem. Eng. J.* **2025**, *519*, 165080. [CrossRef]
31. Fu, S.; Li, D.; Fu, K.; Zhang, X.; Zhu, Y.; Yang, S.; Zeng, H.; Zhang, J. Towards a high-rate operation of contact stabilization process: A microscopic view of carbon capture properties. *Environ. Res.* **2024**, *263*, 120113. [CrossRef] [PubMed]
32. Fu, S.; Li, D.; Fu, K.; Zhu, Y.; Yang, S.; Zhang, X.; Zhang, J. Towards a high-rate operation of contact stabilization process: Challenges of flocculation and floc stability. *J. Environ. Manag.* **2024**, *370*, 123014. [CrossRef] [PubMed]
33. Jiang, Z.; Deng, M.; Qiu, S.; Fu, M.; Yuan, M.; Wen, Q.; Jia, X. Indicative impacts of sludge properties and biological metabolic characteristics on high-rate contact stabilization process performance. *J. Environ. Manag.* **2024**, *370*, 122569. [CrossRef] [PubMed]
34. Torfs, E.; Nopens, I.; Winkler, M.K.H.; Vanrolleghem, P.A.; Balemans, S.; Smets, I.Y. Settling tests. In *Experimental Methods in Wastewater Treatment*; van Loosdrecht, M.C.M., Nielsen, P.H., Lopez-Vazquez, C.M., Brdjanovic, D., Eds.; IWA Publishing: London, UK, 2016.
35. Mancell-Egala, W.; Kinnear, D.J.; Jones, K.L.; De Clippeleir, H.; Takacs, I.; Murthy, S.N. Limit of stokesian settling concentration characterizes sludge settling velocity. *Water Res.* **2016**, *90*, 100–110. [CrossRef] [PubMed]
36. Fau, Z.; Azais, A.; Gillot, S.; Chazarenc, F.; Derlon, N. Impact of slowly biodegradable COD and loosely bound polymeric substances accumulation in high-rate activated sludge: Implications for biofloculation and organic matter harvesting. *ACS EST Eng.* **2025**, *5*, 3632–3644. [CrossRef] [PubMed]
37. Cao, S.; Lu, D.; Phua, K.; Yan, W.; Le, C.; Tao, G.; Zhou, Y. Organics transformation and energy production potential in a high rate A-stage system: A demo-scale study. *Bioresour. Technol.* **2020**, *295*, 122300. [CrossRef] [PubMed]
38. Canals, J.; Cabrera-Codony, A.; Carbo, O.; Toran, J.; Martin, M.; Baldi, M.; Gutierrez, B.; Poch, M.; Ordonez, A.; Monclus, H. High-rate activated sludge at very short SRT: Key factors for process stability and performance of COD fractions removal. *Water Res.* **2023**, *231*, 119610. [CrossRef] [PubMed]

39. Hauduc, H.; Al-Omari, A.; Wett, B.; Jimenez, J.; De Clippeleir, H.; Rahman, A.; Wadhawan, T.; Takacs, I. Colloids, flocculation and carbon capture—A comprehensive plant-wide model. *Water Sci. Technol.* **2019**, *79*, 15–25. [[CrossRef](#)] [[PubMed](#)]
40. Song, M.; Chun, M.; Park, J.; Jeong, S.; Lee, T.; Shin, Y.-U.; Kim, J.; Bae, H. Effects of contact/stabilization time ratio on organic carbon-methane conversion in high-rate contact stabilization system. *J. Water Process Eng.* **2024**, *60*, 105247. [[CrossRef](#)]
41. Sakurai, K.; Abe, C. Enhanced biomethane production from organic matter recovered from municipal wastewater by a pilot-scale plant using continuous high-rate contact stabilization process. *Sci. Rep.* **2026**, *16*, 11078. [[CrossRef](#)] [[PubMed](#)]
42. Henze, M.; Gujer, W.; Mino, T.; van Loosdrecht, M. *Activated Sludge Models ASM1, ASM2, ASM2d and ASM3*; IWA Publishing: London, UK, 2006.
43. Heidrich, E.S.; Curtis, T.P.; Doling, J. Determination of the internal chemical energy of wastewater. *Environ. Sci. Technol.* **2011**, *45*, 827–832. [[CrossRef](#)] [[PubMed](#)]
44. McCarty, P.L.; Bae, J.; Kim, J. Domestic wastewater treatment as a net energy producer—Can this be achieved? *Environ. Sci. Technol.* **2011**, *45*, 7100–7106. [[CrossRef](#)] [[PubMed](#)]
45. Sakurai, K.; Abe, C. Enhanced organic matter recovery in the high-rate contact stabilization process by addition of waste activated sludge: A pilot-scale study. *Water* **2026**, *18*, 1127. [[CrossRef](#)]
46. Ekama, G.A.; Wentzel, M.C. Organic matter removal. In *Biological Wastewater Treatment: Principles, Modelling and Design*; Chen, G., van Loosdrecht, M.C.M., Ekama, G.A., Brdjanovic, D., Eds.; IWA Publishing: London, UK, 2023; pp. 111–160.
47. Sun, H.; Cai, C.; Chen, J.; Liu, C.; Wang, G.; Li, X.; Zhao, H. Effect of temperatures and alternating anoxic/oxic sequencing batch reactor (SBR) operating modes on extracellular polymeric substances in activated sludge. *Water Sci. Technol.* **2020**, *82*, 120–130. [[CrossRef](#)] [[PubMed](#)]
48. Li, X.Y.; Yang, S.F. Influence of loosely bound extracellular polymeric substances (EPS) on the flocculation, sedimentation and dewaterability of activated sludge. *Water Res.* **2007**, *41*, 1022–1030. [[CrossRef](#)] [[PubMed](#)]

Disclaimer/Publisher’s Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.