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Juhani Suvilampi

Aerobic Wastewater Treatment under High and Varying Temperatures

Thermophilic Process Performance and Effluent Quality

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UNIVERSITY OF JYVÄSKYLÄ

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ABSTRACT

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Industries, such as the pulp and paper industry, generate high-temperature process waters and wastewaters. Biological treatment at high temperatures may be an attractive option for the treatment of hot or concentrated wastewaters. The objective of this study was to evaluate the feasibility of thermophilic aerobic wastewater treatment in laboratory experiments, in which thermophilic processes were compared with mesophilic processes. A combined thermophilic-mesophilic treatment was used to improve thermophilic effluent quality. Also the performance of a pilot thermophilic aerobic suspended carrier biofilm process (SCBP) treating groundwood mill circulation water (GWM) was studied on mill premises. Thermophilic and mesophilic activated sludge processes (ASPs) were compared under different hydraulic retention times (HRTs) 18-8 h and volumetric loading rates (VLRs) 2.5-6 kg COD_{filt} $m^{-3}d^{-1}$. In thermophilic ASPs total COD (COD_{tot}) and GFA-filtered COD (COD_{filt}) removals were lower (65-75%) than in the mesophilic (85-90%). Both ASPs gave similar COD_{sol} (0.45 σm-filtrated COD) removals (90%), whereas effluents from the thermophilic ASPs had notably higher COD_{col} (calculated COD between 0.45-1.6 σm) values (460∂170 mg l^{-1}) than mesophilic effluents (6∂5 mg l^{-1}). The increased COD_{col} was due to the high density of dispersed particles, which were unable to aggregate and settle under thermophilic conditions. These bacteria were mostly non-viable gram-positive, whereas small (<1 σm) thermophilic gram-negative bacteria were dominant in the settled flocs. Combined thermophilic-mesophilic treatment had COD_{filt} removals of 80-85% with HRTs from 36 to 18 h. Thermophilic treatment removed COD_{sol} (90%), whereas mesophilic post-treatment removed free bacteria from the thermophilic effluent, measured as 90-100% COD_{col} removals. Between HRTs of 12-18 h the thermophilic ASP and SCBP removed 60∂13% and 62∂7% of COD_{filt}, respectively, whereas with HRT of 7-8 h the removals were 48∂1% and 69∂4%. A two-stage thermophilic SCBP produced 49-77% dissolved organic carbon (DOC) removals at VLRs of 3-14 kg COD_{filt} $m^{-3}d^{-1}$ and HRTs of 2-8 h, whereas the single reactor produced 40-67% DOC removals at VLRs of 7-28 kg COD_{filt} $m^{-3}d^{-1}$ and HRTs of 1-4 h. The two-stage thermophilic process was particularly resistant to process upsets. In conclusion, thermophilic aerobic wastewater treatment proved operable under varying parameters and yielded COD_{sol} removals comparable to those obtained from mesophilic treatment. Increased temperature reduced COD_{tot} and COD_{filt} removals.

Key words: Thermophilic; aerobic; wastewater treatment; activated sludge; suspended carrier biofilm; process performance; effluent quality.

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THE LIST OF ORIGINAL PUBLICATIONS

This thesis is a summary and discussion of the following articles and manuscripts, which are referred to by their Roman numerals in the text. The studies I, III, and IV were carried out with assistance of Annimari Lehtomäki, II with Tanja-Riitta Oksa. The experiments and methodologies were planned by Jukka Rintala and myself. The materials for all the papers were brought together, processed, and wrote by myself with a supervision of Jukka Rintala.

I Suvilampi, J. Lehtomäki, A. and Rintala, J. Comparative study of mesophilic and thermophilic activated sludge processes. Submitted.

II Suvilampi, J. and Rintala, J. Comparison of activated sludge processes at different temperatures: 35∀C, 27-55∀C, and 55∀C. Env. Technol. 23 (10), 1127-1134.

III Suvilampi, J., Lehtomäki, A. and Rintala, J. Comparison of laboratory-scale thermophilic biofilm and activated sludge processes integrated with a mesophilic activated sludge process. Accepted for publication in Bioresource Technology.

IV Suvilampi, J., Lehtomäki, A. and Rintala, J. Thermophilic-mesophilic wastewater treatment and biomass characterization. Submitted.

V Suvilampi, J., Rintala, J. and Nuortila-Jokinen, J. On-site aerobic suspended carrier biofilm treatment for pulp and paper mill process water under high and varying temperatures. Accepted for publication in Tappi Journal.

VI Suvilampi, J. and Rintala, J. Review – Thermophilic aerobic wastewater treatment, process performance and effluent quality. Submitted.

CONTENTS

ABBREVIATIONS

ASP	Activated sludge process	-
BOD_7	Biological oxygen demand	mg l^{-1}
COD	Chemical oxygen demand	mg l^{-1}
COD_{tot}	Total chemical oxygen demand	mg l^{-1}
COD_{filt}	Filtered chemical oxygen demand	mg l^{-1}
COD_{susp}	Suspended solids chemical oxygen demand	mg l^{-1}
COD_{col}	Colloidal solids chemical oxygen demand	mg l^{-1}
COD_{sol}	Soluble chemical oxygen demand	mg l^{-1}
DO	Dissolved oxygen	mg l^{-1}
DOC	Dissolved organic carbon	mg l^{-1}
GWM	Groundwood mill process water	-
EPS	Exocellular polymeric substances	-
HRT	Hydraulic retention time	h
MBBR	Moving bed biofilm reactor	-
MBR	Membrane bioreactor	-
MLSS	Mixed liquor suspended solids	g l^{-1}
MLVSS	Mixed liquor volatile suspended solids	g l^{-1}
N	Number of samples	-
SBR	Sequencing batch reactor	-
SCBP	Suspended carrier biofilm process	-
SLR	Sludge loading rate	kg COD_{filt} kg $VSS^{-1}d^{-1}$
SRT	Sludge retention time	d
SS	Suspended solid	mg l^{-1}
SVI	Sludge volume index	ml g^{-1}
TS-fix	Attached total solids	g l^{-1}
VLR	Volumetric loading rate	kg COD_{filt} m^{-3}d^{-1}
VS-fix	Attached volatile solids	g l^{-1}
VSS	Volatile suspended solids	mg l^{-1}
χ_c	SRT	d
V_r	reactor volume	l
V_s	sedimentation tank volume	l
Q_w	flowrate of wasted sludge	l d^{-1}
Q_e	effluent flowrate	l d^{-1}
X_f	concentration of SS in feed wastewater	g l^{-1}
X_r	concentration of SS in reactor	g l^{-1}
X_e	concentration of solids in the effluent	g l^{-1}
S_0	COD_{filt} in feed wastewater	g l^{-1}
S	COD_{filt} in effluent	g l^{-1}
$r_g^{'}$	net rate of microbial growth	g SS d^{-1}

1 INTRODUCTION

Biological treatment of hot industrials wastewaters and process waters under thermophilic conditions is an attractive alternative in many cases. The minimised need to use heat exchangers renders configuration of the process simpler, i.e. more cost-efficient and reliable. Thermophilic aerobic treatment is particularly suitable for operating as a high-rate wastewater treatment since the degradation rates achieved are higher than they are under mesophilic conditions, which in turn mean more compact reactor configurations (Jahren 1999, LaPara & Alleman 1999). Low sludge yield under thermophilic conditions has obvious benefits due to reduced sludge disposal and handling costs.

With increased production and the need to treat more wastewaters, an already existing mesophilic wastewater treatment plant could take the advantage of the high loading capacity of thermophilic treatment, by bringing it into partial operation at high temperatures and subsequently treating the remaining load under mesophilic conditions. In the pulp and paper industry, reduced water consumption and closed water cycles make thermophilic treatment attractive as a kidney, reducing thereby the amount of easily biodegradable substances, which without treatment would accumulate in water circuits and disturb the paper-making processes (Suvilampi et al. 1999, Huuhilo et al. 2001, Ramaekers et al. 2001, Jahren et al. 2002).

A number of aerobic thermophilic wastewater treatment processes treating different wastewaters under high VLRs, low HRTs, and resulting in high COD removals have been reported (Rintala & Lepistö 1993, Malmqvist et al. 1996, Ragona & Hall 1998, Becker et al. 1999, Jahren & Ødegaard 1999a, 1999b, Suvilampi et al. 1999, Huuhilo et al. 2002, Jahren et al. 2002, Rozich & Bordacs 2002). Many of these studies have focused on determining the feasibility of thermophilic aerobic wastewater treatment in the case of different industrial or synthetic wastewaters. Other studies have focused on comparing the operation of aerobic wastewater treatment under both mesophilic and thermophilic temperatures (Couillard & Zhu 1993, Barr et al. 1996, Tardif &

Hall 1997, Banat et al. 1999, Malmqvist et al. 1999, Tripathi & Allen 1999, Cibis et al. 2002, Vogelaar et al. 2002a, 2002b, Vogelaar et al. in press a). During these studies, with few exceptions, thermophilic aerobic wastewater treatment has proven to be a comparable alternative to mesophilic treatment. However, it is commonly held that operating biological wastewater treatment processes under higher temperatures leads to a deterioration in performance, measured as, e.g., lower COD removals and poorer sludge settling properties.

Thermophilic treatment has been claimed to have the advantage over mesophilic treatment in several aspects, e.g., higher loading rates, faster chemical reaction rates, faster microbial growth rates, lower net sludge yield, increased solubility of organics, increased removal of specific substrates, and increased destruction of pathogens (Brock 1986, Sundaram 1986, Schwarzenbach et al.1993, LaPara & Alleman 1999, Skjelhaugen 1999, Kosseva 2001, Rozich & Bordacs 2002). Laboratory-scale studies have demonstrated the truth of many of these claims (Tardif & Hall 1997, Becker et al. 1999, Banat et al. 1999, Bérubé & Hall 2000, LaPara et al. 2000b, Graham et al. 2000, Lim et al. 2001, Vogelaar et al. in press a, b). In addition, where the biological treatment has been augmented by an ultra- or nanofiltration unit, operating the membrane filtration unit under higher temperatures has provided a higher permeate flux (Tardif & Hall 1997, Suvilampi et al. 1999, Huuhilo et al. 2001, Huuhilo et al. 2002). The results appear to be case-dependent and may point to either thermophilic or mesophilic treatment as more beneficial.

Compared to the mesophilic aerobic processes, thermophilic aerobic processes are also thought to have some disadvantages, such as increased oxygen consumption along with a lower oxygen transfer rate, foaming problems, lower effluent quality, and poorer sludge settling characteristics (Sürücü et al. 1976, Tripathi & Allen 1999, LaPara et al. 2001, Vogelaar et al. 2002a). Higher effluent COD values and poor sludge settling properties under thermophilic conditions are well-established facts (Tripathi & Allen 1999, LaPara et al. 2001, Vogelaar et al. 2002a, 2002b). However, the reasons offered for high COD values and poor sludge settling properties vary from study to study. Tripathi & Allen (1999) and LaPara et al. (2001) suggested lower COD removal under thermophilic conditions is related to lower diversity of the thermophilic microbial population, the increase of complex substances at higher temperatures, and production of soluble microbial products from thermophilic bacteria lyses. Vogelaar et al. (2002a, 2002b) suggested that lower COD removal is due to colloidal matter in the initial wastewater and due to the erosion of flocs. Poor sludge settling properties are apparently due to poor floc formation. Possible reasons for poor floc formation under increased temperatures are the higher shear sensitivity of flocs leading to erosion of flocs, a decrease in the cell hydrophobicity, and the absence of floc-forming bacteria (Zita & Hermansson 1997, LaPara & Alleman 1999, Mikkelsen & Keiding 2002a).

Thermophilic organisms in wastewater treatment are representatives of both the *Archaebacteria* and *Eubacteria* kingdoms. *Eubacteria* are dominant in moderate (45-60ºC) thermophilic aerobic processes (Madigan et al. 1998). In

1999 LaPara & Alleman reviewed thermophilic aerobic wastewater treatment processes. Since then the microbial research in the field of thermophilic aerobic wastewater treatment has benefited from a wealth of new data, and several new strains have been isolated. In the recent thermophilic aerobic studies *Bacillus* species and beta-*Proteobacteria* have most often been isolated (LaPara et al. 2000a, LaPara et al. 2002, Lim et al. 2001, Kurisu et al. 2002, Tiirola et al. in press). A distinct difference in microbial diversity between thermophilic and mesophilic processes has also been shown (LaPara et al. 2001, Vogelaar et al 2002b). Floc formation under thermophilic conditions has been reported by, e.g., Tripathi & Allen (1999) and Vogelaar et al. (2002b).

2 OBJECTIVES

The main objective of this study was to evaluate the feasibility of thermophilic aerobic wastewater treatment. Laboratory experiments were conducted to compare the performance of mesophilic and thermophilic processes, measured as different COD removals under different operational conditions, such as volumetric loading rates (VLRs) and hydraulic retention times (HRTs), and as biomass characteristics. In addition, different pilot-scale feasibility studies were conducted to assess the performance of the thermophilic aerobic suspended carrier biofilm process (SCBP) on groundwood mill premises.

The main objective can be divided into:

- comparison of the performance of mesophilic and thermophilic activated sludge process (ASP) in different COD removals and in the use of a cation-based polymer to evaluate a possible method of promoting thermophilic floc formation and improving thermophilic effluent quality (I)
- comparison of ASP performance under constant mesophilic (35°C) and thermophilic (55°C) conditions, and at varying temperatures (27-56°C) (II)
- comparison of continuously operated laboratory-scale combined aerobic thermophilic – mesophilic wastewater treatments and the performance of thermophilic suspended carrier process (SCBP) and ASP (III)
- comparison of biomass characteristics and floc formation in continuously operated laboratory-scale aerobic thermophilic-mesophilic treatments (IV)
- evaluation of the feasibility of a pilot-scale aerobic SCBP under prevailing pulp mill conditions, e.g., elevated and variable process water temperatures and varying water characteristics (V)

3 MATERIALS AND METHODS

Experiments presented in this thesis were conducted in the laboratory (I-IV) and on mill premises (V).

3.1 Wastewaters

In the laboratory experiments diluted molasses was used as a synthetic wastewater. The molasses was purchased from Lännen Tehtaat Ltd., Iso-Vimma, Säkylä, Finland. Molasses was selected for laboratory use due its high organic load (COD_{filt} 800 000 - 1000 000 mg l^{-1}), yielding a high volume of diluted wastewater which nonetheless had a COD level of 2000 mg l^{-1}. The characteristics of the diluted molasses (1:300 - 1:600) are presented in Table 1. Feed wastewater was prepared 2-3 times in week in a feed container which was kept at 4°C under nitrogen atmosphere (I, III, IV) or at ambient room temperature (II). The diluted molasses acidified rapidly when exposed to room temperature, which led to varying pH values in the feed entering reactors and, subsequently to daily maintenance of the feed lines. Nutrients NH_4Cl and K_2HPO_4 were added into the feed container to obtain COD:N:P ratio of 200:5:1.

TABLE 1 Characteristics of diluted molasses (I-IV). Samples from the feed lines before entry into reactors

	unit	N	Average ∂ sd	Range
Dilution factor			1:500	1:600 - 1:300
$\mathrm{COD}_{\mathrm{tot}}$	mg l^{-1}	59	2 060∂210	1 700 – 2 400
$\mathrm{COD}_{\mathrm{filt}}$	mg l^{-1}	132	1 780∂275	1 200 – 2 700
$\mathrm{COD}_{\mathrm{susp}}$	mg l^{-1}	41	240∂160	50 – 700
$\mathrm{COD}_{\mathrm{col}}$	mg l^{-1}	16	70∂25	50 – 100
$\mathrm{COD}_{\mathrm{sol}}$	mg l^{-1}	21	1660∂180	1 400 – 2 000
SS	mg l^{-1}	91	160∂110	30 – 470
VSS	mg l^{-1}	79	150∂90	30 – 450
TS	g l^{-1}	12	1.8∂0.4	1.3 – 2.4

Pilot-scale experiments were conducted in a groundwood mill (UPM-Kymmene Ltd, Kaukas, Lappeenranta, Finland), where the wastewater was groundwood mill circulation water (GWM), obtained from the grinder whitewater pipe system. GWM also contained varying amounts of whitewater from the paper machine. The wastewater chemical characteristics showed seasonal variation and the physical characteristics varied on daily basis. The characteristics of the water after 1-mm pre-screening are presented in Table 2. A screen was installed in the feed line to avoid occasional fibre washouts to the reactors from the whitewater circuit.

TABLE 2 Characteristics of GWM after 1-mm pre-screening (V)

		Spring trial I			Autumn trial II		
	unit	N	Average ∂ sd	Range	N	Average ∂ sd	Range
Temperature	°C	81	60∂ 5	38 – 68	55	62 ∂ 3	52 – 70
SS	mg l^{-1}	18	460∂270	250 – 1 300	22	170 ∂ 40	120 – 270
VSS	mg l^{-1}	18	440∂ 260	230 – 1 270	22	150 ∂ 20	110 – 190
DOC	mg l^{-1}	32	430∂ 90	250 – 600	42	500 ∂ 80	350 – 720
$\mathrm{COD}_{\mathrm{tot}}$	mg l^{-1}	9	1300∂260	800 – 1 600	-	nd.	
$\mathrm{COD}_{\mathrm{filt}}$	mg l^{-1}	9	1100∂250	690 – 1 500	11	1430 ∂ 170	1 100 – 1 750
$\mathrm{BOD}_{7\,\mathrm{filt}}$	mg l^{-1}	9	650∂140	430 – 900	6	770 ∂ 80	740 – 870
Lignin-like compounds, $\mathrm{COD}_{\mathrm{filt}}$	mg l^{-1}	6	340∂56	270 – 400	6	410 ∂ 35	360 – 440
Sugars, $\mathrm{COD}_{\mathrm{filt}}$	mg l^{-1}	-	nd.		6	330 ∂ 50	285 - 405

N = number of samples.

3.2 Inoculums

All inoculums used as seed sludge were collected from mesophilic full-scale ASP plants treating pulp and paper mill wastewaters (Kaipola (III, IV), Kaukas (V), and Jämsänkoski (I, II) pulp and paper mills, UPM-Kymmene Ltd., Finland). Inoculum MLSS varied between 5 and 14 g l^{-1} and MLVSS between 4 and 12 g l^{-1}. Reactors were inoculated either at the ambient room temperature (20°C: I-IV) or at operating temperature (50-55°C: V). In the thermophilic reactors the temperature was increased rapidly after seeding performed at room temperature, and thermophilic conditions (temperature >50°C) were achieved within 18 h in all laboratory studies. In the second study (II) inoculum was first introduced into diluted molasses wastewater for 21 d, then mixed, after which the temperature in thermophilic reactor was increased from 35°C to 55°C.

3.3 Experimental set-up

3.3.1 Activated sludge and suspended carrier biofilm processes

ASPs and a SCBP were used in the laboratory experiments; the liquid volumes of the reactors varied from 1.5 l (I, III, IV) to 8 l (II). All the laboratory processes were ASPs, except in III and IV one of the processes was SCBP. The ASPs had separate settling units and sludge recirculation ratio to feed flow from settling unit to aeration unit was between the ratios 1:1 to 3:1. The SCBP had no settling unit; the biofilm carriers were KMT carriers (Kaldnes Miljøteknologi) (Fig. 1), which were filled to 50% of the reactor volume, providing a specific carrier surface area of 250 m^2m^{-3}. All the reactors and settling units were placed in temperature-controlled water baths.

The thermophilic processes were kept at 55°C, the mesophilic processes at 20-35°C, and one ASP was operated under a temperature fluctuating from 27 to 56°C (II). Aeration was provided with aquarium aeration stones connected to aquarium aerators (I, III, IV) or to an air-pressured line (II). All the reactors were covered with aluminium foil to prevent or to minimise evaporation. Evaporation in the mesophilic and thermophilic processes was 0-10% and 10-20%, respectively. Due to the daily variation in evaporation and its negligible effect on effluent COD values, the COD values were not corrected for evaporation.

SRT varied in all processes and in all studies between 1 and 18 d, due to the influence of high effluent SS values. SRT was controlled not to exceed 15 d (I, III, IV) or 18 d (II) by decanting a measured volume of mixed liquor from the aeration unit. SRT was calculated according to the mass of solids in the reactor and in the effluent, except in the second study (II), where solids in both the reactor and settling unit volumes were taken into account.

Feed samples were taken from the feed line before entry into the reactors. Composite (12 – 24 h) effluent samples were collected in effluent containers, except in the case thermophilic ASP and thermophilic SCBP placed before mesophilic ASP, where samples were grabbed before mesophilic processes (III, IV). Reactors were started immediately at their operating temperatures, except in the second study (II), where the reactors were first run at 35°C for 21 d.

Subsequently, the contents of the three reactors were combined and mixed and then re-divided equally among all three reactors to ensure the presence of a similar microbial population in each reactor.

In the pilot trials two serial aerobic reactors (referred to as R1 and R2 in the case of single reactors and R12 in the case of two-stage reactor) of total volume 2 m^3 were run in 37-day (referred as trial I, from April to May 1999) and 63-day (trial II, from October to November 1999) trials. The reactors were filled to 50% of volume with carrier elements (Flootek RF 438, Fig. 1) of near water density, which provided a total surface area of 190 m^2m^{-3} in both reactors. The RF 438 carriers, in 36 mm length and in 44 mm diameter, were made of recycled polyethylene and had two internally crossed fins. The carriers had been used before and apparently contained some attached biomass. The pilot plant was a Floobed® unit (Fig. 5), where pH adjustment, temperature measurement, nutrient addition, and wastewater sampling were performed automatically.

In the first reactor (R1), pH was adjusted to 7.0 with 50% NaOH-solution, and technical urea and phosphoric acid were added to maintain a COD:N:P ratio of 100:5:1 in trial I and 150:5:1 in trial II until day 37 and 150:4:1 thereafter. In trial I a 250 l lamella settling unit with sludge recirculation was used; in trial II neither sludge settling nor recirculation was applied. The reactors were aerated with an air-blower to exceed 3.0 mg l^{-1} of DO in R1. Manually controlled valves were used to maintain an airflow sufficient to keep the carriers in movement in the reactors. Composite samples (24 h) from the feed line after screening, effluent from R1, and final effluent from R2 were taken for analysis.

FIGURE 1 Biofilm carriers used in suspended carrier biofilm processes. Left: Kaldnes KMT carrier used in laboratory, diameter 10 mm. Right: Flootek RF 438 carriers used in pilot studies, diameter = 44 mm.

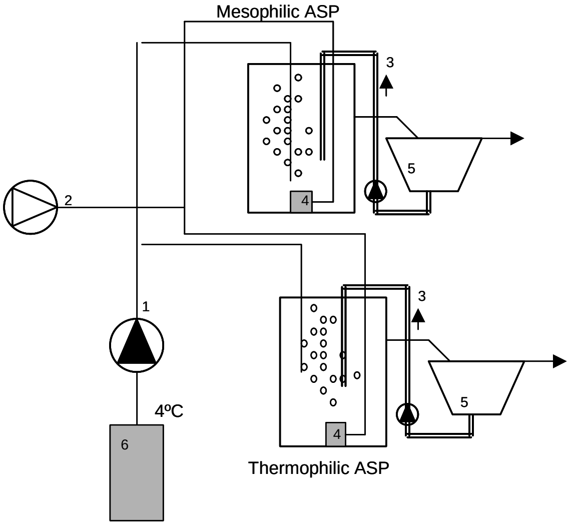

FIGURE 2 Experimental set-up in laboratory study of thermophilic and mesophilic ASP. (1) feed pump; (2) aeration pump; (3) sludge return line; (4) aeration stone; (5) settling unit; (6) refrigerator. (I).

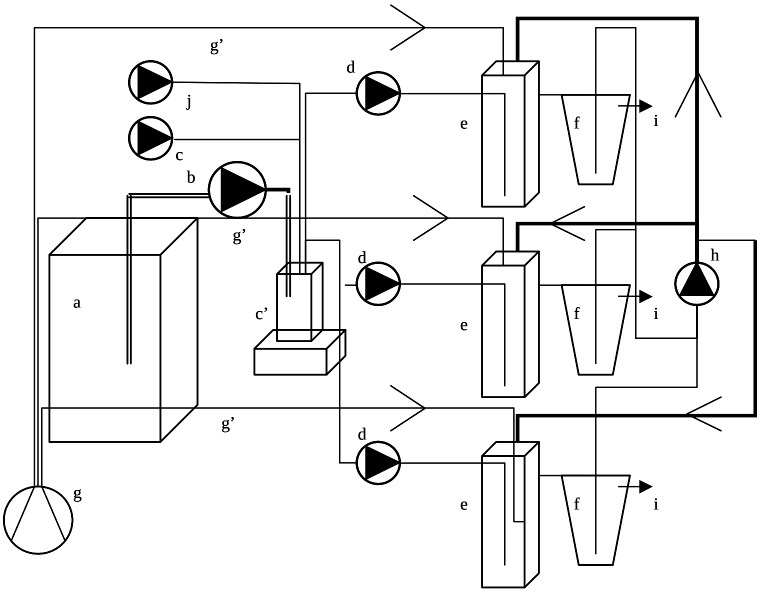

FIGURE 3 Experimental set-up in laboratory study of comparative study of thermophilic, mesophilic, and fluctuating temperature ASP. (a) feed container 200 l; (b) feed pump; (c) NaOH addition, (c') pH adjustment, (d) peristaltic pumps; (e) reactors 8 l; (f) sedimentation tanks 8 l (1.5 l after day 60); (g) air pump; (g') airflow; (h) sludge recycling pump; (i) clarified effluent; (j) nutrient addition. (II).

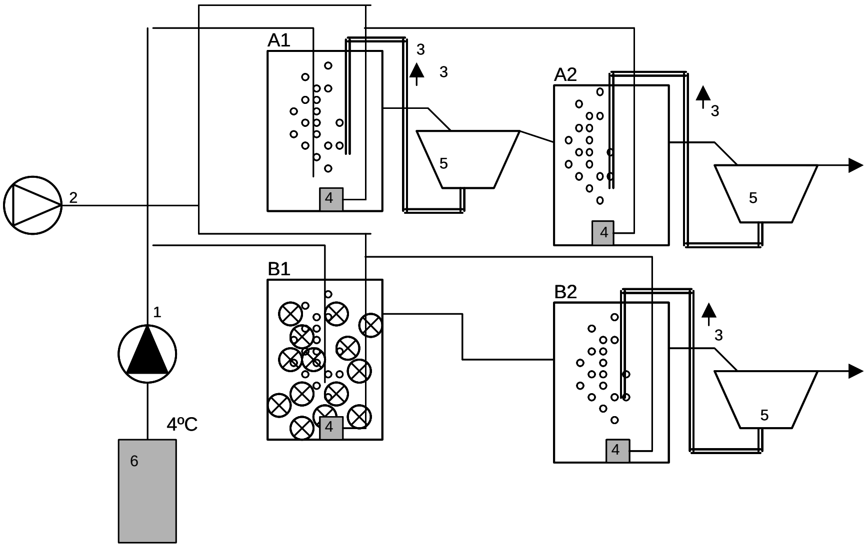

FIGURE 4 Experimental set-up in laboratory study of combined thermophilic-mesophilic ASP-ASP and thermophilic-mesophilic SCBP-ASP. (1) feed pump; (2) aeration pump; (3) sludge return line; (4) aeration stone; (5) settling unit; (6) refrigerator. (III).

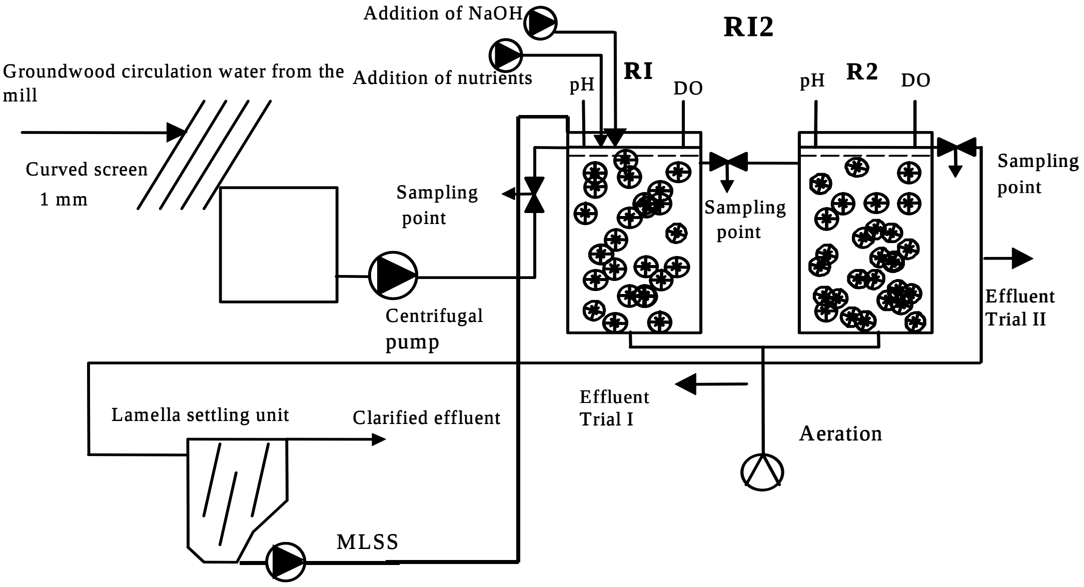

FIGURE 5 Experimental set-up in pilot study of two-stage thermophilic aerobic SCBP (V).

3.3.2 Batch experiments

Post-aeration studies were conducted in batch tests, in which aeration was provided by Rena aquarium-aerators connected to aeration stones. Duplicate 2 l Pyrex bottles and 2 l plastic vessels (covered with aluminium foil) were used (I and II). COD values were corrected for detected water evaporation (10% of volume at 35\C and 20% at 55∀(I); no evaporation was detected (II)). One assay was done at both temperatures with polymer addition of 4.5 ml l^{-1} (PAX-18, 3%) and without polymer (I), samples from 0 and 24 h were GF/A and 0.45 σm – filtrated, and analysed for COD_{tot}, COD_{susp}, COD_{col}, and COD_{sol} (I). The floc size distribution and the density of the free bacteria were also determined (I). In the second assay samples from 0, 4, 12, and 24 h were GF50-filtered and analysed for COD_{filt} (II).

3.4 Calculations

3.4.1 Sludge Yield

Sludge yields were calculated using two different methods: one for the determination of daily sludge yield (3-1) and one for the determination of net sludge production over a longer period (3-2), thereby assuring that the effect of daily variations was minimised. The first equation was taken from Tchobanoglous & Burton (1991), as follows:

$$\frac{V_r r_g^{'}}{Q(S_0\ 4\ S)} \mid \frac{dX_r}{dt} V_r\ 4\ QX_f\ 2\ QX_e \qquad (3\text{-}1)$$

Where $\frac{V_r r_g^{'}}{Q(S_0\ 4\ S)}$ = reactor volume and the net rate of microbial growth (g SS d^{-1}), divided by net substrate removal (g $COD_{filt\ removed}$)

$\frac{dX_r}{dt} V_r$ = rate of change of suspended solids (g SS) concentration in the reactor measured in terms of g SS l^{-1} unit volume x time (d)

QX_f = flowrate l d^{-1} x concentration of suspended solids (g SS) in the feed wastewater

QX_e = flowrate l d^{-1} x concentration of suspended solids (g SS) in the effluent wastewater

In the net sludge yield measured during longer periods, the change in microbial concentration in the reactors, ($\frac{dX}{dt} V_r$), was considered negligible (0), which gave the following equation:

$$\frac{V_r r_g^{'}}{Q(S_0\ 4\ S)} \mid 4QX_f\ 2\ QX_e \qquad (3\text{-}2)$$

3.4.2 SRT

Solids retention time, i.e., sludge age or mean cell residence time was calculated for the single reactors (I) and for the combined treatment lines (III), according to Tchobanoglous & Burton (1991), as follows:

$$\chi_c \mid \frac{V_r X_r}{Q_w X_r \; 2 \; Q_e X_e} \tag{3-3}$$

Where χ_c = SRT (d)
V_r = reactor volume (l)
X_r = concentration of SS in reactor (g l^{-1})
Q_w = flowrate of wasted sludge (l d^{-1})
Q_e = effluent flowrate (l d^{-1})
X_e = concentration of SS in the effluent (g l^{-1})

In the combined treatment systems the total reactor volume was the sum of the two reactors in the treatment line and the reactor solids concentration was the mean value of the two reactors (III). In one study the equation was modified to include solids in the sedimentation tanks (II), giving:

$$\chi_c \mid \frac{V_r X_r \; 2 \; V_s X_s}{Q_w X \; 2 \; Q_e X_e} \tag{3-4}$$

Where V_s = volume of sedimentation tank (l)
X_s = solids concentration in sedimentation tank (g l^{-1})

3.5 Analyses

3.5.1 COD, lignin-like and sugar COD, and DOC

Chemical oxygen demand (COD_{Cr}) was analysed according to SFS 5504 (Finnish Standard Association 1988). In the COD_{Cr} analyses potassium dichromate is used as an oxidizer, samples are boiled at 150\C for two hours with sulphuric acid. After boiling the samples, iron sulphate solution is added until a change in colour is observed. For process follow-up and performance total COD (COD_{tot}) and filtered COD (COD_{filt}) (also referred as SCOD in Paper II) typically were analysed, and for COD_{filt} both Schleicher and Schuell GF50 and Whatman GF/A filters were used. Both filters have an approximate pore size of 1.6 σm. Soluble COD (COD_{sol}) samples were filtrated with a 0.45 σm

Schleicher and Schuell membrane filter and pre-filtered with GF/A or GF50 filters. COD was also characterised into more detailed fractions, such COD$_{tot}$, suspended COD (COD$_{susp}$), colloidal COD (COD$_{col}$), and COD$_{sol}$, of which COD$_{susp}$ and COD$_{col}$ were calculated values. COD$_{susp}$ is the difference between COD$_{filt}$ and COD$_{tot}$ and COD$_{col}$ is the difference between COD$_{sol}$ and COD$_{filt}$.

Lignin and other aromatic compounds were determined by ultraviolet absorbency at 280 nm (UV$_{280}$) according to Rintala & Lepistö (1992). Samples were diluted to an absorbency of less than 0.8. The amount of soluble lignin-like substances measured as COD$_{filt}$ was estimated with an absorbtivity coefficient of 22.3 l g^{-1}cm^{-1} and oxygen demand of 1.9 g O_2 g^{-1} lignin (Sierra-Alvarez et al. 1991). Sugars were measured by the Antron method (UV$_{620}$), which determines the cellulose and hemicellulose content of the sample, by changing both of these into monosaccharides, which react with antron under high temperature conditions and turn green in colour. The intensity of the green colour indicates the sugar content. Sugar content (mg l^{-1}) was changed to DOC (mg l^{-1}) with a factor of 0.4 and subsequently to COD (mg l^{-1}) with a factor of 2.7.

Dissolved organic carbon (DOC) was measured with a Shimadzu TOC-5050A Total Organic Carbon analyser. Samples were GF/A filtered and diluted with deionised and 0.2 σm-filtrated Millipore water so as not to exceed total carbon (TC) values of 700 mg l^{-1}. The analyser measures the total carbon and inorganic carbon present in the sample. Organic carbon is the difference between total carbon and inorganic carbon. A high correlation ($r^2=0.98$) was found between the DOC and COD$_{filt}$ of the feed samples (feed COD$_{filt}$ = 2.6 x DOC) and hence the DOC values were corrected for COD$_{filt}$ values.

3.5.2 Solids analyses

Total solids, (TS) and volatile solids (VS) were analysed according to *Standard Methods* (APHA 1998). Suspended solids (SS), volatile suspended solids (VSS), mixed liquor SS (MLSS), and MLVSS were measured according to *Standard Methods* (APHA 1998) using Schleicher and Schuell GF50 and Whatman GF/A filters (approximate pore size 1.6 σm). In the laboratory-scale experiment (III) total solids (TS-fix) as attached biomass in SCBP was measured by weighing 50 dried (4 h at 105∀C) carriers from the reactor and 50 unused carriers, TS-fix is the difference between a clean and used carrier multiplied by the amount of carriers (950) in one litre. TS-fix and VS-fix in the pilot-scale experiments (V) were analysed by weighing 6 to 8 carriers after drying them at 105∀C for 4 h, scraping off the bulk of the biomass and brushing off the remainder in a soap solution, then weighing the carriers again after 4 h at 105∀C. TS-fix is the difference between a clean and used carrier multiplied by the amount of carriers (6.25) in one litre. The scraped biomass was used to determine the VS/TS –ratio and to calculate VS-fix.

3.5.3 Other analyses

Dissolved oxygen (DO), pH, and temperature in the reactors were measured typically on a daily basis. In the laboratory studies DO was measured using a YSI Jenway 9300 DO-meter, pH and temperature were measured using a Hanna Instrument 6028 pH meter. The sludge volume index (SVI) was measured in 250-500 ml graduated glasses by settling samples, with known SS, for 30 minutes. BOD_7-ATU was measured according to SFS 3019 (Finnish Standard Association 1979).

3.6 Microbial analyses

3.6.1 Phase-contrast microscopy

A phase-contrast microscope (Olympus 1 x 70, 60 to 150-fold magnification) was used to estimate filamentous bacteria density, floc size distribution, and the presence of dispersed particles, such as free bacteria, according to Table 6 (I, IV). Also, microbial diversity and floc morphology were evaluated during the phase-microscopy. Floc sizes were measured with the help of an eyepiece micrometer, and the flocs were classified into four sizes: <50 σm, 50-150 σm, 150-500 σm, and >500 σm. Samples were taken on a weekly basis from the feed wastewater, all the reactors, and from the effluents. The inoculum was also examined.

3.6.2 Fluorescence microscopy

A fluorescence microscope (Leitz DM RBE, 1000-fold magnification) was used to estimate the relative quantities of gram-positive and -negative, as well as viable and dead bacteria (IV). Samples were taken weekly from the biofilm and from the ASP settling units. Two samples were taken from each settling unit, suspended sludge was taken from the surface and settled sludge from the bottom. Samples were first treated ultrasonically (High Intensity Ultrasonic Processor, 375 Watt Model, Sonics & Materials, Inc.) to disintegrate the floc or biofilm structure and then stained with a ViaGram™ Red^+ Bacterial Gram Strain and Viability Kit (Molecular Probes Inc.). The kit differentiates gram-positive and gram-negative bacterial species and distinguishes viable from dead cells on the basis of their plasma membrane integrity. Stained samples were filtered through 0.2 µm black polycarbonate filters (Nucleopore). With the aid of the SPOT Advanced 3.1 software, the bacteria on the filter were photographed and divided into three size classes; <1 σm, 1-5 σm, and >5 σm.

TABLE 3 Scales used to estimate of the density of filamentous organisms (according to Jenkins et al. 1993), density of free bacteria, and density of higher organisms (modified from Eikelboom & van Buijsen 1983) in the phase-contrast microscopy. (I, IV).

Value	Description
	Filamentous organisms
0	No filamentous bacteria present in sample
1	Filamentous bacteria present, but only observed in an occasional floc
2	Filamentous bacteria commonly observed, but not present in all flocs
3	Filamentous bacteria observed at low density in all flocs (1-5 filamentous bacteria per floc)
4	Filamentous bacteria observed at medium density in all flocs (5-20 per floc)
5	Filamentous bacteria observed at high density in all flocs (>20 per floc)
6	Filamentous bacteria present in all flocs - more filament than floc and / or filamentous bacteria growing in high abundance in bulk solution
	Free dispersed cells
0	No free cells in sample
1	Low density; few free cells per field of view
2	Medium density; some tens per field of view
3	High density; hundreds per field of view
4	Extremely high density; thousands per field of view
	Higher organisms
0	No organisms in sample
1	Low density; random occurrence
2	Medium density; few organisms (5-10) per preparation
3	High density; > 10 organisms per preparation
4	The organism is a dominant species

3.7 Statistical analyses

Statistical analyses were performed to investigate the differences between two samples, two sample groups, or correlations with operational parameters. Microsoft® Excel 2000 was used for one-way analyses of variance (one-way ANOVA), which is an extended version of the t-test. Linear regression analysis was used for evaluation of the correlations.

4 RESULTS AND DISCUSSION

4.1 Comparison of thermophilic and mesophilic ASP

The performance, effluent quality, and biomass characteristics of the thermophilic and mesophilic ASPs were compared in two laboratory studies (I, II). In the first study (I) two ASPs were run, one at 35°C and the other at 55°C, with a constant HRT of 12 h corresponding to VLR of 3.2-4.0 kg COD_{filt} $m^{-3}d^{-1}$ (Fig. 6, Table 4) (I). In the latter study, three ASPs were first operated at 35°C for 21 d, after which reactor contents were mixed to ensure a similar microbial population in all reactors. After mixing the reactor contents, reactors were run at 35°C, 55°C, and under a fluctuating (27-56°C) temperature (II). In both runs a rise in temperature to 55°C occurred within 18 h. In the latter study HRT was gradually decreased in both the mesophilic and thermophilic ASPs from 18 to 3.5 h, corresponding to an increase in VLR from 2.4 to 9.7 kg COD_{filt} $m^{-3}d^{-1}$ (Fig. 6, Table 4) (II). The results for the fluctuating temperature ASP are given in a different section (4.1.3). The effect of polymer dosing on thermophilic and mesophilic ASP operation was evaluated in one study (I), in which the reactors were first run without (days 1-29) and then with polymer dosing (days 30-42) (I).

4.1.1 Thermophilic and mesophilic ASP performance

In both the comparative studies the thermophilic and mesophilic ASPs were operated under similar HRTs and VLRs (Fig. 6), whereas the sludge loading rates (SLRs) were lower in the mesophilic ASPs due to higher MLSS (Tables 4 and 8) (I, II). Throughout the runs the mesophilic ASPs gave higher (85-90%) COD_{filt} removals than did the thermophilic ASPs (67-74%) (Fig. 6). Thermophilic ASP COD_{filt} removals showed more variation (Table 4) and were intermittently almost as high as these from the mesophilic ASPs (Fig. 6). In the latter study the thermophilic and mesophilic ASPs experienced a short-term starvation period (days 45-47) due to technical malfunction (II), which caused a drop in thermophilic ASP $\mathrm{COD_{filt}}$ removal to 30% while mesophilic ASP showed no reduction in $\mathrm{COD_{filt}}$ removals (Fig. 6 b). Reducing HRT from 12 to 8 h (corresponding to a increase in VLR from 3.5 to 5.7 kg $\mathrm{COD_{filt}}$ $\mathrm{m^{-3}d^{-1}}$) decreased mesophilic ASP $\mathrm{COD_{filt}}$ removals to some extent, whereas in the thermophilic ASP they remained. However, under the highest VLR (10 kg $\mathrm{COD_{filt}}$ $\mathrm{m^{-3}d^{-1}}$) the thermophilic ASP experienced process failure, which was seen as lowered pH (5) and poor (19%) $\mathrm{COD_{filt}}$ removal (Fig. 6) (II). This was apparently due to a markedly high SLR (>17 kg $\mathrm{COD_{filt}}$ kg $\mathrm{SS^{-1}d^{-1}}$, Table 4) due to low MLSS (0.6 g $\mathrm{l^{-1}}$). The effect of polymer dosing on the thermophilic and mesophilic ASP $\mathrm{COD_{filt}}$ removals was negligible (Table 4). However, with polymer dosing, both the thermophilic and mesophilic ASP MLSS values showed some increase (Table 8), which can be seen in the lower SLR values (Table 4).

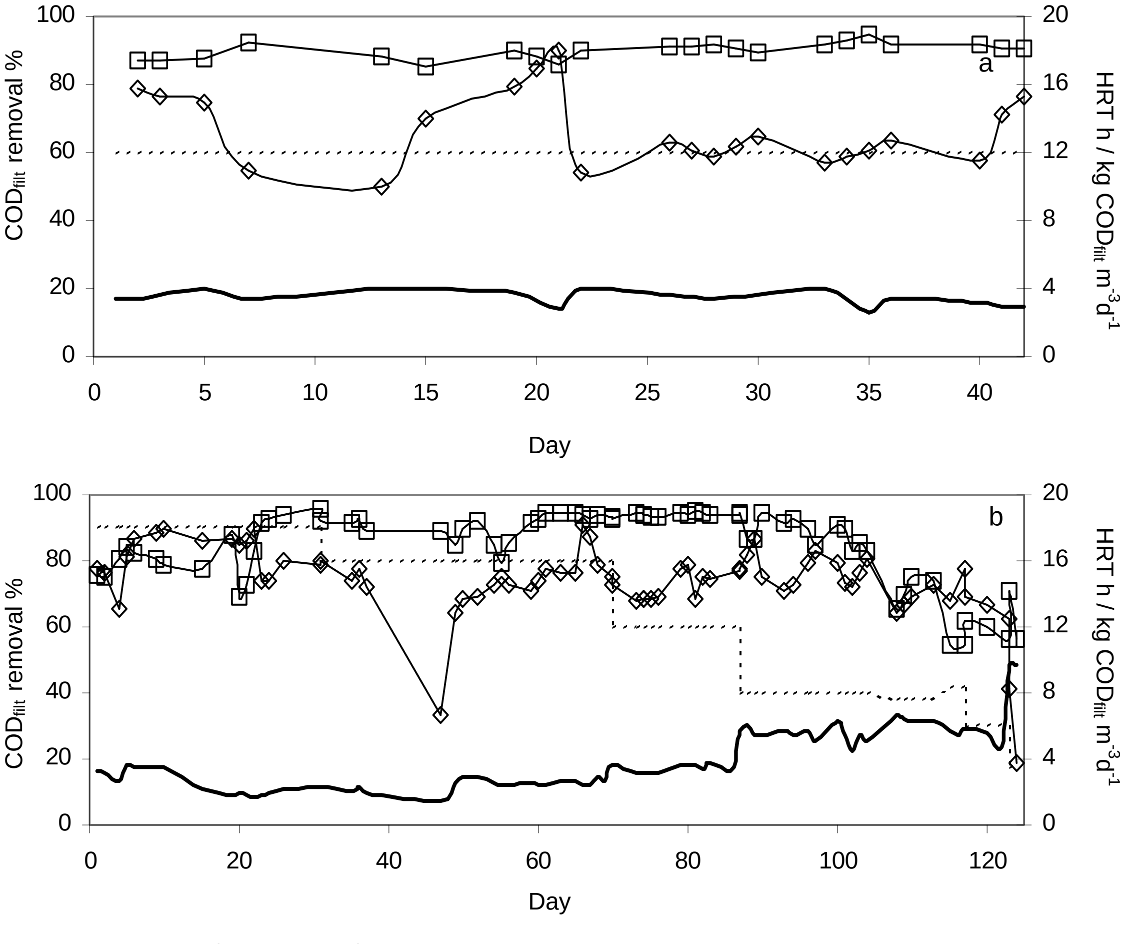

FIGURE 6 Performance of thermophilic and mesophilic ASPs as $\mathrm{COD_{filt}}$ removals under different HRTs and VLRs. Figure a: thermophilic (÷) and mesophilic (D) ASP $\mathrm{COD_{filt}}$ removal, HRT (--), and VLR (ʃ) (I). Figure b: thermophilic (÷) and mesophilic (D) ASP $\mathrm{COD_{filt}}$ removal, HRT (--), and VLR (ʃ) (II).

TABLE 4 Performance of thermophilic (55°C) and mesophilic (35°C) ASPs as COD$_{filt}$ removals without or with polymer dosing (I) and under different VLRs, SLRs, and HRTs (II). Average ∂ standard deviation for selected periods. (Fig. 6).

Process	Days	VLR kg COD$_{filt}$ m^{-3}d^{-1}	SLR kg COD$_{filt}$ kg SS^{-1}d^{-1}	HRT h	COD$_{filt}$ removal %	Ref.
Thermophilic ASP	1-29	3.7∂0.2^1	3.5∂3.2^1	12^1	68∂12^1	I
	30-42	3.4∂0.3^2	2.4∂1.2^2	12^2	64∂7^2	
Mesophilic ASP	1-29	3.7∂0.2^1	1.8∂0.8^1	12^1	89∂2^1	
	30-42	3.4∂0.3^2	1.4∂0.5^2	12^2	92∂2^2	
Thermophilic ASP	23-69	2.4∂0.3	5.5∂2.2	16+0.8	74∂10	II
	70-86	3.4∂0.2	2.5∂0.3	12∂0.0	72∂4	
	87-113	5.7∂0.6	7.0∂4.0	8∂0.2	75∂6	
	115-122	5.5∂0.4	9.9∂0.7	7∂1.2	69∂5	
	123-214	9.7∂0.0	17.5∂0.0	3.5∂0.0	30∂16	
Mesophilic ASP	23-69	2.4∂0.3	1.3∂0.4	16+0.8	91∂4	
	70-86	3.4∂0.2	1.2∂0.1	12∂0.0	94∂1	
	87-113	5.7∂0.6	5.2∂3.5	8∂0.2	85∂9	
	115-122	5.5∂0.4	9.7∂2.0	7∂1.2	58∂3	
	123-214	9.7∂0.0	15.6∂0.0	3.5∂0.0	64∂10	

ASPs ^{1}without polymer, ^{2}with polymer

Studies comparing mesophilic and thermophilic aerobic processes or a single reactor operated at different (20-70°C) temperatures have variously reported either mesophilic or thermophilic conditions as more advantageous (Table 5). Couillard & Zhu (1993) found similar and high (90%) COD removals under different temperatures (45-58°C) and high loading rates up to 12 kg COD m^{-3}d^{-1}. Johnson & Hall (1996) reported SBR treatment of synthetic TMP whitewater to fail at 50°C; however, their process experienced a massive sludge wash-out with increased temperature, which apparently caused the process failure. Tripathi & Allen (1999) found slightly lower COD removal at 55-60°C (62-63%) than at 35-45°C (73-75%) in aerobic sequencing batch reactors treating bleach kraft mill effluent (BKME). They suggested the lower removals under higher temperatures to be due to the inability of the thermophilic microorganisms to remove the same range of compounds as their mesophilic counterparts. The average AOX removals were also lower at 55-60°C (60∂12%) than at 35-45°C (70∂6%). Vogelaar et al. (2002a) studied thermophilic and mesophilic ASP performance under different SRTs and found that changing the SRT from 20 to 10 d had only some effect on the both thermophilic and mesophilic ASP COD$_{tot}$ removals, whereas more characteristically COD$_{col}$ and COD$_{sol}$ removals were increased with decreased SRT. Measured as COD$_{tot}$, COD$_{susp}$, COD$_{col}$, and COD$_{sol}$ thermophilic ASP gave lower removals than mesophilic ASP (2002a, 2002b). Most studies have measured higher COD removals in mesophilic than in thermophilic processes (Table 5). Mesophilic ASPs have also manifested more stable performance as COD$_{filt}$ removals than thermophilic ASPs (Vogelaar et al. 2002a).

TABLE 5 Studies of comparative mesophilic and thermophilic aerobic suspended sludge treatments.

Process	T °C	Scale	Wastewater	VLR kg COD $m^{-3}d^{-1}$	HRT h	COD_{filt} removal %	Reference
SCAS	45	lab	Slaughterhouse effluent	2.4-12	6-30	>90	Couillard & Zhu 1993
	52			2.4-12	6-30	>90	
	58			2.4-6	12-30	86	
SBR	20	lab	TMP whitewater	1.4	48	69	Tardif & Hall 1997
	30					76	
	40					59	
	45					63	
	50					9	
SBR	35	lab	Bleached kraft	0.9-1.4	12	75	Tripathi & Allen 1999
	45					73	
	55					62	
	60					63	
ASP	30	lab	Anaerobically pre-treated paper mill process water	4.1	12	57-60[1]	Vogelaar et al. 2002a
	55					43-56[1]	
ASP	30	lab	Anaerobically pre-treated paper mill process water	3.3-6.0	12	64-70[1]	Vogelaar et al. 2002b
	55					53-54[1]	

ASP = activated sludge process, SBR = sequencing batch reactor, SCAS = semi-continuous activated sludge process, SCBP = suspended carrier biofilm process, [1]COD_{tot} removals.

4.1.2 Fluctuating temperature ASP

The effect of varying temperature on APS performance was studied by changing the temperature randomly and rapidly between 27-56°C (Fig. 7) under different VLRs and HRTs. In the previous chapter (4.1.1.) ASPs, which were run at 35°C and 55°C as reference processes, were described (Fig. 6) (II). In the fluctuating temperature ASP the HRT was gradually reduced from 18 h to 4.5 h, corresponding to an increase in VLR from 2 to 7.5 kg COD_{filt} $m^{-3}d^{-1}$. Owing to the more fluctuating process conditions (temperature) compared to the mesophilic and thermophilic ASPs, the changes in HRT and VLR were conducted more moderately (Fig. 7) (II). The average COD_{filt} removals under mesophilic and thermophilic conditions were close to those under the respective conditions (Table 6). The fluctuating temperature ASP experienced the same starvation period as the thermophilic and mesophilic ASPs. The starvation affected the COD_{filt} removal of the fluctuating temperature ASP, which dropped from 63 to 56% for one day thereafter returning to 69% (Fig. 7). The reactor temperature at that time was 55°C.

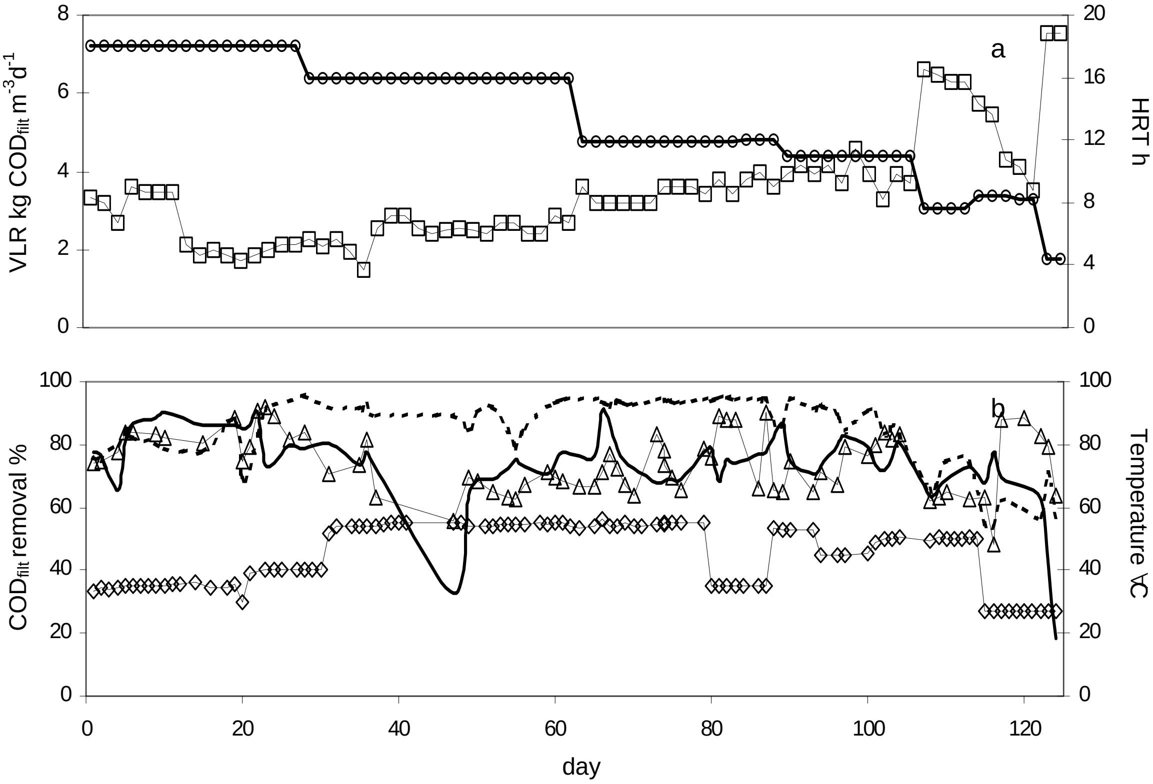

FIGURE 7 Performance of fluctuating temperature ASP. Figure a: HRT h (') and VLR kg COD_{filt} $m^{-3}d^{-1}$ (D). Figure b: Fluctuating temperature ASP temperature ∀C (÷) and COD_{filt} removal % (æ), thermophilic ASP COD_{filt} removal (ĭ), and mesophilic ASP COD_{filt} removal (--). (II).

TABLE 6 Fluctuating temperature ASP COD_{filt} removals and reference COD_{filt} removals in the thermophilic (55∀C) and mesophilic (35∀C) ASPs (II).

Process conditions	Temperature ∀C	N	COD_{filt} removal %	Reference COD_{filt} removal % at 35∀C and 55∀C
Mesophilic	27	4	85∂4	88∂9
	35	6	83∂10	
	40-45	8	80∂8	
Thermophilic	50-52	14	72∂9	74∂8
	55	25	70∂5	

Data for individual temperatures are from different VLRs and HRTs (Fig. 7).
N = Number of samples.

The fluctuating temperature ASP was capable of producing stable, COD_{filt} removals comparable to those of the reference ASPs, which was apparently shown for the first time. Apparently, the microbial population in the thermophilic aerobic processes was flexible and tolerated sudden short and long-term temperature changes. Barr et al. (1996) found in a thermophilic (50∀C) ASP that an 8 to 10-hour drop in temperature to 43, 33.5, 18, and 9.5°C caused BOD removal to drop from 90% to 80-85% at 43-33.5°C, and to 70-55% at 18-9.5°C. After all the temperature drops 1-3 days was a sufficient period for the process to return to removing 90% BOD.

4.1.3 Thermophilic and mesophilic ASP effluent quality

Thermophilic and mesophilic ASP effluent quality was established in different CODs and studied in the reactors describe earlier (I). Different CODs (COD_{tot}, COD_{susp}, COD_{col}, and COD_{sol}) for selected samples (during normal operation) were characterised for the feed and effluents from both the thermophilic and mesophilic ASPs (Fig. 8).

The thermophilic effluent showed markedly higher COD_{tot} values than did the mesophilic effluent. This was due, especially, to the increase in COD_{col} in the thermophilic treatment, as both treatments gave varying and high effluent COD_{susp} values. COD_{susp} indicates the unsettleable fraction of suspended solids in the effluents. Both reactors suffered from poor sludge settling properties, measured as varying effluent COD_{susp} values. In both processes the COD_{susp} values were higher compared to those of the feed wastewater. The thermophilic ASP produced slightly higher effluent COD_{susp} than the mesophilic ASP (Fig. 8).

The mesophilic ASP gave 99∂21% COD_{col} removal with effluent values of 6∂5 mg l^{-1}, whereas in the thermophilic ASP the COD_{col} values were increased to 460∂170 mg l^{-1} (I). COD_{sol} removals were 91∂1% and 89∂2% in the mesophilic and thermophilic ASPs, respectively. The polymer dosing effect on the effluent quality measured as different COD characteristics was negligible. Compared to the mesophilic ASP effluent, COD_{col} had a clearly greater influence to the lower thermophilic COD removals than COD_{susp} (Fig. 8). The possible reasons for higher COD_{col} values and/or high turbidity in thermophilic treatment processes are, e.g., the composition of the feed wastewater, high density of free bacteria and poor floc formation, and higher proportion of complex substances in the wastewater (Tripathi & Allen 1999, Vogelaar et al. 2002a, 2002b).

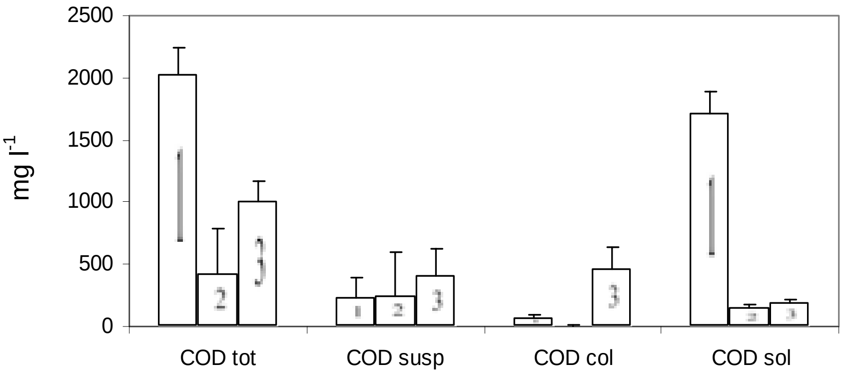

FIGURE 8 Different CODs in the feed wastewater (1), the mesophilic ASP effluent (2) and thermophilic ASP effluent (3) as average + standard deviation (I). Number of samples = 8.

Both the thermophilic and mesophilic ASPs efficiently removed COD_{sol}. Thermophilic COD_{sol} removal may depend on wastewater biodegradability, and microorganisms using the substrate also produce organic substances, soluble microbial products, which may have an inert nature. Thermophilic (55°C) treatment of easily degradable, acetate wastewater, gave slightly higher COD_{sol} removal than mesophilic (30°C) treatment (Vogelaar et al. in press a). With more complex wastewaters, such as pre-treated paper mill effluent or pharmaceutical wastewater, thermophilic aerobic processes have removed less COD_{sol} than mesophilic aerobic processes (LaPara et al. 2001, Vogelaar et al. 2002a, 2002b) (Table 7). In the treatment of pharmaceutical wastewater at 30°C and 60°C more COD_{sol} was removed at 30°C (62%) (LaPara et al. 2001). LaPara et al. (2001) suggested that lower removals (38%) under thermophilic conditions were related to the lower diversity of the thermophilic population. They also reported that mesophilic-mesophilic (30°C) treatment resulted in a greater reduction in COD_{sol} (80%) than thermophilic-mesophilic (55 / 30°C) treatment (75%) and suggested that this was related to the increased proportion of non-degradable COD caused by the bacterial decay at higher temperatures. However, the differences between the two effluent COD_{sol} values were rather small, compared to the initial feed wastewater COD_{sol}. Even though Vogelaar et al. (2002a) reported lower thermophilic than mesophilic COD_{sol} removal, their COD_{sol} removals under both the thermophilic (14-27%) and mesophilic (26-33%) conditions were markedly low, owing to the low amount of biodegradable substrate, measured as COD_{sol} in the anaerobically pre-treated wastewater. However, even with higher-COD wastewater, Vogelaar et al. (2002b) obtained a similar result.

TABLE 7 Comparison of COD_{sol} removals from different wastewaters in thermophilic and mesophilic aerobic processes.

Process	T°C	Wastewater	COD_{sol} removal %	Reference
ASP	30	Anaerobically pre-treated paper mill effluent	26-33	Vogelaar *et al.* 2002a
	55		14-27	
ASP	30	Anaerobically pre-treated paper mill effluent	21-35	Vogelaar *et al.* 2002b
	55		28-45	
Batch	30	pharmaceutical wastewater	65^1	LaPara *et al.* 2001
	35		58^1	
	40		52^1	
	45		52^1	
	50		54^1	
	55		42^1	
	60		37^1	
CSTR	30	acetate-wastewater	$96\text{-}97^2$	Vogelaar *et al.* in press a
	55		$97\text{-}98^2$	

^{1}values estimated from figure in original article, ^{2}VFA removal.

COD_{col} values and removals are rarely reported in wastewater treatment studies. However, effluent COD_{col} values can be related to higher effluent turbidity (Vogelaar et al. 2002b). Turbidity is reported more often in thermophilic aerobic treatment studies than COD_{col} values (reviewed by LaPara

& Alleman 1999, Jahren 1999, Suvilampi et al. 2001, Vogelaar 2002). In the present thesis COD$_{filt}$ and COD$_{sol}$ were fractioned by GF/A (1.6 σm) and 0.45 σm filters, whereas Vogelaar et al. (2002a, 2002b, in press b) fractioned COD similarly to COD$_{tot}$, COD$_{susp}$, COD$_{col}$, and COD$_{sol}$. However, they used different pore-size filters (4.5 σm for COD$_{filt}$ and 0.6 σm for COD$_{sol}$). In a batch experiment they found COD$_{col}$ to be higher after thermophilic than mesophilic treatment (Vogelaar et al. in press b), whereas in continuously operated ASPs the thermophilic ASP gave lower effluent COD$_{col}$ than the mesophilic ASP (Vogelaar et al. 2002a, 2002b). More often, only COD$_{tot}$, COD$_{filt}$, or COD$_{sol}$ values, with or without effluent turbidity measurements, have been reported in thermophilic wastewater treatment studies, and calculated values, such as COD$_{col}$, have not. In many cases the high COD$_{filt}$ values in thermophilic effluents are probably due to higher COD$_{col}$ values, since, at least for easily biodegradable wastewaters, thermophilic bacteria can remove the same amount of COD$_{sol}$ as mesophilic bacteria (Vogelaar et al. in press a, Table 7). Colloidal particles in the diluted molasses wastewater did not cause high thermophilic effluent COD$_{col}$ values in the laboratory experiments, since the feed wastewater COD$_{col}$ values were markedly lower (70∂25 mg l^{-1}) than they were in the thermophilic effluents (350∂220 mg l^{-1}) (I-III). Methods of improving thermophilic ASP floc formation and sludge settleability have not been proposed so far, but it appears that a cationic agent might be one way of achieving better floc formation under thermophilic conditions. Divalent and trivalent cations are used to enhance mesophilic ASP performance and effluent quality, as showed by Murthy & Novak (1998).

In the present study the use of polymer did not have any obvious effect on thermophilic effluent quality (I). Apparently the polymer did not interact with the dispersed, free bacteria. The polymer was expected to join dispersed particles together, which should particularly have been seen in a difference in thermophilic COD$_{col}$ values: because small dispersed particles, such as free bacteria of size 0.45-1.6 σm, would be expected to adsorb onto the polymer and to the flocs bridged by the polymer, their removal should be seen in decreased COD$_{col}$ values, and probably in the form of a shift from COD$_{col}$ to COD$_{susp}$.

The effect of prolonged (24 h) aeration and temperature on thermophilic effluent COD$_{filt}$ was studied in batch experiments at 35∀C and 55∀C (I, II) in order to study the higher CODs in thermophilic effluent, which were assumed to be due to either the lower COD removal of the thermophilic microorganisms or some physico-chemical difference in a high-temperature environment. Thermophilic effluents had markedly higher COD$_{tot}$ and COD$_{filt}$ than the mesophilic effluents (Chapter 4.1.1. Fig. 6; this Chapter, Fig. 8). In more detail, effluent COD$_{susp}$ and COD$_{col}$ values were higher for thermophilic effluent (Fig. 8). Both of these CODs were considered to be higher in the thermophilic effluent due to the high amount of free and dispersed bacteria (COD$_{col}$) and to the poor settling properties of the bigger aggregates (COD$_{susp}$). Aeration at 55∀C did not reduce COD$_{filt}$, whereas at 35°C COD$_{filt}$ removal was 38-43% (Fig. 9). The thermophilic effluent COD$_{col}$ values remained the same at 55∀C, whereas at

35\C 90% of COD_{col} was removed (Fig. 10). Some COD_{sol} was removed at 55\C, whereas at 35\C the COD_{sol} values rose from an initial 230 mg l^{-1} to 380 mg l^{-1}. The density of free bacteria in the thermophilic effluent fell from class 4 (the highest density) to 2 (mediate density) under mesophilic conditions and remained the same (4) under thermophilic conditions (I).

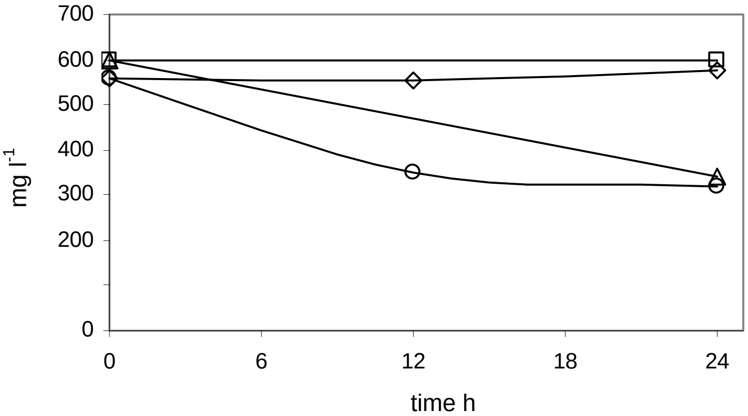

FIGURE 9 The effect of temperature on COD_{filt} in post-aeration of thermophilic ASP effluent (D) at 55\C (I), (÷) at 55\C (II), (æ) at 35\C (I), (') at 35\C (II).

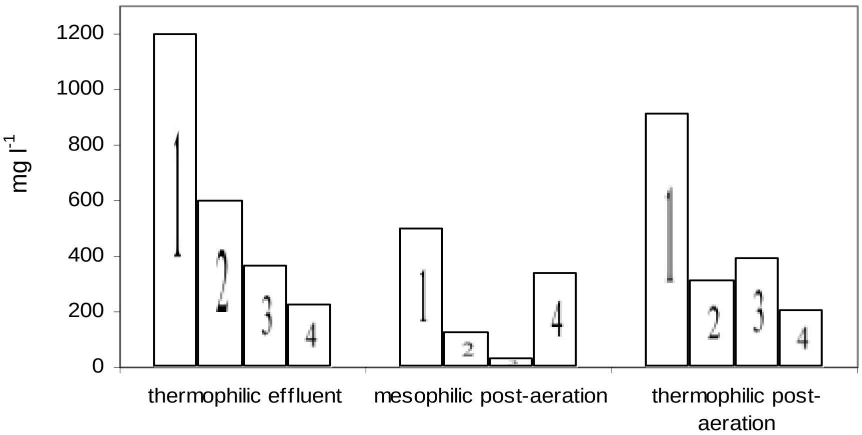

FIGURE 10 The effect of temperature on different fractioned CODs in thermophilic ASP effluent post-aeration at 35\C and 55\C. (1) COD_{tot}, (2) COD_{susp}, (3) COD_{col}, (4) COD_{sol}. (I).

Apparently the reduced COD_{filt} and COD_{col} values were due to the aggregation of dispersed particles and not caused by biological activity, which was supported by the simultaneous decrease in COD_{col} (aggregation) and increase in COD_{sol} (microbial lyses) at 35\C (Fig. 12). COD_{sol} increase has been related to released soluble microbial products (Murthy & Novak 1998). Aeration at 55\C did reduce the COD_{tot} to some extent (24%), probably by dissolution of suspended solids, whereas COD_{filt} (Fig. 11) and COD_{sol} (Fig. 12) remained unchanged. Apparently in all of the batches thermophilic bacteria decayed and at 55\C the remaining bacteria were able to utilise the SMP from the decayed bacteria, whereas at 35∀C this did not occur. This leads one to assume that releasing thermophilic effluent at a lower temperature improves the settling properties of thermophilic sludge by physical aggregation, and further that if the mesophilic bacteria are able to utilise the SMP from the decay of the thermophilic bacteria, the total effluent quality will improve.

4.1.4 Thermophilic and mesophilic ASP sludge settleability

The ASPs were also used to compare thermophilic and mesophilic sludge settling properties and MLSS values (I, II). In the first study the influence of polymer dosing on SVIs was also studied (I). Under both the thermophilic and mesophilic conditions MLSS varied markedly (Fig. 11) due to problems in the operation of settling units and sludge recycling. However, operating the reactors under increased VLR led to higher MLSS. The thermophilic ASPs had lower MLSS values than the mesophilic ASPs (Table 8). The thermophilic ASPs gave better SVIs than the mesophilic ASPs. After dosing with cationic polymer, the SVI values showed drastic change: in the thermophilic ASP SVIs increased, whereas in the mesophilic ASP SVIs were reduced (I).

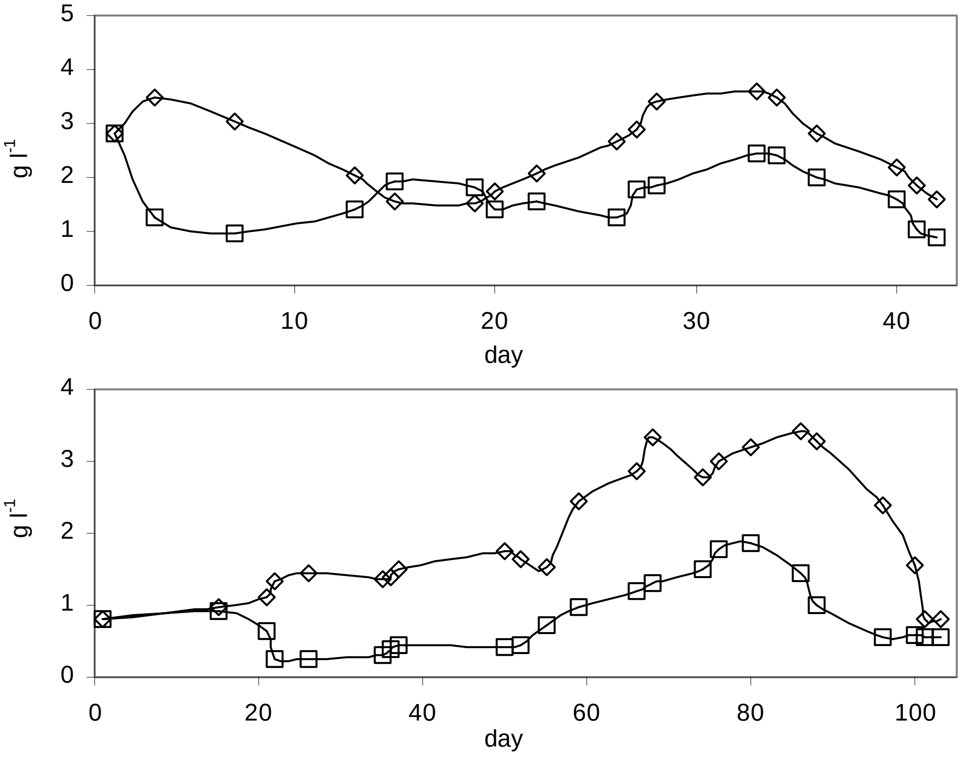

FIGURE 11 Thermophilic (D) and mesophilic (÷) MLSS. Figure a: ASPs operated under constant HRT and VLR (Fig. 6, Table 5, I), figure b: ASPs operated under different HRTs and VLRs (Fig. 6, Table 5, II).

TABLE 8 Thermophilic and mesophilic ASPs sludge settleability as SVI values and MLSS (I, II).

Process	T ∀C	SVI ml g^{-1}	MLSS g l^{-1}	Reference
ASP	35	740∂160^1	2.5∂1.0^1	I
		145∂40^2	2.7∂1.1^2	
	55	40∂30^1	1.6∂0.8^1	
		90∂40^2	1.7∂0.8^2	
ASP	35	220∂160	2.0∂1.1	II
	55	280∂240	0.9∂0.6	

^{1}ASP without polymer, ^{2}ASP with polymer dose

During the SVI measurements the thermophilic sludge bed settled rapidly into the bottom of the settling column (data not shown). Typically no difference was found in sludge bed height after 15 min in the case of thermophilic sludge whereas for mesophilic sludge it took 30 min to settle (visual perception, data not shown). Apparently the thermophilic aggregates which were able to settle and were also visually detectable, had a higher settling velocity than the mesophilic aggregates under lower temperatures. As temperature rises viscosity falls, which increases the settling velocity of particles (Schwarzenbach et al. 1993). Consequently better sludge settling properties could be expected under thermophilic than mesophilic conditions. However, many laboratory thermophilic aerobic wastewater treatment processes have suffered from poor sludge settling properties (reviewed by LaPara & Alleman 1999). A few exceptions have, however been reported, such as Barr et al. (1996), who found no difference in the sludge settling properties of mesophilic (35∀C) and thermophilic (50∀C) ASPs treating bleached kraft mill effluent and Vogelaar et al. (2002a), who reported excellent sludge settling properties (SVI 12∂8 ml g^{-1}) in a thermophilic ASP treating anaerobically pre-treated paper mill wastewater. Also, the mesophilic ASP in the same study had a markedly low SVI (21∂8 ml g^{-1}). It would seem that the excellent sludge settling properties in the study by Vogelaar et al. (2002a) were due to a high calcium content (400 mg l^{-1}) in the wastewater. Ca^{2+} creates cationic bridges with EPS within the floc and can be considered as an effective flocculant (Sobeck & Higgins 2002).

The reason why thermophilic compared to mesophilic sludge settling properties may be poorer is due to the weaker structure of flocs in the formers, which in this study was revealed by microscopy (IV). However, both the thermophilic and mesophilic ASPs had high SVI values in the laboratory study described earlier (II). A marked difference was also measured between two separated runs (I, II). The differences between the two runs were in their operational parameters, such as HRT and VLR, and also in feed wastewater pre-treatment: in the latter study the feed was stored at room temperature and to avoid biological activity the pH was lowered to 5. In the former study the feed was stored in the refrigerator and the pH was kept high (7.5). Consequently, in the latter case markedly more pH regulation was required for the feed before it was fed into the reactors. Besides high temperature, a high monovalent to divalent (M/D) cation ratio may be the reason for the impaired floc structure and poor sludge settling properties in ASPs, since pH regulation by NaOH increases the M/D ratio (Murthy 1998). Monovalent ions, such as sodium, decrease the binding strength of the EPS to the floc structure, releasing colloidal particles into the solution (Murthy 1998). Mikkelsen & Keiding (2002b) suggested that extracellular polymeric substances (EPS) are the most important factor in floc structure. On the basis of their study Vogelaar (2002) suggested that EPS production correlates with temperature and a decreased amount of EPS inhibits floc formation. Liao et al. (2001) suggested that sludge hydrophobicity and surface charge correlate positively with protein EPS and negatively with total carbohydrate EPS. They also found that of the EPS components protein and DNA-EPS had an influence on SVI but that the amount of dispersed particles (measured as effluent suspended solids) did not, whereas total carbohydrate EPS had a correlation with dispersed particles but not with SVI.

4.2 Combined thermophilic-mesophilic aerobic process

4.2.1 Performance of combined thermophilic-mesophilic process

A combined thermophilic-mesophilic wastewater treatment was studied using a laboratory thermophilic ASP followed by mesophilic ASP and a thermophilic SCBP followed by a mesophilic ASP (III). Both treatment lines were fed with diluted molasses wastewater and operated under similar conditions, including HRT, SRT, VLR, and temperature (Fig. 13). The HRT was gradually reduced in both treatment lines in three stages; from 36 h to 24 h to 16 h.

Throughout the runs the thermophilic-mesophilic ASP-ASP treatment gave average COD_{tot} and COD_{filt} removals of 81±7% and 85±5%, respectively, and the thermophilic-mesophilic SCBP-ASP treatment gave COD_{tot} and COD_{filt} removals of 82±8% and 87±3%, respectively (Fig. 12). Increased VLR and reduced HRT had no effect on COD_{filt} removal in either of the treatment lines. Thermophilic treatment efficiently removed 90% COD_{sol} (Table 9). Mesophilic post-treatment removed the colloidal COD (difference between COD_{filt} and COD_{sol}) present in the thermophilic effluents. Table 9 summarises the performance of both treatment lines.

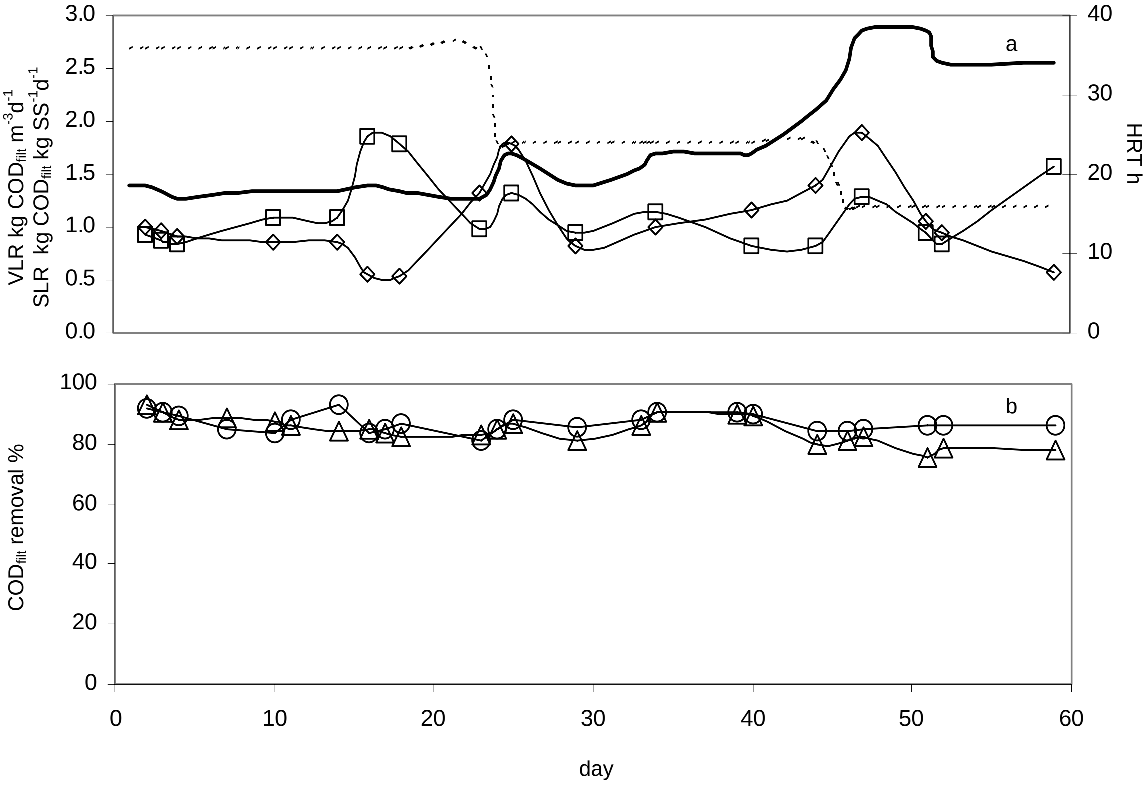

FIGURE 12 Performance of combined thermophilic-mesophilic processes. Figure a: combined processes HRT (--), VLR (—), thermophilic-mesophilic ASP-ASP SLR (÷), and thermophilic-mesophilic SCBP-ASP SLR (D); b: ASP-ASP COD_{filt} removal (æ) and SCBP-ASP COD_{filt} removal (') (III).

TABLE 9 Performance of combined thermophilic-mesophilic aerobic processes (A12 as thermophilic ASP followed by mesophilic ASP and B12 as thermophilic SCBP followed by mesophilic ASP) under different HRTs, VLRs, and SLRs (III). (Fig. 12).

		Period I days 1-23		Period II days 24-45		Period III days 46-59	
	unit	A12	B12	A12	B12	A12	B12
HRT	h	36∂0	36∂0	24∂0	24∂0	16∂0	15∂0
VLR	kg COD_{filt} m^{-3} d^{-1}	1.4	1.4	1.6	1.6	2.7	2.8
SLR	kg COD_{filt} kg MLVSS d^{-1}	1.5	1.2	1.2	1.0	1.1	1.2
COD_{tot} removal	%	86∂5	79∂11	79∂7	84∂4	75∂5	86∂1
COD_{filt} removal	%	87∂4	87∂4	86∂5	88∂3	79∂3	86∂1
COD_{sol} removal	%	84[a]	88[a]	78∂6	84∂0	77[a]	84[a].
Effluent SS	mg l^{-1}	18∂19	80∂53	55∂38	40∂42	160 ∂68	120 ∂125

[a]data for one sample only

The present results show that the combined thermophilic-mesophilic treatment of diluted molasses wastewater gave high (80-90%) COD removals. Thermophilic treatment efficiently removed COD_{sol}, whereas COD_{col} was increased, which was manifested as higher COD_{filt} values in the comparative studies (4.1: I, II). Mesophilic post-treatment removed the COD_{col} present in the thermophilic effluents and subsequently lowered COD_{filt} values. However, even when the combined treatment achieved 90% COD_{filt} removal, the one-stage mesophilic ASP treating the same wastewater also gave 90% COD_{filt} removal whereas in the same study the thermophilic ASP gave 67% COD_{filt} removal (I). In a study by LaPara et al. (2001) mesophilic-mesophilic (30 / 30°C) treatment removed more COD_{sol} (80%) than thermophilic-mesophilic (55 / 30°C) treatment (75%), which they suggested was due to the higher portion of recalcitrant SMP-originated COD in the thermophilic effluent than in the mesophilic effluent. Apparently combined thermophilic-mesophilic aerobic wastewater treatment of easily degradable wastewater does not necessarily yield higher effluent quality than single mesophilic treatment but can produce better effluent quality than thermophilic treatment alone.

4.2.2 Comparison of thermophilic ASP and thermophilic SCBP

The thermophilic ASP and SCBP, the first stages in the combined thermophilic-mesophilic processes shown in the previous section (4.2.1), were compared in more detail in respect of COD removals and sludge characteristics (III). HRT was gradually reduced in both reactors in three stages, from 18 h to 12 h to 8 h (to 7 h in the SCBP); corresponding to an increasing VLR from 2.7 to 3.3 to 5.7-6.0 kg COD_{filt} $m^{-3}d^{-1}$. Between HRTs of 12-18 h the thermophilic ASP and SCBP removed 60±13% and 62±7% of COD_{filt}, respectively, and with HRT of 8 h the corresponding removals were 48±1% and 69±4% (Fig. 13). The SCBP removed slightly more COD than the ASP and produced markedly better sludge settling properties (lower SVI values) than the ASP. During the first few days after start-up COD_{filt} removals were markedly higher in the SCBP than in the ASP. Table 10 show thermophilic ASP and SCBP performance under similar operational conditions.

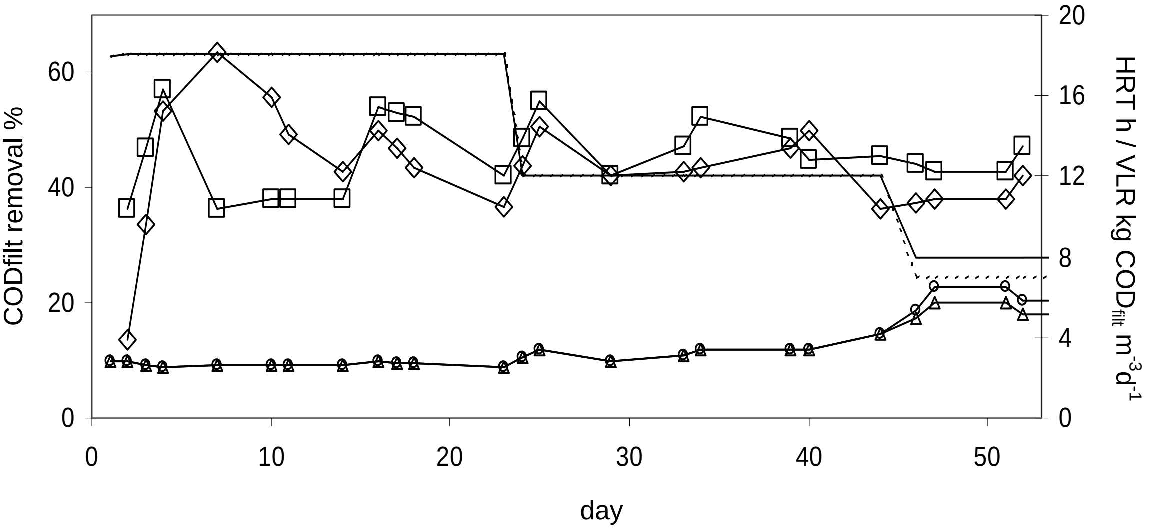

FIGURE 13 Performance of thermophilic ASP and SCBP. ASP HRT (—), SCBP HRT (--)., ASP VLR (æ), SCBP VLR (ʹ), ASP COD_{filt} removal (÷), and SCBP COD_{filt} removal (Ⴇ). (III).

TABLE 10 Performance of thermophilic aerobic ASP and SCBP under different loading rates and HRTs (III).

		Period I days 1-23		Period II days 24-45		Period III days 46-59	
	unit	ASP	SCBP	ASP	SCBP	ASP	SCBP
HRT	h	18	18	12	12	8	7
VLR	kg COD_{filt} m^{-3} d^{-1}	2.7∂0.1	2.7∂0.1	3.3∂0.4	3.3∂0.4	5.3∂0.4	6.0∂0.3
SLR	kg COD_{filt} kg MLSS d^{-1}	7.9∂6.4	7.1∂2.7	3.4∂1.1	1.8∂0.2	1.8∂0.6	2.2∂0.3
COD_{tot} removal	%	44∂13	45∂8	44∂5	48∂4	39∂2	44∂2
COD_{filt} removal	%	58∂14	63∂7	66∂5	61∂6	51∂9	69∂3
COD_{sol} removal	%	84^1	88^1	81∂12	85∂4	nd.	nd.
SVI	ml g^{-1}	nd.	nd.	190∂105	26∂11	167	25∂1

In the SCBP, COD_{filt} removal increased with increasing VLR, whereas ASP COD_{filt} removal decreased, which suggests that SCBPs might require a specific loading rate, higher than that in ASPs, to maintain high COD removal. This is apparently not related to temperature. Several authors have reported SCBPs (or moving bed bioreactors (MBBRs)) as well as ASPs and SBRs treating industrial wastewaters, operating under widely varying loading rates and achieving widely varying COD removals (Couillard & Zhu 1993, Barr et al. 1996, Malmqvist et al. 1996, Tardif & Hall 1997, Broch-Due et al. 1997, Becker et al. 1999, Jahren & Ødegaard 1999a,b, Malmqvist et al. 1999, Rusten et al. 1999, Tripathi & Allen 1999, Jahren et al. 2002, Vogelaar et al. 2002b). It appears that biofilm reactors can operate under higher loading rates than processes based on suspended sludge. However, a thermophilic process with no specific sludge circulation or biofilm fixation was reported to have an extremely high (80-150 kg COD $m^{-3}d^{-1}$) VLR (Becker et al. 1999). One of the reasons why biofilm processes have been claimed to have a higher loading capacity is due to their higher biomass concentrations (Jahren 1999). In this study in both thermophilic reactors the average MLSS (in a biofilm reactor the MLSS includes both suspended sludge and attached sludge) was approximately the same. This suggests that biofilm processes have an advantage at higher loading rates over the suspended sludge process, such as better mass transfer capabilities (Lazarova & Manem 1994), which might be enhanced under thermophilic conditions due to increased dissolution rates (Schwarzenbach et al.1993).

4.2.3 Characterisation of biomass in thermophilic-mesophilic aerobic processes

The previously described (4.2.1) thermophilic-mesophilic wastewater treatments (combination of the thermophilic ASP, followed by the mesophilic ASP and the thermophilic SCBP, followed by the mesophilic ASP) were used to study sludge characteristics and floc formation (IV).

Thermophilic bacteria in both the ASP and SCBP were able to form flocs. However, these were characterized as small (<50 σm) and had weak structure and irregular shape (data not shown). Flocs in both the mesophilic ASPs were bigger (50-500 σm) than those in the thermophilic processes and had more compact structures. Filamentous bacteria joined the small flocs into bigger, settleable groups in both the thermophilic and mesophilic processes. It seems that the growth of thermophilic filamentous bacteria was promoted by decreased HRT, whereas mesophilic filamentous bacteria were suppressed by decreased HRT (Fig. 14). Both thermophilic processes showed a high density of dispersed particles, such as free bacteria (Fig 14) (IV).

In both mesophilic processes the floc sizes were structured differently from those in the thermophilic processes (IV). Flocs were notably larger, firmer and round in shape (IV). The mesophilic ASP after thermophilic SCBP produced larger flocs than the mesophilic ASP after thermophilic ASP, apparently due to good flocculation in the mesophilic reactor combined with detached biofilm particles from the thermophilic reactor, which may adsorb dispersed cells and thereby participate in the flocculation process. Reducing the HRT led to an increase in MLSS in both processes (Fig. 14).

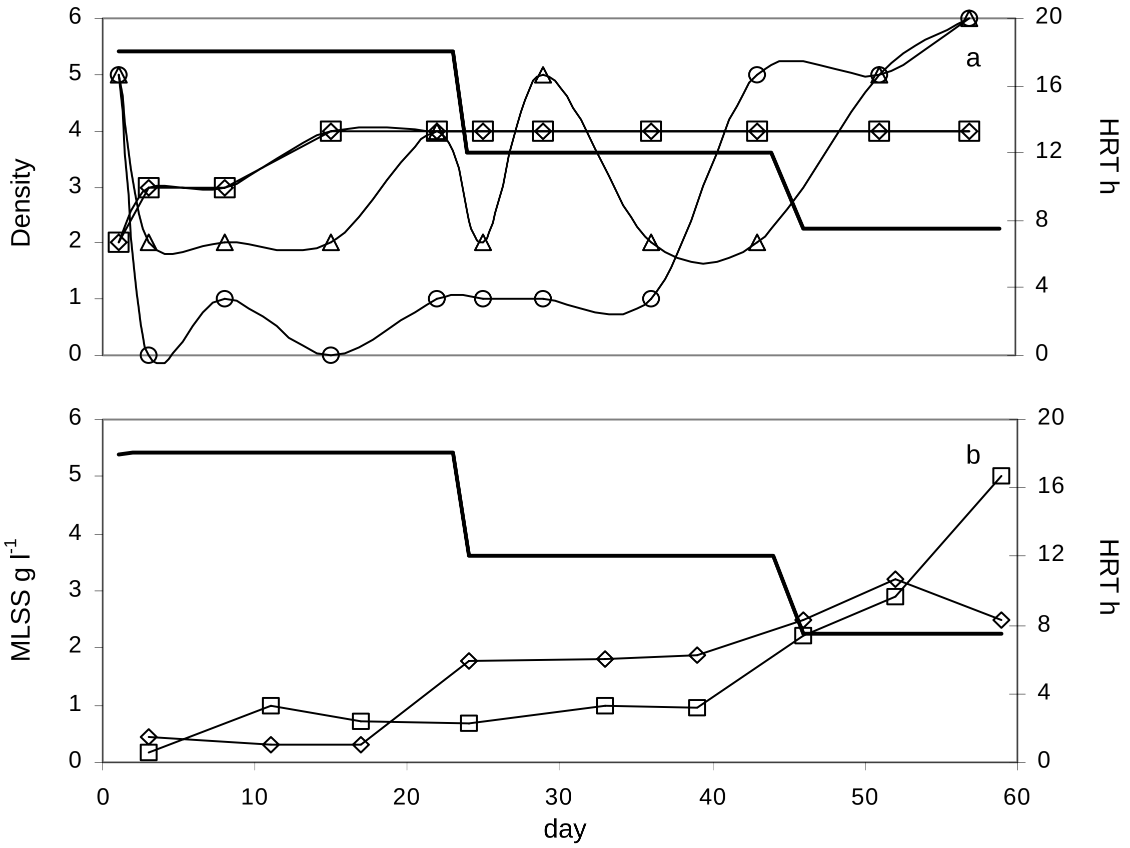

FIGURE 14 Characteristics of thermophilic ASP and SCBP biomass under different HRTs (ʄ). Figure a: density of free bacteria in ASP (Ɗ) and in SCBP (÷) and density of filamentous bacteria in ASP (æ) and in SCBP (ˈ) (IV). Figure b: ASP MLSS (Ɗ) and SCBP MLSS (÷). SCBP MLSS include both the biofilm SS and suspended SS.

Gram-negative bacteria dominated in all processes (Fig. 15), and gram-negative bacteria of <1 σm in size was the most prominent class in the settled flocs. Gram-positive bacteria were also present in both processes, increasing with reduced HRT in both the thermophilic ASP suspension and thermophilic SCBP biofilm (IV). Approximately 20% of gram-positive bacteria in all of the processes were viable.

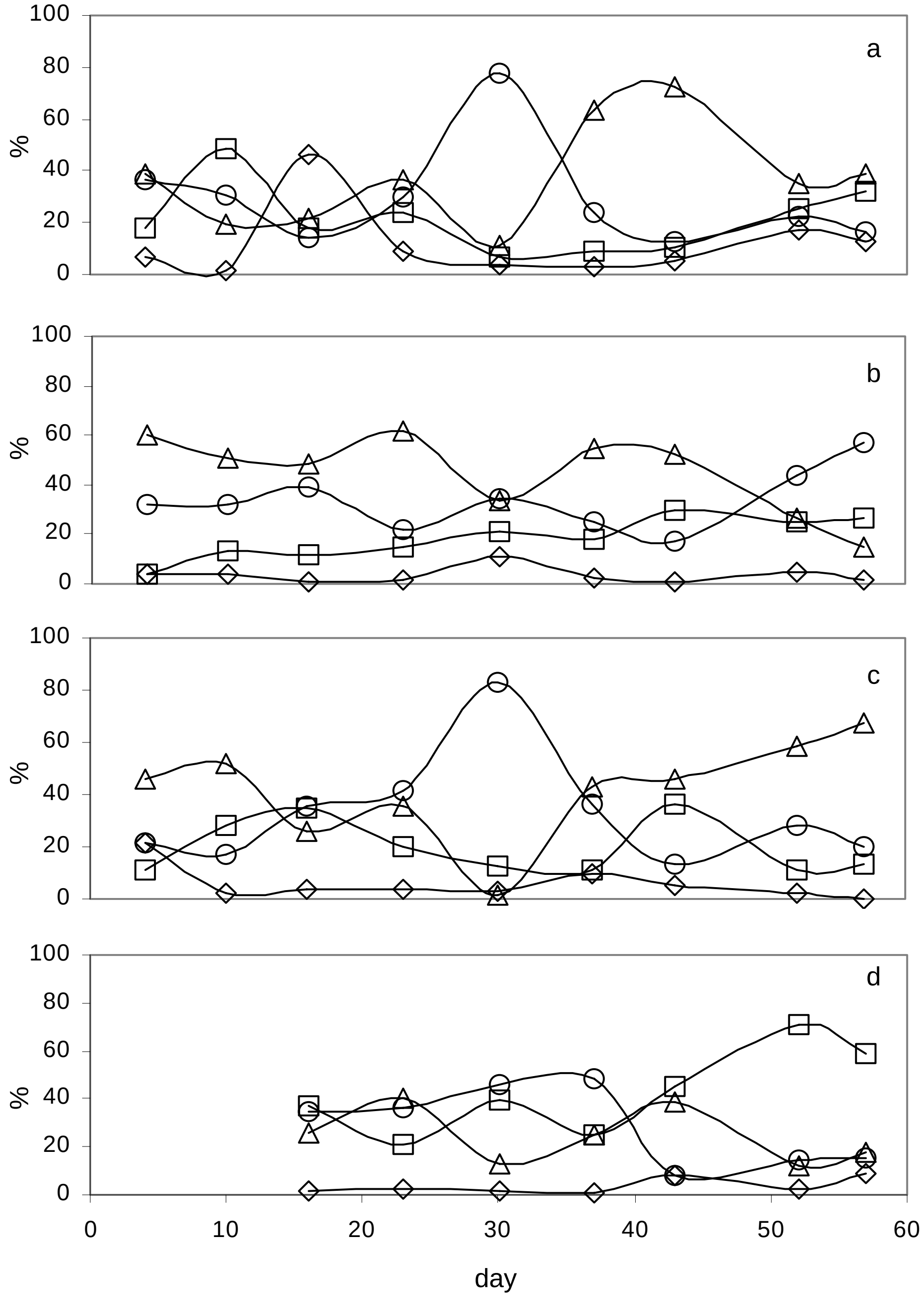

FIGURE 15 Gram-positive viable (÷) and dead (𝔻) bacteria, gram-negative viable (æ) and dead (ˈ) bacteria in thermophilic ASP and SCBP. Figure a: ASP suspension, figure b: ASP settled sludge, figure c: SCBP suspension, figure d: SCBP biofilm. (IV).

The smallest bacteria in the thermophilic ASP were most eager to form flocs, and those flocs were denser than flocs formed by bigger bacteria. Small aggregates had better settling properties than the bigger aggregates since they were dominant in the settled samples. Rattanakawin & Hogg (2001) also suggested that the smallest aggregates yielded denser flocs than the bigger ones. Most of the bacteria in suspended particles from both the thermophilic and mesophilic processes were in the size range of 1-5 σm, whereas in settled flocs the dominant size was <1 σm (IV). Floc sizes in thermophilic ASP have so far been reported in one study, most flocs having sizes between 25-250 σm (Vogelaar 2002). In another thermophilic aerobic process pinpoint flocs were reported, with no particular size determination (Tripathi & Allen 1999). Previously it was believed that thermophilic bacteria are unable to aggregate and form flocs (reviewed by LaPara & Alleman 1999). In the present study the samples from the mesophilic processes had different microbial populations than the samples from the thermophilic processes, which is in agreement with studies of microbial populations in mesophilic and thermophilic processes (Tripathi & Allen 1999, LaPara et al. 2000a). Tiirola et al. (in press) detected in a LH-PCR profile that a thermophilic SCBP had a totally different microbial population in the biofilm than in suspension. They found that beta-*Proteobacteria, Cytophagales,* and gamma-*Proteobacteria* were the most dominant peaks in LH-PCR, but that during process upset *Bacillus* rapidly dominated in both suspension and biofilm. LaPara et al. (2000a) found in a full-scale thermophilic-mesophilic aerobic pharmacy wastewater treatment that microbial diversity decreased with increased temperatures and that beta- *Proteobacteria* were dominant. In the present study, in the mesophilic ASPs gram-negative bacteria also dominated, but, as in the thermophilic processes, gram-positive bacteria were also present.

No higher organisms were present in either of the thermophilic processes, which is in agreement with both theory and previous studies on thermophilic wastewater treatment (Madigan et al. 1998, LaPara & Alleman 1999). In both mesophilic ASPs higher organisms were present and diversity was high. In the ASP after thermophilic ASP ciliates and flagellates were dominant, whereas in the ASP after thermophilic SCBP rotifers dominated the process, indicating good process stability.

4.2.4 Thermophilic and mesophilic ASP sludge yield

Sludge yields were measured in laboratory studies (I, II, III) from thermophilic ASPs as daily values and as net sludge yields. Daily sludge yields had high variation due to variation in effluent SS and MLSS values and therefore are not shown here. Table 11 shows both the thermophilic and mesophilic ASPs net sludge yields (I-III) along with a few comparative values from other studies.

TABLE 11 Aerobic thermophilic and mesophilic ASP sludge yields in aerobic suspended sludge wastewater treatment studies. Results are from the present study (I-III) and from the literature.

Process	T °C	Wastewater	Sludge yield $\text{g SS g COD}_{\text{filt removed}}^{-1}$	Reference
Thermophilic ASP	55	diluted molasses	0.18-0.19	I
Mesophilic ASP	30	diluted molasses	0.15-0.19	
Thermophilic ASP	55	diluted molasses	0.23	II
Mesophilic ASP	30	diluted molasses	0.47	
Thermophilic ASP	55	diluted molasses	0.06	III
Mesophilic ASP after thermophilic ASP	30	diluted molasses	0.07	
Mesophilic ASP after thermophilic SCBP	30	diluted molasses	0.03	
SCNR	45	slaughterhouse effluent	0.35	Gariépy et al. 1989
	52		0.29	
	58		0.28	
SBR	20	synthetic whitewater	0.13	Johnson & Hall 1996
	30		0.10	
	40		-0.005	
	45		0.022	
	50		-0.14	
MBR	55	synthetic kraft pulp mill condensate	0.16	Bérubé & Hall 2000
	60		0.14	
	65		0.12	
	70		0.12	
CSTR	55	synthetic wastewater	0.19	LaPara et al. 2000b
Solid-phase bioreactor	30	synthetic wastewater	0.47	Lim et al. 2001
	40		0.79	
	50		0.79	
Mesophilic ASP	30	anaerobically pre-treated paper mill effluent	0.18	Vogelaar et al. 2002a
Thermophilic ASP	55	anaerobically pre-treated paper mill effluent	0.21	Vogelaar et al. 2002a

Thermophiles are capable of faster growth rates than mesophiles (Sundaram 1986). However, this does not lead to higher sludge yield under higher temperatures, as an increase in temperature decreases microbial net yield due to energy uncoupling (higher maintenance energy, higher cell death rate). Thus, the net sludge yield in biological wastewater treatment processes is expected and has been reported to be lower under thermophilic than mesophilic conditions (Tchobanoglous & Burton 1991, Jahren 1999, LaPara & Alleman 1999, LaPara et al. 2000b). Lower sludge yields with increasing temperatures has been reported under mesophilic conditions as well (Krishna & van Loosdrecht 1999). Several experimental studies have reported lower excess sludge production for thermophilic aerobic treatment than for mesophilic treatment. A few full-scale thermophilic aerobic treatments have indicated rather low sludge yields (Rozich & Bordacs 2002).

The sludge yields in the thermophilic ASPs were low compared to typical values in the mesophilic ASPs (Tchobanoglous & Burton 1991). However, in these studies, the mesophilic ASPs also manifested similarly low yields. Thermophilic aerobic treatment appears to generate low sludge yields, in some cases lower than in analogous mesophilic systems. From the laboratory experiments it can be concluded that sludge yield is difficult to determine reliably, because the variation of measurements of small samples is high. Therefore pilot or more preferably full-scale determinations are needed to reveal the true thermophilic aerobic sludge yield.

4.3 In-mill aerobic thermophilic SCBP treatment of GWM

A two-stage thermophilic aerobic SCBP for treatment of groundwood mill circulation water (GWM) under high and varying process temperature was studied in-mill premises (V). A rapid start-up, measured as DOC removal (comparable to COD_{filt} removal) was achieved using inoculum from an existing mesophilic activated sludge plant treating the pulp and paper mill wastewater (Figs. 16-17). The two-stage SCBP (R12) produced 49-77% DOC removals at VLRs of 3-14 kg COD_{filt} $m^{-3}d^{-1}$ and HRTs of 2-8 h, whereas the single reactor (first stage, R1) produced 40-67% DOC removals at VLRs of 7-28 kg COD_{filt} $m^{-3}d^{-1}$ and HRTs of 1-4 h (Figs. 16-17, Tables 12-13). Few exceptions to stable DOC removals occurred in the pilot trials and were due to either mill upsets or malfunctions in the pilot plant (Figs. 16-17). These disturbances led usually to high pH values of up to 11-12 in the first reactor; in trial II pH failure was accompanied by aeration failures (DO 0 mg l^{-1}). Duration of disturbances was 8-12 h for single days and approx. 20-30 h for two-day shocks. During the pH failures DOC removal in the first reactor dropped to less than 20% whereas the second stage reactor maintained some removal. During plant upsets, attached biomass was also washed out (data not shown). Thermophilic SCBP tolerated periodical temperature variations along with caustic shocks, and COD_{filt} removal returned to normal within 24 hrs.

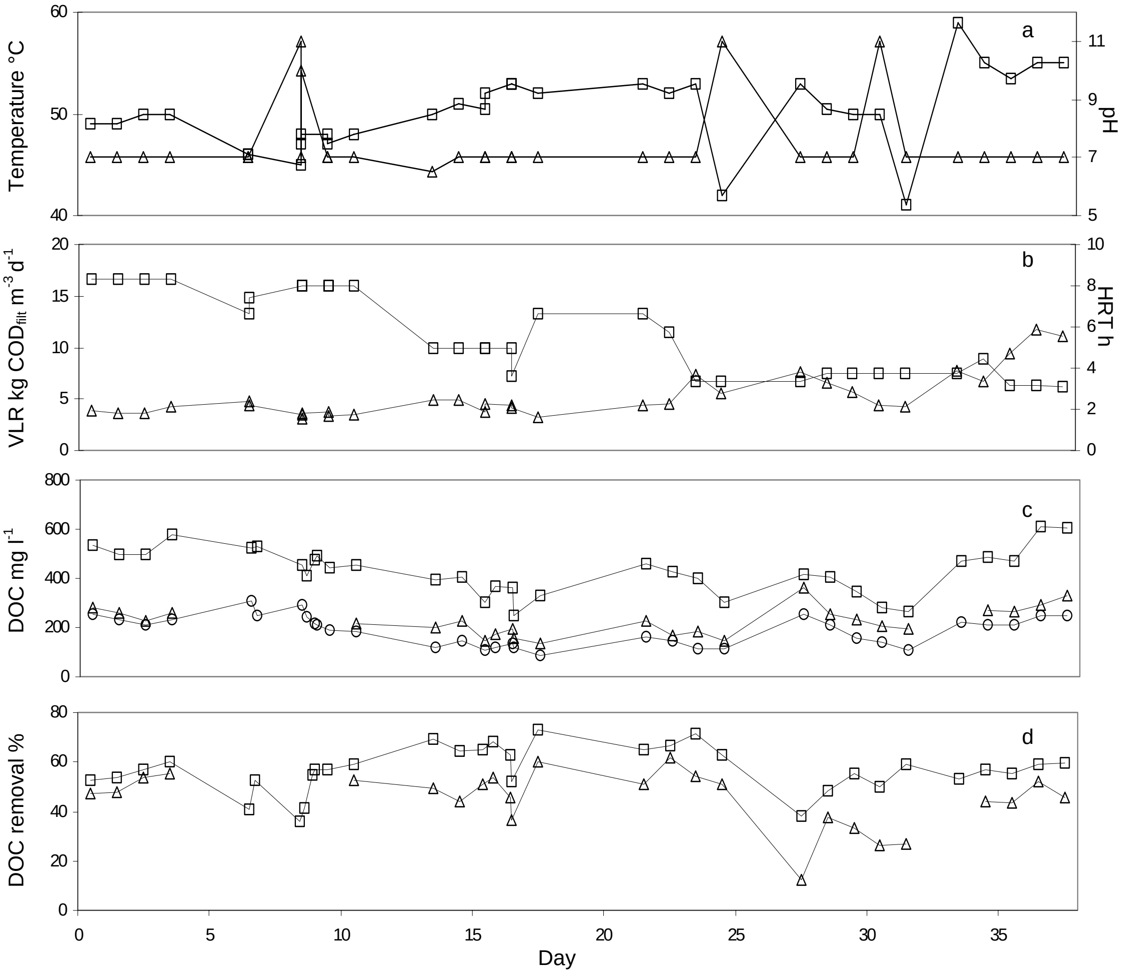

FIGURE 16 Performance of thermophilic aerobic SCBP in pilot trial I. Temperature (ⅅ) and pH (æ) (fig. a), volumetric loading rate (VLR) (æ) and HRT (ⅅ) of two-stage thermophilic SCBP (R12) (fig. b), DOC of GWM (ⅅ), after R1 (æ), and after R2 (ˈ) (fig. c), and DOC removals of R1 (æ) and R12 (ⅅ) (fig. d). R1 VLR and HRT are not presented in the figure, VLR was twice the value for R1 than for R12, and HRT was half the figure for R1 than for R12. (V).

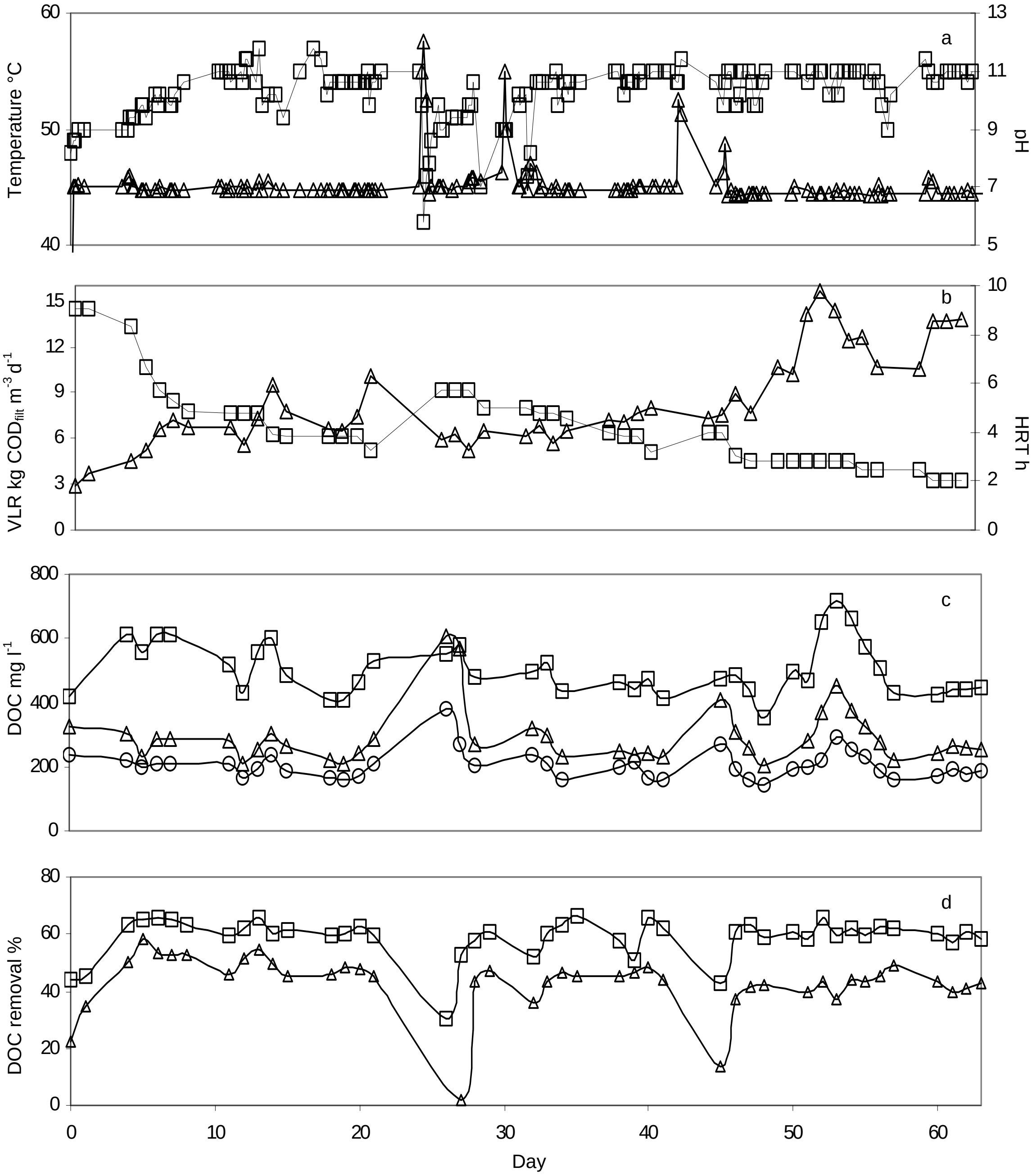

FIGURE 17 Performance of thermophilic aerobic SCBP in pilot trial II. Temperature (D) and pH (æ) (fig. a), volumetric loading rate (VLR) (æ) and HRT (D) of two-stage thermophilic SCBP (R12) (fig. b), DOC of GWM (D), after R1 (æ), and after R2 (') (fig. c), and DOC removals of R1 (æ) and R12 (D) (fig. d). R1 VLR and HRT are not presented in the figure, VLR was twice the figure for R1 than for R12, and HRT was half the figure for R1 than for R12. (V).

TABLE 12 Aerobic thermophilic SCBP performance in trial I under different loading rates in a single reactor (R1) and in a two-stage reactor (R12) (average ∂ standard deviation) (V).

			Reactor R1	Reactor R12	Reactor R1	Reactor R12
Parameter	Unit	Days	17 – 21		35 - 37	
Temperature	°C		52.5 ∂ 0.7		54 ∂ 1	
Loading rate	kg COD_{filt} $m^{-3}d^{-1}$		7.6 ∂ 1.6	3.8 ∂ 0.8	20.5 ∂ 2.3	10.3 ∂ 1.2
Sludge load	kg COD_{filt} kg $VSS^{-1}d^{-1}$		3.0 ∂ 1.4	1.4 ∂ 0.4	11.2 ∂ 2.3	3.8 ∂ 0.5
HRT	h		3.3 ∂ 0.0	6.7 ∂ 0.0	1.6 ∂ 0.1	3.1 ∂ 0.0
Removal rate	kg COD_{filt} $m^{-3}d^{-1}$		4.4 ∂ 0.1	2.7 ∂ 0.3	9.7 ∂ 1.3	6.2 ∂ 1.0
DOC removal	%		55 ∂ 6	70 ∂ 6	45 ∂ 2	57 ∂ 3
COD_{filt} removal	%		59 ∂ 11	72 ∂ 7	47 ∂ 1	60 ∂ 3
COD_{tot} removal	%		53 ∂ 7	64 ∂ 3	42 ∂ 2	50 ∂ 0
BOD_7 removal	%		64 ∂ 9	89 ∂ 4	57 ∂ 2	72 ∂ 1
UV_{280} removal	%		26 ∂ 17	36 ∂ 17	-[a]	8[b]
Effluent SS	g l^{-1}		0.76 ∂ 0.18	0.48 ∂ 0.12	0.69 ∂ 0.07	0.66 ∂ 0.14

[a]negative value, [b]data from one day only

TABLE 13 Aerobic SCBP performance in trial II (average ∂ standard deviation) under different loading rates in a single reactor (R1) and in a two-stage reactor (R12) (V).

			Reactor R1	Reactor R12	Reactor R1	Reactor R12	Reactor R1	Reactor R12
Parameter	Unit	Days	11-13		53-55		61-63	
Temperature	°C		55 ∂ 0.4	53 ∂ 1.7	55 ∂ 0.9	54 ∂ 0.5	55 ∂ 0.9	54 ∂ 0.5
Loading rate	kg COD_{filt} $m^{-3}d^{-1}$		13.3 ∂ 1.8	6.6 ∂ 0.9	27.4 ∂ 2.0	13.7 ∂ 1.0	27.4 ∂ 0.2	13.7 ∂ 0.1
Sludge load	kg COD_{filt} kg $VSS^{-1}d^{-1}$		8.1 ∂ 2.1	2.8 ∂ 0.4	19.6 ∂ 3.1	4.6 ∂ 0.5	15.6 ∂ 0.1	4.3 ∂ 0.0
HRT	h		2.4 ∂ 0	4.8 ∂ 0	1.45 ∂ 0	2.9 ∂ 0	1 ∂ 0	2 ∂ 0
Removal rate	kg COD_{filt} $m^{-3}d^{-1}$		6.7 ∂ 1.1	4.2 ∂ 0.7	11.3 ∂ 06	8.3 ∂ 0.6	11.3 ∂ 0.5	8.0 ∂ 0.2
DOC removal	%		51 ∂ 4	62 ∂ 3	41 ∂ 4	60 ∂ 1	41 ∂ 2	58 ∂ 2
COD removal	%		49 ∂ 4	65 ∂ 3	36 ∂ 3	55 ∂ 2	nd.	nd.
BOD_7 removal	%		68 ∂ 5	80 ∂ 3	45 ∂ 2	76 ∂ 1	nd.	nd.
UV_{280} removal	%		13 ∂ 3	35 ∂ 4	9 ∂ 8	7 ∂ 8	nd.	nd.
Sugars removal	%		82 ∂ 5	88 ∂ 2	76 ∂ 8	82 ∂ 24	nd.	nd.
Effluent NH_4-N	mg l^{-1}		31 ∂ 13	30 ∂ 11	5.4 ∂ 2.9	1.2 ∂ 1.3	nd.	nd.
Effluent SS	g l^{-1}		0.32 ∂ 0.01	0.28 ∂ 0.06	0.34 ∂ 0.03	0.37 ∂ 0.04	0.37[a]	0.22[a]
Sludge yield	kg VSS kg $COD_{filt\ removed}^{-1}$		0.21 ∂ 0.04	0.16 ∂ 0.12	0.21 ∂ 0.01	0.09 ∂ 0.09	0.53[a]	0.14[a]

[a]standard deviation not presented for one analysis only, nd. = not determined

The highest removal rate for the carrier surface area in the pilot-scale experiments (V) was 65 g COD_{filt} $m^{-2}d^{-1}$ under a loading rate of 148 g COD_{filt} m^{-}

$^{2}d^{-1}$. The thermophilic laboratory SCBP was operated under moderate load and the maximum removal rate was 19 g COD_{filt} $m^{-2}d^{-1}$ under a loading rate of 26 g COD_{filt} $m^{-2}d^{-1}$ (III). Maximum removal rates of 95 g COD_{filt} $m^{-2}d^{-1}$ (Jahren & Ødegaard 1999a), 48 g COD_{filt} $m^{-2}d^{-1}$ (Rusten et al. 1999), and 25-30 g COD_{filt} $m^{-2}d^{-1}$ (Ødegaard et al. 2000) have been reported for a laboratory-scale 55°C moving bed biofilm reactor (MBBR) treating high strength wastewater (diluted molasses), a pilot-scale mesophilic (34°C) MBBR treating mixed chemical plant wastewater, and a low-temperature (10-15°C) MBBR treating synthetic wastewater, respectively. A higher temperature increases the COD removal rates according to measurements based on carrier surface area.

Several authors have reported on the operation of SCBPs (or MBBRs) treating industrial wastewaters under wide range of different loading rates and COD removals (Table 14). Biofilm reactors seem to be able to operate under considerably higher loading rates than processes based on suspended sludge. However, a thermophilic process with no specific sludge circulation or biofilm fixation was reported to have an extremely high (80-150 kg COD $m^{-3}d^{-1}$) VLR (Becker et al. 1999). A high-rate process, being a relatively small reactor, is an attractive alternative when a pulp and paper mill is considering implementing in-mill treatment of process waters.

The two-stage SCBP tolerated the different operational conditions and disturbances well even under thermophilic conditions. The operation of a 55°C SCBP with 70 to 90% COD_{filt} removals has been reported even at a pH of 3.2 to 4.5 (Malmqvist et al. 1999). In our study, COD_{filt} removals in R1 recovered rapidly (within 1-3 days) after a 2-day, high pH shock (pH 10-11), even without reduced loading rates (Figs. 16-17). The shocks decreased DOC removals in R1 while those in R2 increased, resulting in only slightly decreased efficiency in R12. No washout of the attached biomass in the latter reactor was measured during disturbed operation. It would appear that the latter reactor thus greatly stabilized the process, suggesting that two-stage biological treatment is capable of producing steady COD_{filt} removals under varying operating conditions.

TABLE 14 Studies on thermophilic aerobic biofilm processes for treatment of pulp and paper mill wastewaters. Some mesophilic processes are given for reference.

Process	T °C	Scale	Wastewater	Loading rate kg COD $m^{-3}d^{-1}$	Sludge load kg COD kg $VSS^{-1}d^{-1}$	HRT h	COD removal %	Reference
BF	meso	pilot	Neutral paper mill effluent	1-10	nr.	0.3-0.65	30-91	Kantardjieff & Jones 1997
MBBR	29-35	pilot	Thermo mechanical GW	7.6-17.8	nr.	4-10	75-78	Broch-Due *et al.* 1997
MBBR	28	pilot	Secondary fiber mill	25 [a]	nr.	1.3	40-50	Dalentoft & Thulin 1997
MBR	55	lab	TMP whitewater	5	0.45	17	77	Tardif & Hall 1997
MBR	55	lab	TMP whitewater	4.1-12.4[a]	nr.	8-24	35-45	Ragona & Hall 1998
MBBR	55	lab	TMP whitewater	2.5-3.5	1.5-2.6	13-22	60-65	Jahren *et al.* 2002
MBBR	55	lab	TMP whitewater	1.6-12	0.8-6.2	8-24	15-55	Jahren & Ødegaard, 1999b
SCBP	40-50	full	CTMP effluent	100	nr.	1	15-25	Malmqvist *et al.* 1996
SCBP	50-52	lab	Liner	nr.	nr.	10-13	86-90	Malmqvist *et al.* 1999

[a]Calculated from reported feed COD and HRT, BF = Aerobic biofilter, CMTP = chemithermomechanical pulp, GW = groundwood pulp, GWM = groundwood mill water, MBR = Membrane bioreactor, MBBR = Moving bed biofilm reactor, meso = mesophilic, SCBP = Suspended carrier biofilm process, TMP = thermomechanical pulp, nr. = not reported.

5 CONCLUSIONS

The results obtained from this study show that thermophilic aerobic wastewater treatment is feasible and operable under different operational conditions, such as HRT and VLR. Both the thermophilic and mesophilic aerobic ASPs gave good COD_{filt} removals (70-90%) under VLRs and HRTs from 2 to 10 kg COD_{filt} $m^{-3}d^{-1}$ and 18 to 3 h, respectively.

Thermophilic treatment can produce the same COD_{sol} removal as that generated by mesophilic treatment, whereas COD_{filt} removal is less efficient. In this study higher effluent COD_{filt} values obtained were due to a high density of dispersed particles, such as free bacteria, and were characterised in more detail under thermophilic conditions as increased COD_{col} values. Mesophilic post-treatment efficiently reduces COD_{col} from thermophilic effluents (95-100% removal). Aeration of thermophilic effluent at 35°C, without introducing mesophilic microorganisms into the aeration vessel, can efficiently reduce COD_{col} (90%). However, in this study COD_{sol} was increased by 50%, apparently by lyses of thermophilic microorganisms. In the mesophilic ASP treating thermophilic effluent the increase in COD_{sol} was less marked. However, the combined process gave the same COD_{filt} removal than as that obtained from the single mesophilic ASP. The use of cationic polymer did not enhance the quality of either the thermophilic or mesophilic ASP effluent in terms of COD_{filt} removal.

ASP can be operated under varying temperatures, and operating conditions can be shifted rapidly from mesophilic to thermophilic and back without process failure. The fluctuating temperature (27-56°C) ASP gave similar COD_{filt} removals to those of the mesophilic ASP at temperatures between 27 and 40°C and the thermophilic ASP at temperatures between 45 and 56°C.

The thermophilic ASP and SCBP showed the same level of performance under moderate loading rates (VLR 2.7 to 3.3 kg COD_{filt} $m^{-3}d^{-1}$; HRT 18 to 12 h) in terms of COD_{filt} removal (60-62%). However, increasing the VLR to 5.7-6.0 kg

$\mathrm{COD_{filt}}$ $\mathrm{m^{-3}d^{-1}}$ (HRT 7-8 h) resulted in higher $\mathrm{COD_{filt}}$ removal (69%) in the thermophilic SCBP and lower $\mathrm{COD_{filt}}$ removal (48%) in the thermophilic ASP.

The thermophilic ASPs seems to have poor sludge settling properties, detected as weak and irregular-shaped flocs, low MLSS, and turbid effluents. However, the SVI values suggested excellent sludge settling properties under thermophilic conditions. Polymer dosing affected sludge settleability: thermophilic SVI values increased and mesophilic values decreased.

The in-mill thermophilic aerobic SCBP treatment of GWM on mill premises, which was done on the pilot-scale, was feasible under varying temperature (39-56°C) conditions. The benefits of a high-rate system appear especially to derive from the compact and relatively small reactor size; the two-stage process gave 60% $\mathrm{COD_{filt}}$ removals with HRT of 2 h. The two-stage thermophilic SCBP seemed particularly highly resistant to short-term process upsets and was operable under high VLRs (up to 15 kg $\mathrm{COD_{filt}}$ $\mathrm{m^{-3}d^{-1}}$).

In the thermophilic aerobic ASP and SCBP processes suspended sludge consisted mainly of dispersed particles, such as free bacteria, and of small flocs. Gram-negative bacteria dominated in both the thermophilic and mesophilic processes, and were more dominant in the settled sludge than in suspension, indicating that they were more likely to form flocs. Gram-positive bacteria increased when the VLRs were increased in the thermophilic processes. However, only 20% of gram-positive bacteria were viable. Gram-positive bacteria were apparently unable to form flocs; on the other hand, they were effectively retained in the biofilm. Filamentous bacteria participated in thermophilic floc formation, the number of filamentous bacteria in the thermophilic processes increasing with decreased HRTs. Unlike under thermophilic conditions, filament growth was suppressed by decreased HRTs in the mesophilic processes. None of the higher organisms were found in the thermophilic processes.

As a final conclusion, treatment of hot industrial wastewaters under aerobic thermophilic conditions appears to be feasible, resulting in similar $\mathrm{COD_{sol}}$ than under mesophilic conditions but requires that colloidal and suspended particles be removed if the effluent quality is to resemble that from mesophilic treatments.

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YHTEENVETO

Tuottavassa teollisuudessa, kuten paperin ja massan valmistuksessa, muodostuu kuumia prosessi- ja jätevesiä. Tyypillisesti vedet viilennetään alle 40°C:een ennen mesofiilista biologista jätevedenkäsittelyä. Termofiilinen, eli korkeassa lämpötilassa (55°C) tapahtuva, aerobinen jätevedenkäsittely tuo monia mahdollisuuksia teollisten jätevesien käsittelyyn. Termofiilinen aerobinen prosessi on yksinkertaisempi kuin mesofiilinen, koska lämmönvaihtimien tarve pienenee, ja siten prosessi on kustannustehokkaampi ja toiminnaltaan varmempi. Lämpötilan noustessa reaktionopeudet kasvavat, joten termofiilista prosessia voidaan operoida korkeammalla kuormituksella kuin mesofiilista, minkä ansiosta termofiilinen aerobinen jätevedenkäsittelyprosessi voidaan toteuttaa pienemmässä reaktorissa kuin mesofiilinen. Termofiiliselle aerobiselle jätevedenkäsittelyprosessille ominaista on matala lietteentuotto, mikä lisää prosessin kokonaiskustannustehokkuutta.

Tämänhetkisen tuotantokapasiteetin mukaan mitoitetut mesofiiliset aerobiset jätevedenkäsittelylaitokset voisivat käsitellä enemmän jätevesiä, esimerkiksi tuotannon kasvaessa, jos osa prosessista muutettaisiin termofiiliseksi. Toisaalta vedenkäytön vähentäminen voi nostaa jäteveden lämpötilaa. Lisäksi esimerkiksi paperi- ja massateollisuudessa on tarve pienentää ominaisvedenkulutusta, jolloin tuotannon sisäisten prosessivesikiertojen sulkeminen lisääntyy. Ilman suljetun kierron veden käsittelyä orgaaniset ja epäorgaaniset haittayhdisteet konsentroituvat vesikiertoon, mikä johtaa ongelmiin tuotannossa. Biologinen kiertovesien käsittely poistaa liukoisia, helposti hajoavia yhdisteitä, mitkä aiheuttavat mm. putkistojen ja laitteiden korroosiota, hajuhaittoja ja mikrobiologisia haittoja. Biologisen sisäisen prosessiveden käsittelyn toteuttaminen termofiilisena on mesofiilista käsittelyä edullisempi, koska käsiteltävää vettä ei tarvitse jäähdyttää ennen käsittelyä eikä lämmittää uudelleen käsittelyn jälkeen.

Tässä työssä tutkittiin termofiilista aerobista jätevedenkäsittelyä. Kokeet tehtiin laboratorio- ja pilot-mittakaavassa. Tutkitut aerobiset jätevedenkäsittelyt olivat joko aktiiviliete- tai kantajaprosesseja, joita operoitiin eri käsittelyviipymillä ja kuormituksilla sekä eri lämpötiloissa. Laboratoriokokeissa vertailuprosesseina käytettiin mesofiilisia aktiivilieteprosesseja. Käsiteltävä jätevesi laboratoriokokeissa oli laimennettu melassi, pilot-kokeissa käsiteltiin hiomon kiertovettä.

Tulosten mukaan termofiilinen aerobinen jätevedenkäsittely toimii eri kuormituksilla ja viipymillä. Termofiiliset ja mesofiiliset aktiivilieteprosessit poistivat hyvin (70-90%) laimennetun melassin GF/A-suodatettua COD:ta ($\mathrm{COD}_{\mathrm{filt}}$, huokoskoko noin 1.6 σm) 2-10 kg $\mathrm{COD}_{\mathrm{filt}}$ $\mathrm{m}^{-3}\mathrm{d}^{-1}$ kuormituksilla ja 18-3 h viipymillä. Termofiilinen prosessi poisti liukoista COD:ta ($\mathrm{COD}_{\mathrm{sol}}$, 0.45 σm –suodatettu COD) yhtä paljon kuin mesofiilinen, mutta $\mathrm{COD}_{\mathrm{filt}}$ vähenemät olivat alhaisempia. Tämä johtui termofiilisen prosessin vapaista bakteereista, jotka eivät muodostaneet flokkeja eivätkä laskeutuneet selkeyttimissä. Korkeammat $\mathrm{COD}_{\mathrm{filt}}$ –arvot olivat tarkemmin havaittavissa kolloidisena COD:na ($\mathrm{COD}_{\mathrm{col}}$, COD$_{filt}$ ja COD$_{sol}$ erotus), mikä kasvoi termofiilisen jätevedenkäsittelyn aikana. Termofiilisessa prosessissa käsitellyn jäteveden mesofiilinen jälkikäsittely poisti 90-100% COD$_{col}$:a. COD$_{col}$-arvo vähentyi pelkässä jälki-ilmastuksessa, jolloin tosin COD$_{sol}$-arvo kasvoi (50%), johtuen termofiilisten mikro-organismien liukoisista kuolemistuotteista. Kun jälkikäsittely toteutettiin mesofiilisessa aktiivilieteprosessissa COD$_{sol}$-arvot eivät nousseet. Yhdistetty termofiilinen-mesofiilinen aktiivilietekäsittely poisti COD$_{filt}$-arvoja yhtä paljon, mutta ei enempää kuin yksivaiheinen mesofiilinen aktiivilieteprosessi. Kationisen polymeerin käyttö rinnakkaissaostuksessa ei vaikuttanut termofiilisesti tai mesofiilisesti käsitellyn jäteveden COD-arvoihin.

Aktiivilieteprosessin toimintaa tutkittiin myös vaihtelevissa lämpötiloissa. Prosessin lämpötilaa vaihdeltiin satunnaisesti 27-56°C välillä. Aktiivilieteprosessi oli toimiva myös vaihtelevissa lämpötiloissa vähentäen COD$_{filt}$-arvoja miltei yhtä paljon korkeissa lämpötiloissa (45-56°C) kuin termofiilinen (55°C) aktiivilieteprosessi ja matalissa lämpötiloissa (27-40°C) kuin mesofiilinen (35°C) aktiivilieteprosessi.

Termofiilinen aktiiviliete- ja kantajaprosessi vähensivät COD$_{filt}$-arvoja yhtä paljon (60-62%) 18-12 h viipymillä (vastaten 2.7-3.3 kg COD$_{filt}$ m^{-3}d^{-1} kuormituksia), kuormituksen kasvaessa (7-8 h viipymä, 5.7-6.0 kg COD$_{filt}$ m^{-3}d^{-1} kuormitus) aktiivilieteprosessin COD$_{filt}$ –vähenemät alenivat (48%) ja kantajaprosessin nousivat (69%). Ilmeisesti kantajaprosessi toimii paremmin korkeammilla kuormituksilla kuin aktiivilieteprosessi.

Termofiilisten aktiivilieteprosessien toimintaa heikensi käsitellyn jäteveden mukana karkaavat vapaat bakteerit sekä lietteenpalautuksen ongelmat, mikä johti reaktorien alhaisiin lietepitoisuuksiin. Tästä huolimatta termofiilisen lietteen SVI-arvot (lieteindeksi, joka kuvaa lietteen laskeutuvuutta) olivat erinomaiset. Kationisen polymeerin käytöllä oli vaikutusta lietteen laskeutuvuuteen, termofiilisen lietteen SVI-arvot kasvoivat ja mesofiilisen laskivat.

Pilot-mittakaavan termofiilinen aerobinen kaksivaiheinen kantajaprosessi oli toimiva poistaen 60-70% hiomon kiertoveden orgaanisesta kuormasta vaihtelevissa lämpötiloissa (39-56°C) ja suurilla kuormituksilla (<15 kg COD$_{filt}$ m^{-3}d^{-1}, 2 h. viipymä). Kaksivaiheinen termofiilinen kantajaprosessi toipui nopeasti (24 h) häiriötilanteista, kuten hetkellisistä (<12 h) pH-shokeista (pH 11-12).

Termofiilisten aerobisten prosessien suspensiossa oleva liete koostui pääosin vapaista bakteereista ja pienistä, heikoista flokeista. Gram-negatiiviset bakteerit olivat vallitsevia erityisesti laskeutuneissa flokeissa. Gram-positiivisten bakteerien osuus kasvoi kuormituksen noustessa, mutta ainoastaan noin 20% gram-negatiivisista bakteereista oli eläviä. Rihmamaiset bakteerit vaikuttivat termofiilisten flokkien muodostumiseen sitomalla flokkeja toisiinsa. Kuormituksen kasvu lisäsi termofiilisten rihmamaisten bakteerien esiintymistiheyttä, toisin kuin mesofiilisissa prosesseissa, joissa tiheys väheni.

Termofiilinen jätevesien käsittely on mahdollista korkeillakin kuormituksilla ja lyhyillä käsittelyajoilla, mutta saavuttaakseen saman laadun kuin mesofiilisissa prosesseissa, on käsittelyä täydennettävä hienoaineksen poistamiseksi.

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