Biological sulphate reduction with primary sewage sludge in an upflow anaerobic sludge bed (UASB) reactor – Part 3: Performance at 20°C and 35°C

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Abstract

The performance of 2 biological sulphate reduction (BSR) upflow anaerobic sludge bed (UASB) reactors fed primary sewage sludge (PSS) and sulphate, one at 20°C (R2) and one at 35°C (R1) is described. To maintain the effluent sulphate concentration below 250 mgSO$_4^{2-}$/ℓ, the hydraulic retention time (HRT) and bed solids retention time (SRT or sludge age) both needed to be longer and the feed primary sewage sludge (PSS) COD to SO$_4^{2-}$ ratio higher at 20°C than at 35°C, viz. 20.4 to 21.0 h, 24 d and 1.75 gCOD/gSO$_4^{2-}$ at 20°C and 16.4 to 17.0 h, 21 d and 1.75 gCOD/gSO$_4^{2-}$ at 35°C respectively. The longer HRT, SRT and higher feed PSS COD/ SO$_4^{2-}$ ratio is a consequence of a slower PSS hydrolysis/acidogenesis rate at 20°C resulting in a lower biodegradable particulate organics conversion to volatile fatty acids (VFA). Solid liquid separation in both systems was good yielding average particulate and soluble organic COD concentrations of (150 and 100 mgCOD/ℓ for R1; 138 and 96 mgCOD/ℓ for R2). The sulphate reduction was >90% in both systems. The UASB reactor R1 (at 35°C) was also operated at an increased influent sulphate concentration (1 800 mgSO$_4^{2-}$/ℓ) to investigate the inhibition effect by un-dissociated hydrogen sulphide generated from the reduction of this high sulphate concentration. It was found that a high sulphate reduction (~ 92%) was maintained even at the relatively low HRT of 18.5 h. The COD and S mass balances above 95% were achieved over both systems indicating that the performance data obtained from them is reliable for developing and calibrating mathematical models.

Keywords: biological sulphate reduction, hydrolysis, hydraulic retention time, UASB reactor

Nomenclature

Alk H$_2$S	alkalinity with respect to the H$_2$S reference species excluding the water species
AMD	acid mine drainage
BPO	biodegradable particulate organics
BRT	bed retention time
BSR	biological sulphate reduction
COD	chemical oxygen demand
FBR	fluidised bed reactor
f_{cv}	COD to VSS ratio
f_n	orgN/VSS ratio
$f_{PS'up}$	influent and biodegradable particulate COD fraction of primary sludge
FRBCOD	fermentable readily biodegradable COD
FSA	free and saline ammonia
H$_2$CO$_3$*Alk	alkalinity with respect to the H$_2$CO$_3$ reference species including the water species
HAc	acetic acid
HRT	hydraulic retention time
K_I	sulphide inhibition kinetic constant
OLR	organic loading rate
PBR	packed bed reactor
pH	negative log of hydrogen ion activity
pK'_{S1}	1st dissociation constant for the sulphide weak acid base system corrected for ionic strength effects.
PSS	primary sewage sludge
R1	UASB Reactor 1
R2	UASB Reactor 2
R_s	sludge age
S_{bp}	biodegradable particulate COD concentration
SBR	sequencing batch reactor
SCFA	short chain fatty acids
SLR	sludge loading rate
SRB	sulphate reducing bacteria
SRT	solids retention time
SS	steady state
SSD	sample standard deviation
S_{up}	un-biodegradable particulate COD concentration
TKN	total Kjeldahl nitrogen
Total Alk	sum of weak acid/base subsystem alkalinities
UASB	upflow anaerobic sludge bed reactor
UPO	un-biodegradable particulate organics
USCOD	un-biodegradable soluble COD
V_b	bed volume
VFA	volatile fatty acids
V_{up}	hydraulic upflow velocity in UASB reactor

Introduction

The feasibility of a novel system for BSR of AMD using PSS as carbon source in a UASB reactor configuration (R1) was described in Part 1 (Poinapen et al., 2009a). From the successful operation of the UASB reactor R1 at 35°C (fed 1 500 mgSO$_4^{2-}$/ℓ for a period of 280 d), the performance of BSR using PSS was evaluated also in a second identical UASB reactor (R2) operated in parallel to R1 but at ambient (20°C)

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Received 13 January 2009; accepted in revised form 31 July 2009.

TABLE 1
Summary of operational conditions for UASB reactor R2 during the 270 d of operation at 20°C (see timeline Fig. 2 in Poinapen et al., 2009a)

Day of investigation	Day of operation of R2	Reactor status/activities performed	HRT (h)	BRT (h)
280-350	**1-70**	**Feed PSS COD to sulphate ratio 1.33 and sludge bed volume 7.1 ℓ**		
280-330	1-50	SRB acclimatisation	28.0	21.8
331-350	51-70	R2 SS 1	28.0	25.5
371-392	91-112	R2 SS 2	24.0	21.8
395-550	**115-270**	**Feed PSS COD to sulphate ratio 1.75 and sludge bed volume 7.4 ℓ**		
398-419	118-139	R2 SS 3	24.0	22.8
424-431	144-151	R2 SS 4	22.0	20.9
434-550	154-270	R2 SS 5	20.4-21.0	19.4-19.9
406-435	126-155	CHON elemental analysis of waste sludge		
448 & 463	168 & 183	Bed profile tests (both on solubles and solids)		
448 & 435	168 & 183	Determination of sludge age	20.4-21.0	19.4
430-538	150-258	H_2S gas capture and measurement*	20.4-21.0	19.4
465-530	185-250	Stopped $NaHCO_3$ dosage to feed	20.4-21.0	19.4
398-538	118-258	COD, S and N mass balances calculated		

** H_2S gas measurement done only when conducting sulphur mass balances over the system.*

temperature. Accordingly, from Day 280 of the investigation, Reactor R2 (T=20°C) was operated with feed sulphate and PSS concentrations of 1 500 mgSO_4^{2-}/ℓ and 1995 mgCOD/ℓ respectively, representing a feed PSS COD to sulphate ratio of 1.33 mgCOD/mgSO_4^{2-}. This ratio for R2 was slightly higher than the R1 ratio of 1.25 because the hydrolysis/acidogenesis rate of PSS at 20°C is slower than at 35°C, but how much slower was not known. Later on Day 395, the PSS COD/SO_4^{2-} R2 ratio was increased to 1.75 (see Fig. 2 in Poinapen et al., 2009a) to reduce the effluent sulphate concentration to below 250 mgSO_4^{2-}/ℓ. In this paper, the operation and performance of UASB R2 at 20°C are assessed, evaluated and compared with that of UASB R1 at 35°C.

After the feasibility study with R1 from Day 1 to 280 (Poinapen et al., 2009a), the influent sulphate concentration to R1 was increased from 1 500 mgSO_4^{2-}/ℓ to 1 800 mgSO_4^{2-}/ℓ. It was expected that the higher feed sulphate concentration would result in a higher sulphide concentration in the system. Even though compared with other anaerobic micro-organisms (namely methanogenic archaea) the sulphate SRB have the highest tolerance to sulphide, their activity is nevertheless affected by the presence of sulphide, especially un-dissociated hydrogen sulphide (H_2S). Un-dissociated H_2S is inhibitory to SRB and decreases their growth activity (Reis et al., 1992; Maillacheruvu et al., 1993; Konishi et al., 1996; Kalyuzhnyi et al., 1997; O'Flaherty et al., 1998). Investigating the effect of high feed sulphate concentration on the system performance was therefore necessary in order to assess the extent of un-dissociated hydrogen sulphide (H_2S) inhibition on BSR using PSS as substrate in a UASB reactor.

Methodology

Reactor R2 (T=20°C) was inoculated with waste sludge from R1 (Day 280) and operated for 270 d to Day 550. Initially, R2 performance was not as good as R1, despite complete seeding with R1 sludge. The relatively short HRT of ~28 h at which R2 was operated during the start-up period was probably too short. It was thought that R1 waste sludge would result in a quick and effective start-up of R2 biological processes since R1 was at its maximum efficiency. However, it appeared that an adaptation period was required for the selected population group of sulphidogens to develop at the lower temperature (20°C). After 50 d of operation to Day 330, granulation was observed and the system performance started to stabilise. The effluent sulphate concentration remained stable from periods Day 333-350 and Day 371-392 with a sulphate removal of 83.7% and 79.8% respectively, even though the HRT was incrementally decreased from 28 h to 24 h (see Table 5 later), implying that the reactor was at steady state. However, the effluent sulphate concentration (303 mgSO_4^{2-}/ℓ) was still above the specified requirement (250 mg SO_4^{2-}/ℓ in South Africa). In addition, during this period the VFA concentration was low suggesting that PSS hydrolysis was incomplete and was the rate-limiting step under the lower temperature conditions.

During the 270 d of operation of R2 (i.e. from investigation Day 280 to 538, or equivalently from R2 Day 0 to Day 258), the influent feed flow rate was varied to vary the HRT. The UASB R2 operational conditions during the 270 d are set out in Table 1. The different steady state periods with respect to the operating HRT with 2 PSS COD/SO_4^{2-} ratios, the days at which bed profile tests were conducted, the period when alkalinity dosage was stopped and the days on which mass balances were determined are listed.

Influent characteristics

Similar to UASB Reactor R1 (Poinapen et al., 2009a), the prepared feed for R2 (from dilution of the PSS) augmented with 1 500 mgSO_4^{2-}/ℓ was regularly analysed for its COD, TKN, FSA, VFA and H_2CO_3* alkalinity concentrations and pH. Two influent PSS COD/SO_4^{2-} ratios (namely 1.33 and 1.75 mg COD/mgSO_4^{2-}) were applied. A summary of the results from the measurements of the above parameters over the 2 steady state periods at feed COD/SO_4^{2-} ratio = 1.33 (SS2, R2 operation Day 371 to Day 392) and 1.75 (SS5, R2 operation Day 520 to Day 538) is listed in Table 2.

It was expected that at 20°C, the hydrolysis/acidogenesis of biodegradable particulate organics (BPO) would not be complete but no information was available to indicate what

TABLE 2
Summary of the influent characteristics of UASB reactor R2 (1 500 mgSO$_4^{2-}$/ℓ at 20°C) for steady state (SS) periods 2 and 5

Feed PSS COD to sulphate ratio (mgCOD/mgSO$_4^{2-}$)	**1.33**		**1.75**	
Steady state period	**SS 2 (R2 Day 371-392)**		**SS 5 (R2 Day 520-538)**	
Parameter	**Average value**	**SSD[1]**	**Average value**	**SSD[1]**
Total COD (mgCOD/ℓ)	1975	7	2596	78
Unbiodegradable particulate COD ($f_{PS'up}$ = 0.36)[2] (mgCOD/ℓ)	711	-	935	-
VFA (or SCFA) (mgHAc/ℓ)[3]	123	7	158	11
Fermentable readily biodegradable (FRBCOD) COD (mgCOD/ℓ) [3, 4]	139	-	162	-
Slowly biodegradable COD (mgCOD/ℓ)	987	-	1322	-
Unbiodegradable soluble (mgCOD/ℓ)[5]	7	-	8	-
TKN (mgN/ℓ)	71	18	78	18
FSA (mgN/ℓ)	8	5	10	4
H_2CO_3* alkalinity (mg/ℓ as $CaCO_3$) before dosage	18	3	23	3
H_2CO_3* alkalinity (mg/ℓ as $CaCO_3$) after dosage	423	6	429	15
Total phosphorous (mgP/ℓ)	28	-	33	-
Ortho-phosphate (mgP/ℓ)	8	-	10	-
pH before dosage	5.95	0.05	5.94	0.05
pH after dosage	7.25	0.10	-	-

[1] SSD = Standard deviation
[2] Accepted from Ristow et al. (2006) for the same PSS source.
[3] VFA and FRBCOD high due to fermentation in PSS during storage at 4°C
[4] From membrane (0.45 µm) filtered COD minus VFA and USCOD
[5] Un-biodegradable soluble COD (USCOD) very low – after dilution with tap water (>10 times)

PSS COD/SO$_4^{2-}$ ratio was appropriate to supply the required VFA from the hydrolysis/acidogenesis of PSS at 20°C. After a few trial ratios, it was found that a ratio of 1.75 appeared most favourable to supply sufficient VFA for BSR, i.e. effluent sulphate < 250 mg SO$_4^{2-}$/ℓ. The results obtained from the 1.33 and 1.75 ratios are discussed below. Also, towards the end of the experimental programme on UASB R2 (R2 Day 465 to 530), influent alkalinity ($NaHCO_3$) dosage (~ 400 mg/ℓ as $CaCO_3$) was stopped in order to ascertain the pH at which the system would stabilise itself.

Results

Effluent sulphate concentration with time

The variation in UASB R2 effluent sulphate concentration with time from R2 Day 290 is shown in Fig. 1 for the whole experimental period of R2 at the 2 feed PSS COD/SO$_4^{2-}$ ratios. For the period R2 Day 331 to Day 392 (comprising SS Periods 1 and 2), on average the effluent sulphate was not reduced below 250 mgSO$_4^{2-}$/ℓ because of the low PSS COD/SO$_4^{2-}$ ratio (1.33) and consequently low VFA generation by hydrolysis/acidogenesis. From R2 Day 395 onwards the feed COD/SO$_4^{2-}$ ratio was increased to 1.75 which resulted in a marked decrease in the effluent sulphate concentration to around 100 mgSO$_4^{2-}$/ℓ. Clearly VFA generation via hydrolysis/ acidogenesis was the limiting rate for BSR.

Effluent H_2CO_3* alk, Alk H_2S and VFA concentrations with time

As expected, the effluent total Alk (sum of H_2CO_3* alk, Alk H_2S and Alk VFA) increased due to the increase in the PSS COD/SO$_4^{2-}$ ratio (R2 Day 395 to Day 450 in Fig. 2) as a direct consequence of an increase in sulphate reduced, hence increasing the total sulphide ($S_T = H_2S+HS^-$), dissociated sulphide (HS^-) and Alk H_2S (proportional to HS^-) concentrations.

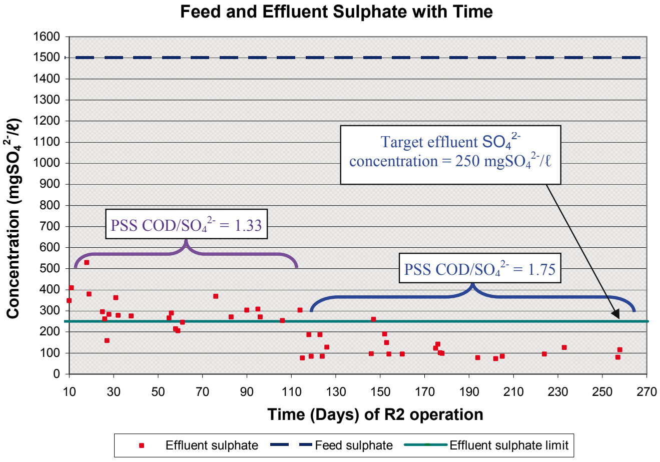

Figure 1
Effluent sulphate concentration-time profiles with PSS as substrate augmented with 1 500 mgSO$_4^{2-}$/ℓ sulphate (R2)

The data points circled in Fig. 2 represent the period (R2 Day 475 onwards) over which the feed $NaHCO_3$ dosage was progressively decreased to zero by R2 Day 510. This elimination of feed alkalinity dosage did not affect the system performance in any way as shown in Figs. 1 and 2 by the low sulphate and VFA concentrations. This is an important result for the UASB BSR performance as it shows that BSR:

- Occurs successfully at low temperature (20°C)
- Regulates the reactor pH near neutrality (~7) by itself even though the sulphate rich AMD is acidic. This is because the BSR bioprocesses reduce sulphate to sulphide and take up 2 protons (H^+) per mole SO$_4^{2-}$, i.e. $SO_4^{2-} + 2H^+ + 8(H^+ + e^-$, supplied by the organics) $\rightarrow H_2S + 4H_2O$.

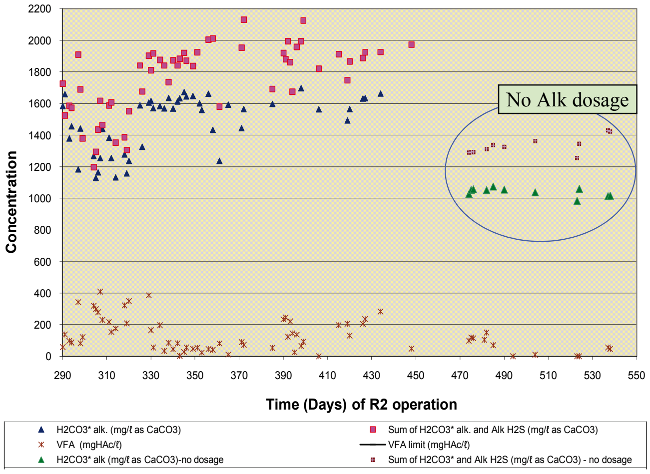

Figure 2

Effluent alkalinity and VFA concentration-time profiles with PSS as substrate augmented with 1 500 mgSO$_4^{2-}$/ℓ sulphate (R2)

The stoichiometry of BSR with organics in general and PSS in particular, are considered in Part 5 of this series, currently in preparation by Poinapen and Ekama.

Reactor performance with COD/SO$_4^{2-}$ ratio of 1.33

Table 3 shows the performance of UASB Reactor R2 when operated at an HRT of 24 h (SS2). It can be seen that R2 performed satisfactorily because it achieved a sulphate removal efficiency of ~80% (± 2%) and an organic removal efficiency of ~77% (± 1%). However, despite the relatively high sulphate removal efficiency, the average effluent sulphate remained above the upper limit of 250 mgSO$_4^{2-}$/ℓ. The cause for this fairly high effluent sulphate concentration (~ 303 mgSO$_4^{2-}$/ℓ) was attributed to the unavailability of sufficient VFA which remained very low in the effluent, despite the higher feed PSS COD/SO$_4^{2-}$ ratio of 1.33 compared with the 1.25 of R1. It was concluded that at low temperature, the hydrolysis rate of the PSS biodegradable particulate organics (BPO) was reduced thereby generating insufficient VFA and H_2 for sulphate reduction.

In order to increase the production of VFA, the PSS COD/SO$_4^{2-}$ ratio was increased to 1.75 which increased the feed BPO COD concentration and consequently the concentration of BPO hydrolysed, thereby generating more VFA and H_2 for BSR. Increasing the PSS COD/SO$_4^{2-}$ ratio also resulted in higher un-biodegradable particulate organics (UPO, S_{up}) and BPO (S_{bp}) not hydrolysed. Hence more particulate organics needed to be wasted to maintain the bed volume. So to provide a larger bed volume (and concomitantly a longer sludge age), the reactor sludge bed volume (and height) was increased from 7.1 to 7.4 ℓ. The increased sludge age allowed more time for the BPO to be hydrolysed as PSS hydrolysis is the rate-limiting process.

R2 performance with a PSS COD/SO$_4^{2-}$ ratio of 1.75 at lowest optimal HRT of 20.4 to 21.0 h

At the higher feed PSS COD/SO$_4^{2-}$ ratio of 1.75, the performance of R2 improved to the extent that the effluent quality concentrations were now met (i.e. sulphate <250 mgSO$_4^{2-}$/ℓ and VFA <100 mgHAc/ℓ). The performance of UASB Reactor R2 at the lowest optimal HRT of 20.4 to 21.0 h with alkalinity dosage (~ 424 mg/ℓ as CaCO$_3$) from R2 Day 434 to Day 464 (SS period 5a) was good (see Table 4) with an average sulphate removal efficiency of 93.3 ± 1.5%.

Influent alkalinity (NaHCO$_3$) dosage was discontinued from R2 Day 465 to the end of the experimental programme on R2 Day 550. Table 4 lists also the effluent parameter values when R2 was operated at an HRT of between 20.4 h and 21.0 h with no alkalinity dosage to the feed for the period Day 520 to Day 538. With the supply of additional carbon (i.e. more PSS), the sulphate removal efficiency increased from 80% (± 2.0%), when fed a PSS CO/SO$_4^{2-}$ ratio of 1.33, to > 93% (± 1.3%), with PSS COD/SO$_4^{2-}$ ratio of 1.75. From the high organic COD removal efficiency of 91% (± 1.3%) it was found that the increase in PSS to meet the VFA demand did not impact the effluent quality in terms of suspended solids. In fact, the particulate organic COD concentration remained low with an average value of 138 (± 25) mgCOD/ℓ.

The aqueous sulphide concentration ($S_T = H_2S + HS^-$) was measured via the COD with the modified method described by Poinapen et al. (2009b). This sulphide concentration matched very closely the theoretical sulphide produced from the sulphate reduced (see below for S mass balance). In addition, discontinuing the alkalinity dosage to the influent (from R2 for the period Day 468 to Day 550) did not affect the system performance in any way. The alkalinity produced by the BSR increased the pH from the influent value of 5.94 to a reactor value of 7.21. This shows that the system is pH self-regulatory, which is a major advantage of BSR to reduce the cost of chemical dosing in AMD treatment.

TABLE 3
Effluent characteristics of UASB reactor R2 with an influent PSS COD to sulphate ratio of 1.33 mgCOD/mgSO$_4^{2-}$ at an HRT of 24 h (bed volume = 7.1 ℓ)

Steady state (SS) period 2 (R2 Day 371 to Day 392)	Influent		Effluent	
Parameter	**Average value**	**SSD**	**Average value**	**SSD**
Total organic COD (mgCOD/ℓ)	1 975	7	453	13
Organic soluble COD (mgCOD/ℓ)	277	7	146	8
Particulate COD (mgCOD/ℓ)	1 698	7	307	9
VFA (or SCFA) (mgCOD/ℓ)	131	7	49	20
H_2CO_3* alkalinity (mg/ℓ as CaCO$_3$) after dosage	424	7	1 553	25
pH	7.25[a]	0.10	7.21	0.02
Sulphate (mgSO$_4^{2-}$/ℓ)	1 500	-	303	30
Aqueous sulphide (mgS/ℓ)	0	-	216	5
Theoretical aqueous sulphide from sulphate reduced (mgS/ℓ)	-	-	399	11

[a]*: Feed pH after dosage with NaHCO$_3$*

TABLE 4
Effluent characteristics of UASB reactor R2 at a feed PSS COD to sulphate ratio of 1.75 mgCOD/mgSO_4^{2-} at the lowest optimal HRT of 20.4-21.0 h (Bed volume = 7.4 ℓ) with and without alkalinity dosage to feed (SS period 5, R2 Day 434 to Day 538)

Steady state (SS) period 5 (Day 434 to Day 538)	Influent		Steady state (SS) period 5a (R2 Day 434 to Day 464) (Alk dosage)		Steady state (SS) period 5b (R2 Day 520 to Day 538) (No Alk dosage)	
			Effluent		Effluent	
Parameters	**Average value**	**SSD**	**Average value**	**SSD**	**Average value**	**SSD**
Total organic COD (mgCOD/ℓ)	2 596	44	664	102	234	27
Organic soluble COD (mgCOD/ℓ)	339	17	305	66	96	8
Particulate COD (mgCOD/ℓ)	2 257	-	359	42	138	25
VFA (or SCFA) (mgCOD/ℓ)	158	13	105	47	62	11
Sulphate (mgSO_4^{2-}/ℓ)	1 500	-	109	18	101	19
Sum of H_2CO_3* alk. and Alk H_2S (mg/ℓ as $CaCO_3$)	424[a] / 23[b]	11 / 3	1 920	27	1558	14
Total alk. (incl. Alk VFA) (mg/ℓ as $CaCO_3$)	548[a] / 148[b]	11 / 9	2 007	29	1610	13
H_2CO_3* alkalinity (mg/ℓ as $CaCO_3$)	424[a] / 23[b]	11 / 3	1 589	11	1144	10
TKN (mgN/ℓ)	78	18	63	9	63	9
FSA (mgN/ℓ)	9.6	4	37	12	38	5
pH	5.94	0.04	7.22	0.02	7.21	0.02
Aqueous sulphide (mgS/ℓ)	0	-	344[c]	39	427	5
H_2S gas in reactor head space (mgS/ℓ influent)	0	-	1.5	1.0	2.1	0.3
Theoretical sulphide from sulphate reduced (mgS/ℓ)	0	-	464	6	466	6

[a]: *With alkalinity dosage to influent*
[b]: *Without alkalinity dosage to influent*
[c]: *H_2S measurement problem. However, S_T was accurately measured from Day 520 to Day 539 using the modified wet chemistry analytical procedures described in Part 2 – Poinapen et al. (2009b).*

TABLE 5
Sulphate and organic COD removal (%) under steady state conditions at each HRT and BRT

Period of operation	SS period	HRT (h)[a]	BRT (h)	Feed flow rate (ℓ/d)	Organic loading rate (g COD/d)	Sulphate loading rate (g SO_4/d)	Sulphate removal (%)	Organic COD removal (%)
Days 333-350	1	28.0	25.8[b]	6.7	13.06	10.03	83.7	75.9
Days 371-392	2	24.0	22.1[c]	7.8	19.88	11.70	79.8	76.8
Days 398-419	3	24.0	22.2[c]	7.8	20.20	11.70	91.1	80.1
Days 424-431	4	20.8	19.2[c]	9.0	23.28	13.48	88.2	74.6
Days 434-464	5a	20.5	19.4[c]	9.1	24.14	13.70	92.7	75.2
Days 520-538[d]	5b	20.4	19.4[c]	9.2	24.71	13.76	93.3	91.2
Days 398-419	3	24.0	22.2[c]	7.8	20.20	11.70	91.1	80.1
Days 424-431	4	20.8	19.2[c]	9.0	23.28	13.48	88.2	74.6
Days 434-464	5a	20.5	19.4[c]	9.1	24.14	13.70	92.7	75.2
Days 520-538[d]	5b	20.4	19.4[c]	9.2	24.71	13.76	93.3	91.2

[a]: *HRT calculated at a liquid volume of 7.8 ℓ*
[b]: *BRT calculated at sludge volume of 7.1 ℓ with feed PSS COD to Sulphate ratio of 1.33*
[c]: *BRT calculated at sludge volume of 7.4 ℓ with feed PSS COD to Sulphate ratio of 1.75*
[d]: *No $NaHCO_3$ dosage to influent*

Summary of UASB R2 performance

A summary of the average experimental data at the different HRTs and BRTs is listed in Table 5. The minimum HRT for which the specified effluent criteria were met was found to be between 20.4 to 21.0 h, where sulphate removal efficiency was ~ 95% at a PSS COD/SO_4^{2-} ratio of 1.75.

Profile of solids – Calculation of sludge age (R_s)

Figure 3 illustrates the variation of R2 solids concentration along the bed axis. Above the 10 cm height there is little difference in the solids concentration along the bed axis probably largely due to the sludge recycle (at half the influent flow). From these measurements, the mass of sludge in the bed was calculated, which in relation to the mass of sludge wasted (see below) and the mass of sludge in the effluent, yielded the sludge age (R_s) of the UASB Reactor R2 at around 24.1 d.

Analysis of R2 waste sludge

UASB R2 waste sludge was analysed on 6 operational days

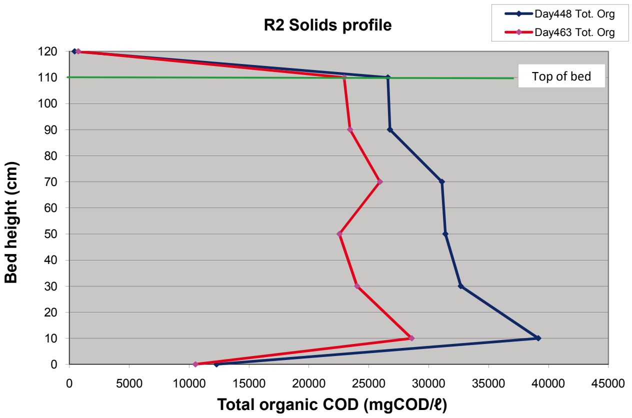

Figure 3
UASB Reactor R2 sludge COD concentration profiles up the sludge bed height (110 cm) for a BRT of 19.3 h on R2 Day 448 and Day 463

(Table 6) and was found to have an average particulate organic COD/VSS (f_{cv}) and TKN/VSS (f_n) of 1.611 ± 0.041 and 0.068 ± 0.016 respectively.

Mass balances on R2

Mass balances were conducted on R2 to check experimental data reliability. Similar to UASB R1 when fed 1 500 $mgSO_4^{2-}$/ℓ during the first 280 d (Poinapen et al., 2009a), prior to the modification of the analytical procedures for accurate measurement of aqueous sulphide, the R2 system COD mass balance was relatively low (91.7 ± 5.2%) while the S mass balance was poor (69.4 ± 6.4%). However, these mass balances, particularly the S mass balance, improved considerably with the modified analytical method for H_2S measurement. The COD mass balance increased to 98.0% (± 0.2%) while the S mass balance averaged 95.9% (± 0.2%). These good mass balances confirm the reliability of the measured data and the modified methods for aqueous H_2S measurement (Poinapen et al., 2009b). Nitrogen (N) mass balance was also very good and averaged 104.5 (± 4.9%).

Operation and performance of R1 fed 1 800 $mgSO_4^{2-}$/ℓ

During the initial phase of this investigation (R1 Day 0 to Day 280), the design and operational parameters for UASB R1 (T=35°C) fed 1 500 $mgSO_4^{2-}$/ℓ were established (R1 Day 1 to Day 280, Poinapen et al., 2009a). Sulphate, H_2CO_3* alkalinity and VFA concentration profiles through the sludge bed were conducted to gain insight into the bioprocess behaviour along the reactor bed. Solids profile tests were used to determine the sludge age of the system at the minimum HRT (13.5 to 14.0 h) at which effluent quality criteria were met (i.e. effluent sulphate < 250 $mgSO_4^{2-}$/ℓ and effluent VFA < 100 mgHAc/ℓ). After the initial phase of R1, the influent sulphate was increased to 1 800 $mgSO_4^{2-}$/ℓ on R1 Day 280 to investigate the effect of higher sulphate concentration on the system. It is well documented that the biological treatment of sulphate-rich wastewater involves the potential effect of sulphide inhibition on bacterial growth and activity. [*In the BioSURE® process design for AMD treatment, the treated effluent is recycled to blend with the raw AMD. The high alkalinity (sulphide and carbonate alkalinities) in the recycle stream neutralises the pH, precipitates the heavy metals and almost halves the inflow sulphate concentration of the raw AMD. Therefore, in a situation where an effluent sulphate of 200* $mgSO_4^{2-}$*/ℓ is recycled on a 1:1 effluent to incoming raw AMD flow rates, the raw AMD can have a feed sulphate concentration of up to 3 400* $mgSO_4^{2-}$*/ℓ*]. Operating R1 with a higher feed sulphate of 1 800 $mgSO_4^{2-}$/ℓ gave insight into the extent of sulphide inhibition on the bioprocesses in the system. The feed PSS COD concentration was also increased to conform to a feed PSS COD/sulphate ratio of 1.44 $mgCOD/mgSO_4^{2-}$. This higher ratio than 1.25 was applied to ensure that BSR in the system was not VFA limited. The operation and performance of R1 at 1 800 $mgSO_4^{2-}$/ℓ are described and analysed below.

R1 experimental programme (fed 1800 $mgSO_4^{2-}$/ℓ)

R1 was fed 1 800 $mgSO_4^{2-}$/ℓ from Day 281 to Day 530 (the day it was stopped). It took around 20 d for the reactor to reach the first steady state at which the effluent sulphate concentration (< 250 $mgSO_4^{2-}$/ℓ) was met at an HRT of 19 h. Thereafter, the HRT was gradually decreased (by increasing the influent flow rate) to establish the lowest optimal HRT. It was found that at an HRT below 18 h, the effluent sulphate concentration could not be maintained below 250 $mgSO_4^{2-}$/ℓ either due to sludge bed expansion (which reduces the bed sludge age) and/or insufficient sulphate contact time with the biomass. Accordingly, the lowest HRT was set at 18.0 to 18.5 h. The BRT was 16.4 to 16.8 h based on a sludge bed volume of 7.1ℓ and the upflow velocity, V_{up}, was 0.093 to 0.096 m/h. The sludge age determined from the bed profile test was 21 d. The various

TABLE 6
Analysis of waste sludge from the UASB Reactor R2 (fed 1 500 $mgSO_4^{2-}$/ℓ)

Day of operation	Total COD (mgCOD/ℓ)	Particulate COD (mgCOD/ℓ)	TSS (mg/ℓ)	VSS (mg/ℓ)	TKN (mgN/ℓ)	COD/VSS (f_{cv})	TKN/VSS (f_n)	Volume wasted per day*, Q_w (ℓ/d)	Operating sludge age (d)
398	21 336	20 056	16 120	12 368	798	1.622	0.065	0.35	22.4
406	20 117	18 862	14 798	11 378	1 113	1.658	0.098	0.35	22.4
419	28 651	27 174	22 138	16 906	1 361	1.607	0.080	0.30	26.2
426	32 458	30 977	26 398	20 150	1 005	1.537	0.050	0.32	24.5
427	27 418	25 745	21 466	16 220	980	1.587	0.060	0.40	20.0
434	37 498	35 832	28 340	21 648	1 253	1.655	0.058	0.31	25.3
Mean	27 913	26 441	21 543	16 445	1 085	1.611	0.068	0.34	23.5
SSD	6 019	5 888	4 917	3 727	185	0.041	0.016	0.03	2.1

** This was the volume of sludge wasted per day on these 6 analysis days. The average volume wasted daily over this operational period was 0.34 ℓ/d to maintain the sludge bed volume at 7.4 ℓ.*

TABLE 7
Summary of operational conditions for UASB Reactor R1 fed 1 800 mgSO$_4^{2-}$/ℓ during the 250 d of operation at 35°C

Day of operation	Reactor status/activities performed	HRT (h)	BRT (h)
281-530	Feed PSS COD to sulphate ratio 1.44 and sludge bed volume 7.1 ℓ		
302-330	R1 Steady state (SS) 1	19.0	17.1
365-530	R1 SS 2	18.0-18.5	16.4-16.8
295, 297, 302, 304, 306, 326, 332, 389 and 478	Bed profile tests (both on solubles and solids)	18.0-19.0	16.4-17.1
362-478	H_2S gas capture and measurement*	18.0-19.0	16.4-17.1
400-473	Stopped $NaHCO_3$ dosage to feed	18.0-18.5	16.4-16.8
475-530	Stopped sludge recycle line	18.0-18.5	16.4-16.8
362-478	COD, S and N mass balances calculated	18.0-18.5	16.4-16.8
295, 297, 304, 306, 332 and 389	Determination of sludge age	18.0-19.0	16.4-17.1
525-530	Batch tests on R1 sludge samples		
Sludge on Days 460 and 461	Measurement of settling rates of solid particles in sludge		

** H_2S gas measurement done only when conducting sulphur mass balances over the system. This was done once because % S mass exiting as H_2S gas was negligible.*

TABLE 8
Effluent characteristics of UASB reactor R1 at the lowest optimal HRT of 18.0 to 18.5 h with no alkalinity dosage to feed

Steady state (SS) period 2 (R1 Day 420-473)	Influent		Effluent	
Parameter	Average value	SSD	Average value	SSD
Total organic COD (mgCOD/ℓ)	2 584	42	460	81
Organic soluble COD (mgCOD/ℓ)	337	6	229	18
Particulate COD (mgCOD/ℓ)	2 249	54	231	72
VFA (or SCFA) (mgCOD/ℓ)	158	10	72	36
Sulphate (mgSO$_4^{2-}$/ℓ)	1 800	-	146	51
Sum of H_2CO_3* alkalinity and Alk H_2S (mg/ℓ as $CaCO_3$)	22	3	1 855	73
H_2CO_3* alkalinity (mg/ℓ as $CaCO_3$)	22	3	1 358	6
TKN (mgN/ℓ)	76	7	55	7
FSA (mgN/ℓ)	9	6	46	12
pH	5.99	0.05	7.08	0.02
Aqueous sulphide (mgS/ℓ)	0	-	502	25
H_2S gas (mgS/ℓ)	0	-	1.1	0.4
Elemental sulphur in sludge (mgS/ℓ sludge)	0	-	21	2
Theoretical aqueous sulphide from sulphate reduced (mgS/ℓ)	0.0	-	539	23

experimental stages and tests conducted on R1 (fed 1 800 mgSO$_4^{2-}$/ℓ) including batch tests during the experimental period from Day 281 to Day 530 are listed in Table 7.

R1 performance (fed 1 800 mgSO$_4^{2-}$/ℓ) at the lowest optimal HRT of 18.0 to 18.5 h

Table 8 summarises the concentrations of the effluent parameters of interest when R1 (fed 1 800 mgSO$_4^{2-}$/ℓ) was operated at its lowest optimal HRT of 18.0 to 18.5 h (Day 420 to Day 473). The influent values are also listed including pH because alkalinity was not dosed to the influent during this period.

The low effluent organic COD, sulphate and VFA concentrations with the concomitant production of high alkalinity underline the very good performance of R1 even at a relatively low HRT and no alkalinity dosage to the feed. The system achieved ~92% sulphate removal efficiency implying that inhibition of un-dissociated hydrogen sulphide on the growth and activity of SRB was negligible. This aspect is important in calibrating the value of hydrogen sulphide inhibition constant (K_I) in the calibration and validation of the dynamic kinetic BSR model (UCTADM1-BSR) (to be presented in Part 6 of this series; currently in preparation by Poinapen and Ekama). Interestingly, the reactor/effluent pH was very close to the pK'_{S1} value of the H_2S/HS^- system suggesting that in the absence of gaseous CO_2 production, the system pH is governed by the sulphide system. This aspect will be explored in greater depth in the steady state model development to be presented in Part 5 (currently in preparation by Poinapen and Ekama). Importantly for model development, the aqueous hydrogen sulphide recovery matches closely the theoretical sulphide from the sulphate reduced and COD, S and N mass balances were all very good averaging 96.5 (± 0.8), 95.8 (± 2.9) and 103.3 (± 8.3) respectively.

With the excellent performance of R1 from Day 280 to Day 400 when fed 1 800 mgSO$_4^{2-}$/ℓ (detailed results not given), $NaHCO_3$ dosage to the feed was stopped on R1 Day 400. Figure 4 shows the variations in feed and effluent pH with time. Particularly striking is that the effluent pH was unaffected implying that the system pH is self-regulatory at pH >7.0 even

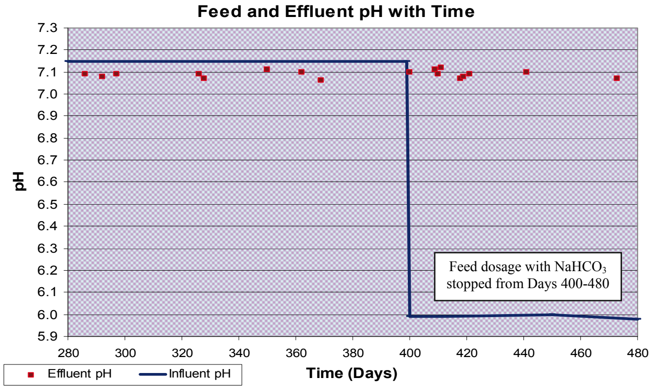

Figure 4

UASB reactor R1 influent and effluent pH-time profile with PSS as substrate augmented with 1 800 mgSO$_4^{2-}$/ℓ sulphate

at the high sulphate feed concentration of 1 800 mgSO$_4^{2-}$/ℓ due to the high alkalinity generation. Because no CO_2 gas was generated (no bubbling was observed in the $FeCl_3$ H_2S gas collection bottle, see Part 1, Poinapen et al., 2009a), it seems that all CO_2 produced by the breakdown of the PSS biodegradable organics remain in solution as HCO_3^-. Because the CO_2 (or HCO_3^-) generation may be insufficient to supply the alkalinity required by the sulphate reduction because the organics donate more electrons and protons than supply C for HCO_3^-, the sulphide system provides the alkalinity shortfall as dissociated sulphide (HS^- which is proportional to Alk H_2S; to be presented in Part 5 of Poinapen and Ekama, in preparation).

Comparison of R1 (35°C) and R2 (20°C) (both fed 1 500 mgSO$_4^{2-}$/ℓ)

Since the objective of operating the UASB Reactor R2 was to study the performance of BSR using PSS at ambient temperature (namely 20°C), a comparison between R1 and R2 both fed 1 500 mgSO$_4^{2-}$/ℓ was important because most full-scale systems are not likely to be heated to 35°C. Table 9 lists the performance of the 2 UASB reactors when both were operated at their lowest optimal HRT. The influent characteristics for both R1 and R2 feeds are also listed because this highlights the carbon requirement when operating BSR using PSS at the 2 reactor temperatures. With regard to the effluent quality, both reactors achieved successful BSR and met the set performance criteria (effluent SO$_4^{2-}$ <250 mgSO$_4^{2-}$/ℓ, VFA <100 mgHAc/ℓ) with excellent solid liquid separation (Table 9). In terms of the design and operating parameters, such as influent flow (Q_i), HRT, BRT, R_s and bed volume (V_b), Reactor R1 at 35°C outperforms R2 at 20°C because the hydrolysis rate of the PSS is strongly decreased by a temperature reduction of 15°C (~ 30% reduction in BPO hydrolysed). The feed PSS COD/SO$_4^{2-}$ ratio of R2 had to be increased to 1.75 to make up for the BPO not hydrolysed. Moreover, R2 had a longer HRT and V_b in order to increase the BRT and sludge age (R_s) thereby improving the utilisation of BPO. Economically these factors favour R1 as it has a more efficient use of BPO (~90%) than R2 (~60%). However, in practical terms, this advantage might be offset as a UASB reactor would need to be heated to 35°C to achieve the R1 performance. A cost-benefit analysis of these 2 reactors' design and operating parameters would be required in order to assess the economics of the 2 systems. The successful

TABLE 9
Comparison of R1 and R2 performance at their respective lowest optimal HRTs

Parameters	**Units**	**R1 at 35°C (1 500 mgSO$_4^{2-}$/ℓ)**		**R2 at 20°C (1 500 mgSO$_4^{2-}$/ℓ)**	
Measured parameters		**Influent**	**Effluent**	**Influent**	**Effluent**
Total COD	mgCOD/ℓ	1880	1165	2596	1141
Un-biodegradable particulate COD ($f_{PS'up}$=0.36) (UPO)	mgCOD/ℓ	677	-	935	-
Total soluble COD	mgCOD/ℓ	236	761^b	339	948
Biodegradable particulate COD (BPO)	mgCOD/ℓ	967	-	1322	-
Un-biodegradable soluble COD (USO)	mgCOD/ℓ	6	-	8	-
SCFA COD	mgCOD/ℓ	126	51	169	66
Sulphide COD	mgCOD/ℓ	0	627^b	0	854
Organic soluble COD	mgCOD/ℓ	230	134^b	331	96
Sulphate	mgSO$_4^{2-}$/ℓ	1 500	149	1500	101
Free and saline ammonia (FSA)	mgN/ℓ	10	32	10	38
pH	-	7.20^a	7.15	5.94	7.21
Sum of H_2CO_3* alkalinity and Alk H_2S	mg/ℓ as $CaCO_3$	456^a	1 938^b	23	1 558
H_2CO_3* alkalinity	mg/ℓ as $CaCO_3$	456^a	1 611^b	23	1 144
Design and operating parameters		R1 at 35°C (1 500 mgSO$_4^{2-}$/ℓ)		R2 at 20°C (1 500 mgSO$_4^{2-}$/ℓ)	
Volume of reactor bed after wastage (V_b)	ℓ	6.7		7.4	
Feed flow rate, Q_i	ℓ/d	13.6		9.2	
Hydraulic retention time (HRT)	h	13.8		20.5	
Sludge age, R_s	d	18		24	
Waste flow rate, Q_w	ℓ/d	0.37		0.32-0.34	
Upflow velocity, V_{up}	m/h	0.127		0.085	

a: *Alkalinity dosage to R1 feed.*
b: *Problem in H_2S measurement during R1 first 280 d of operation.*

TABLE 10
Performance of various bioreactors used for the biological reduction of sulphate-rich wastewaters

Reactor and process type	Substrate(s) utilised	Tempe-rature (°C)	Hydraulic retention time, HRT (h)	pH		Sulphate			Reference(s)
				Feed	Effluent	Feed ($mgSO_4^{2-}/ℓ$)	Volumetric reduction rate (g/m³/d)	Removal (%)	
UASB	PSS	35	13.5	7.2	7.2	1500	2 792	90	Poinapen et al. (2009a)
UASB	PSS	35	18.0	6.0	7.1	1800	2 423	92	Current paper
UASB	PSS	20	20.5	5.9	7.2	1500	1 737	93	Current paper
UASB	Lactate	35	16.0	3.0	7.7	1650	1 860	75	Kaksonen (2004)
UASB with a calm zone above sludge	Ethanol, nutrients (+ flocculant)	30	4.0	3.2	7.0	1480	7 010	79	Barnes et al. (1991)
FBR	Lactate	35	16.0	2.5	7.8	2290	2 220	65	Kaksonen (2004)
FBR	Ethanol	35	16.0	2.5	7.7	1920	2 320	81	Kaksonen (2004)
FBR	Ethanol	35	6.5	3.0	7.9	2080	4 290	57	Kaksonen (2004)
SBR	Molasses	31	15.0	-	7.0	2500	2 712	68	Maree and Strydom (1987)
PBR	Molasses	31	20.0	-	7.0	900	525	58	Maree and Strydom (1987)

UASB = upflow anaerobic sludge bed; FBR = fluidised bed reactor; SBR = sludge bed reactor; PBR = packed bed reactor

TABLE 11
Design and operating parameters for BSR UASB system using PSS as carbon and energy source

Design and operating parameters	Unit	Average values		
		R1 (35°C)	R1 (35°C)	R2 (20°C)
Feed PSS COD based on $f_{PS'up}$ of 0.36	mgCOD/ℓ	1880	2584	2596
Feed sulphate	$mgSO_4^{2-}/ℓ$	1500	1800	1500
Influent pH	-	7.20[a]	5.99	5.94
Influent flow rate, Q_i	ℓ/d	13.6	10.4	9.2
Organic loading rate, OLR	g/(m³·d)	3806	3785	3224
Sulphate loading rate, SLR	g/(m³·d)	3037	2637	1863
Reactor liquid volume	ℓ	7.8	7.8	7.8
Reactor bed volume	ℓ	6.7	7.1	7.4
Sludge recycle ratio (relative to Q_i)	-	0.5	0.5	0.5
Upflow velocity, V_{up}	m/h	0.125	0.096	0.085
Hydraulic retention time, HRT	h	13.8	18.0	20.4
Sludge aspect ratio (bed height/diameter)	-	10.5	11.2	11.6
Bed hydraulic retention time, BRT	h	11.9	16.4	19.3
Sludge retention time (sludge age), R_s	d	18	21	24
Waste flow rate, Q_w	ℓ/d	0.37	0.34	0.31
Expected sulphate removal rate	g/(m³·d)	2735	2423	1737
Expected sulphate reduction	%	90.1	91.9	93.3

[a]*: With alkalinity dosage to influent but this proved unnecessary because the system pH is self-regulatory*

operation of R2 demonstrates that sulphate reduction using PSS at ambient temperature (20°C) can be achieved provided a higher feed PSS COD/SO_4^{2-} ratio and/or a longer R_s is applied.

Comparison of operating parameters and performance with previous studies

A summary of the performance of the UASB reactors operated in this study along with the performance of various one-stage bioreactors for the treatment of sulphate-rich waters is listed in Table 10. Overall the use of PSS as carbon source (substrate) in UASB systems matches closely (and sometimes exceeds) the performance of systems fed with soluble substrates (namely ethanol, lactate and molasses). This demonstrates the benefits of PSS as a low cost energy source for BSR.

Design and operating parameters

A summary of the different design and operating parameters applied to the 2 BSR UASB systems of this investigation is listed in Table 11. Two of these parameters are critical and govern the system performance and economics:

- The sludge age (R_s) which controls the sludge aspect ratio (bed height/diameter) and hence the reactor bed volume. This in turn will govern the biomass concentration for BPO maximum hydrolysis rate, which is the limiting rate of the system; Ristow et al. (2006) showed that this rate is closely similar to that in methanogenic AD systems treating PSS.
- The HRT controls the reactor volume comprising the bed and liquid phases. A short HRT is always desirable because it reduces the reactor volume. However, too short an HRT

leads to high upflow velocity and disproportionate bed expansion and sludge wash out. Therefore, the optimum HRT should be established so as to allow sufficient contact time of the sulphate with the sludge bed (biomass) and to optimise the upflow velocity (V_{up}) which governs the bed expansion ratio which in turn dictates the sludge age if a constant bed height is required. Sludge settleability and bed expansion are considered by Poinapen et al. (2009c).

The remaining parameters will then be a consequence of how the sludge age (R_s) and HRT are optimised. As far as temperature is concerned, it was found that the feed PSS COD/SO_4^{2-} ratio had to be increased to 1.75 to cater for incomplete biodegradable particulate organics (BPO) hydrolysis (and consequently low VFA generation) due to the slower hydrolysis/acidogenesis rate at a temperature of 20°C compared with 35°C. If a high % of biodegradable organics utilisation is required at the lower temperature, the sludge age of the UASB reactor needs to be increased by increasing the reactor area so that a higher sludge mass is retained in the reactor in relation to that wasted to maintain the bed height, i.e. in effect reducing the volumetric COD (and SO_4^{2-}) loading rate.

Conclusions

The successful operation of the UASB Reactor R2 (fed 1 500 mgSO_4^{2-}/ℓ) at 20°C is of significant importance because full-scale systems are not likely to be heated due to the absence of methane gas. Besides the 2 critical parameters (R_s and HRT) which are both longer at 20°C than at 35°C, the other important parameter in the operation of BSR systems using PSS is the feed PSS COD/SO_4^{2-} ratio. By decreasing the temperature from 35°C to 20°C, the PSS biodegradable particulate organics (BPO) hydrolysis rate is decreased and less than 70% of the BPO is hydrolysed thereby generating less VFA for BSR. Increasing the PSS COD/SO_4^{2-} from 1.25 (applied to R1) to 1.75 (applied to R2) increased the BPO concentration hydrolysed to meet the VFA demand for sulphate reduction. Alternatively, increasing the sludge age of the system by reducing the volumetric COD and sulphate load would increase the % BPO hydrolysed while maintaining the feed PSS COD/SO_4^{2-} at 1.25.

UASB R1 was operated at an increased influent sulphate concentration (1 800 mgSO_4^{2-}/ℓ) to investigate the inhibition effect by un-dissociated hydrogen sulphide generated from the reduction of this sulphate concentration level. It was found that high sulphate reduction (~ 92%) was achieved even at the relatively low HRT of 18.5 h. This observation is important in calibrating the value of the acetotrophic (acetate using) SRB inhibition constant (K_I) in the dynamic kinetic simulation model (UCTADM1-BSR) developed in Part 6 of this series, which is currently in preparation by Poinapen and Ekama.

Acknowledgements

This research was supported financially by the Water Research Commission, the National Research Foundation and the University of Cape Town and is published with their permission.

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