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Analysis of immobilized cell bioreactors for desulfurization of flue gases and sulfite/sulfate-laden wastewater

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Abstract

Sulfur dioxide (SO_2) is one of the major pollutants in the atmosphere that cause acid rain. Microbial processes for reducing SO_2 to hydrogen sulfide (H_2S) have previously been demonstrated by utilizing mixed cultures of sulfate-reducing bacteria (SRB) with municipal sewage digest as the carbon and energy source. To maximize the productivity of the SO_2-reducing bioreactor in this study, various immobilized cell bioreactors were investigated: a stirred tank with SRB flocs and columnar reactors with cells immobilized in either κ-carrageenan gel matrix or polymeric porous BIO-SEP^{TM} beads. The maximum volumetric productivity for SO_2 reduction in the continuous stirred-tank reactor (CSTR) with SRB flocs was 2.1 mmol SO_2/h·l. The κ-carrageenan gel matrix used for cell immobilization was not durable at feed sulfite concentrations greater than 2000 mg/l or at sulfite feed rate of 1.7 mmol/h·l. A columnar reactor with mixed SRB cells that had been allowed to grow into highly stable BIO-SEP polymeric beads exhibited the highest sulfite conversion rates, in the range of 16.5 mmol/h·l (with 100% conversion) to 20 mmol/h·l (with 95% conversion). In addition to flue gas desulfurization, potential applications of this microbial process include the treatment of sulfate/sulfite-laden wastewater from the pulp and paper, petroleum, mining, and chemical industries.

Introduction

Sulfur dioxide (SO_2), the most abundant air pollutant in the United States, is one of the major constituents that cause acid rain. The quantity emitted in the United States in 1992 was estimated to be 22.73 million tons [1]. Nearly two-thirds of the SO_2 emission arises from fossil fuel combustion, mainly from coal-fired power plants. In the atmosphere, SO_2 reacts photochemically or catalytically with other constituents to form sulfuric acid. Conventional flue gas desulfurization (FGD) processes use either disposable sorbents, such as limestone, or regenerable sorbents, such as copper oxide. A microbial process of reducing SO_2 first to hydrogen sulfide and then elemental sulfur has been considered as a promising technology that can replace a part of the conventional FGD processes economically and efficiently [6].

Sulfate- and sulfite-reducing bioreactors have been proposed for a number of industrially relevant processes, including treatment of FGD waste gypsum [2–5], desulfurization of flue gas [6–9], and precipitation of metals [10, 11]. Perhaps the process that is closest to commercial realization is a new regenerable FGD process, termed BIO-FDG, that was developed by Biostar Development, a joint venture of Paques B.V. and Hoogovens Technical Services [8]. This process combines a conventional caustic scrubber for the removal of SO_2 from flue gas and two biological reactors for processing the liquid from the absorber. In the biological reactors, the sulfites and sulfates from the absorber liquid are first converted to sulfide and, subsequently, to elemental sulfur, while caustic scrubbing liquor is being regenerated. In this process, as in any potential application of SRB bioreactors, the selection of electron donor source is expected to have an impact on the economics of the process and will

be a site-specific design criterion. Lactic acid, ethanol, hydrogen, synthesis gas and sewage digest have been utilized as electron donors [2–11, 15–18]. While lactic acid and ethanol may be utilized in very specific situations where their availability is fortuitous, they are probably too expensive to be used as feedstocks for the process.

Synthesis gas, a mixture primarily of CO, H_2 and CO_2, would be an attractive alternative feedstock for the SRB process due to its wide availability, particularly at the utility plant, and its zero COD discharge. Various groups have demonstrated that SRB could be supported by carbon dioxide and/or carbon monoxide as the sole carbon source and hydrogen as the energy source [4, 12–16]. Du Preez et al. [15] operated a sulfate-reducing reactor with a mixed SRB population to demonstrate the feasibility of using synthesis-gas as the feed source for SRB. Recently, Van Houten et al. [16] reported the operation of a gas-lift sulfate-reducing reactor that was fed a CO-H_2 mixture (up to 20% CO) and yielded a maximum sulfate conversion rate of 10 g SO_4^{2-}/l·d.

Municipal sewage, which is readily available at negative or near-zero cost, has been proposed as an alternative inexpensive feedstock for SRB reactors [17]. The only real cost incurred would be that involved in transporting the sewage from the sewage plant to the flue gas treatment facility, which may vary with the particular plant and application. However, the sewage sludge would have to be chemically or biologically processed in order for the SO_2-reducing cultures to utilize the organic materials present in this feed. Selvaraj and Sublette operated a bench-scale continuous reactor with a mixed septic culture of the sulfate-reducing bacterium *Desulfovibrio desulfuricans* immobilized by coculturing with floc-forming anaerobes having SO_2 as the terminal electron acceptor [17]. Anaerobically digested-municipal sewage sludge (AD-MSS) medium with inhibition of methanogenesis by 250 ppm of chloroform was used as sole carbon and energy source for the SO_2-reducing process culture. A gravity settler served as the means for recycling the biomass. An economic analysis of this process, with AD-MSS medium as the electron donor and carbon source rather than glucose (which is expensive), revealed that the SO_2-reducing microbial process would be competitive if the total biomass concentration was 50 g/l, comprising greater than 30% of the SRB concentration [18]. A further requirement for economic viability is that the concentration of fermentable substrates in the AD-MSS medium should be as such that the chemical oxygen demand (COD) is above 2500 mg/l [18].

It is thus seen that the concentrations of biocatalyst in the reactor and of fermentable substrates in the AD-MSS medium are key parameters affecting the economic viability of a microbial SO_2-reduction process as compared with conventional hydrogenation. In an earlier study, research focused on increasing the fermentable substrates concentration of AD-MSS medium with decreased cost of medium preparation. A continuous process for producing AD-MSS medium yielded a substrate concentration of 5000 mg/l of COD [19], twice that obtained previously [17]. This increased production was achieved by using only 50 ppm of chloroform. In an effort to increase the biocatalyst concentration, immobilization of the process culture in a gel matrix was considered. Immobilization of biocatalyst with alginate and κ-carrageenan-polyethylenimine (PEI) beads was investigated to assess the durability of the beads and the effects of immobilization on biocatalytic performance [19]. Results of preliminary studies demonstrated that the alginate-cell beads, while not impairing the biocatalytic performance as compared with cells in free culture, disintegrated even at a feed sulfite throughput as low as 0.19 mmol/h·l. This caused the elutriation of biocatalyst, resulting in a low SO_2 conversion rate. On the other hand, κ-carrageenan-PEI cell beads were more stable and more active than the alginate beads under the same operating conditions. Therefore, it was suggested that the operation of a columnar reactor with higher biomass concentration in the κ-carrageenan-PEI beads would increase the volumetric productivity of the reactor.

In the present study, our objective of the research was to obtain maximum reactor productivity with increased biocatalyst concentration and simple reactor configuration to make the biodesulfurization process economically viable. First, a columnar reactor with κ-carrageenan-PEI cell beads was operated to study the integrity and productivity of the beads. Subsequently, polymeric porous beads were considered as an attractive alternative immobilization medium because of their durability, large internal voids, and extensive microporous surface area (which would augment mass transport). The polymeric bead's high content of activated carbon (i.e., of 50–80%) protects the biocatalyst from impetuous high toxic effects of the pollutants. A columnar reactor with SRB cells immobilized in the polymeric porous beads was operated to study its productivity and its utilization of AD-MSS medium as compared with the κ-carrageenan cell reactor and

a CSTR with SRB flocs. Furthermore, this work has been focused on developing a generic technology that could be implemented to treat any sulfite/sulfate-laden waste from various applications such as pulp and paper, mining, and chemical industries, in addition to FGD.

Materials and methods

Organisms and culture

Mixed SRB cultures were isolated from sewage solids obtained from the diffused air flotation (DAF) unit of a municipal sewage treatment plant at Oak Ridge, Tennessee. A serum bottle containing 100 ml of AD-MSS medium [19] and 0.15 g of sodium sulfate was incubated at 30 °C. The indigenous SRB in the sewage medium utilized the added sulfate as the terminal electron acceptor and reduced it to H_2S. The lower-chain fatty acids in the medium served as the electron donor. The stock culture of SRB was maintained by transferring this positive culture into the AD-MSS medium (containing added sodium sulfate) on a weekly basis.

Biomass growth for use in reactor trials

A 1-L Omni-Culture fermenter (Virtis Co., Gardiner, NY) with temperature and agitation controls was used to grow working cultures of mixed SRB. A pH controller (Chemcadet, Cole-Parmer, Niles, IL) using 6 *N* phosphoric acid and 6 *N* sodium hydroxide was used to maintain a constant pH of 7.0. Prior to growth of the culture on SO_2, chemostat operation was initiated with 1 l of AD-MSS medium with sulfate added (1.5 g/l of Na_2SO_4) as the electron acceptor. A 5-ml sample of the mixed SRB stock culture developed as described previously was used to inoculate this reactor, which was operated at an agitation rate of 200 rpm and a temperature of 30 °C. Nitrogen was purged at a flow rate of 300 ml/min in order to scrub the produced H_2S from the system. When the H_2S concentration in the off-gas reached about 1000 ppm, the biomass was harvested by centrifugation and resuspended in fresh AD-MSS medium (with no sulfate added). At this time, 1% SO_2, 5% CO_2, balance N_2 gas mixture (Matheson Gas Products, Marrow, GA) was fed to the reactor at a rate of 7.6 ml/min. This corresponded to a molar SO_2 feed rate of 0.19 mmol/h. The culture was then operated in a continuous mode with the AD-MSS medium at a feed rate of 0.4 ml/min. The reactor effluent was collected under a nitrogen blanket and used as a biomass source for additional experiments. A 10-l glass-bottle batch reactor was also operated in a similar fashion (as described above) to obtain a large amouont of biomass needed for producing κ-carrageenan-PEI encapsulated beads.

SRB flocs

Mixed SRB flocs were developed by co-culturing the mixed SRB free cells with anaerobic flocs obtained from an anaerobic digester of a sewage treatment plant [17]. A 2-l Omni-Culture fermenter (Virtis Co., Gardiner, NY) was operated as a CSTR utilizing the SRB flocs with AD-MSS medium as the electron donor. A 13-cm-long × 3.5 cm-diam plastic settling tube was attached to the effluent tube in the chemostat to retain the immobilized cells in the reactor. This enabled the reactor to operate with a higher cell density than a free-cell reactor. The temperature, pH, and agitation rate of the reactor were maintained at 30 °C, 7.0 and 150 rpm, respectively. Initially the reactor was fed with 1% SO_2, 5% CO_2, and balance N_2 gas mixture at a rate of 63.5 ml/min (1.6 mmol/h of SO_2) and with AD-MSS medium at a rate of 12 ml/h. However, as the concentration of the cells in the reactor increased, the SO_2 feed rate was also gradually increased. During this period, the flow rate of the AD-MSS medium was augmented to meet the requirements of the carbon and energy sources for the process culture. The reactor was monitored routinely for sulfite, organic acids, and mixed liquor suspended solids (MLSS). The AD-MSS feed and the reactor effluent samples were analyzed for both COD and organic acids.

κ-Carrageenan beads

In experiments using carrageenan beads, the immobilized biocatalyst was prepared using 4% κ-carrageenan (Type NJAL-798, FMC Corp., Chicago, IL) with 0.5% polyethylenimine (50% PEI solution, Sigma Chemical Co., St. Louis, MO) in an aqueous solution at 40 °C [20]. A 10-l SO_2 reactor was operated, as described above, at an SO_2 flow rate of 2.5 mmol/h. During the log phase of their growth, the cells were collected anaerobically using a N_2 blanket by centrifugation. The beads were prepared from 300 ml of κ-carrageenan-PEI solution with 75 g (wet wt.) of cell paste and pumped through a syringe needle (22 gauge) as droplets. The κ-carrageenan-PEI solution with cell paste was maintained at 40 °C during the period of bead preparation to prevent solidification of the gel pri-

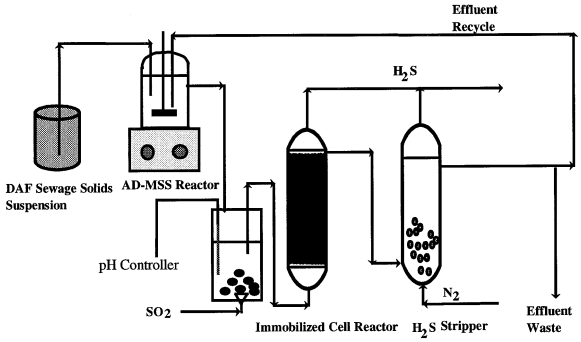

Figure 1. Schematic diagram of an SO_2-reducing columnar reactor with a mixed SRB culture immobilized in BIO-SEP^{TM} polymeric porous beads. The immobilized mixed culture utilizes anaerobically digested municipal sewage solids (AD-MSS) medium as the carbon and energy source. The feed sulfite solution was prepared by sparging SO_2 gas into AD-MSS medium.

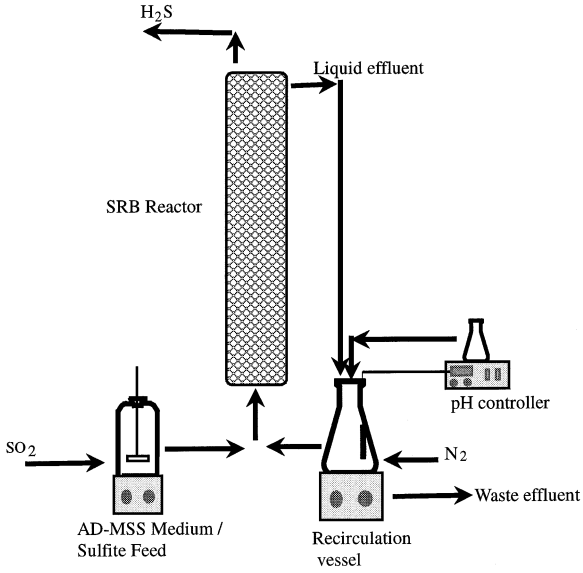

Figure 2. Immobilized cell recycle reactor. To maintain the pH within this reactor, the total liquid flow rate through the reactor was increased to 60 ml/h and the process effluent was adjusted to pH 7.0 before being recirculated to the reactor. The feed rate of the SO_2-AD-MSS solution was the same as the rate of the liquid discharge from the recirculation vessel in order to maintain a constant liquid volume. The residence time of the sulfite feed solution in the reactor was calculated as the liquid volume of the reactor divided by the fresh SO_2-AD-MSS solution feed rate.

or to pumping through the needle. The droplets were fixed as beads (2–3 mm) in cold 0.3 *M* KCl solution at 10–15 °C. The immobilized κ-carrageenan cell beads were mixed in the KCl solution for about 2 h for complete cross-linking. A fully jacketed glass columnar reactor (5821-24, Ace Glass, Vineland, NJ) of internal dimensions 2.5 × 30 cm was used as a fixed-bed reactor with the immobilized cell beads. The resulting fixed bed reactor had a total volume of 181 ml and a liquid volume in the active portion of the reactor of 81 ml. The feedstock for the reactor was prepared by dissolving SO_2 gas in AD-MSS medium with the pH maintained at 6.8–6.9 using 6 *N* NaOH. The reactor was fed with the above solution (393 mg/l sulfite, 0.32 mmol/h·l) at a rate of 12 ml/h. The feed and effluent samples were routinely analyzed for sulfite, COD, and organic acids. The sulfite concentration in the feed was increased gradually when complete sulfite conversion was demonstrated by the low levels (< 5 mg/l) of sulfite in the effluent. A similar experiment with carrageenan beads without any cells was conducted for 7 d as a control run to investigate possible abiotic removal of sulfite.

BIO-SEP^{TM} beads

BIO-SEP^{TM} beads, encapsulated activated carbon (50–80%) in aromatic polyamide (Aramid) (20–50%) were obtained as the kind gift of Dr. Carl Camp from the DuPont Chemical Co. (Glasgow, DE). These beads were expected to have superior performance relative to gelatin beads due to their high toxic resistance, high durability, and large internal voids in which microbes can grow well protected from hydraulic shear.

BIO-SEP beads (2.5–3.5 mm) were packed in a similar, jacketed glass columnar reactor as described above (Figure 1). Initially, an abiotic control experiment was conducted with beads, but without cells, for 5 d at a sulfite feed rate of 0.5 mmol/h·l. To prepare immobilized biocatalyst, the beads in the reactor were initially soaked with 182 ml of AD-MSS medium with 0.4 g of sodium sulfate and 5 ml of a mixed SRB inoculum developed from the 1-l chemostat. After incubation for 5 d at 30 °C, the reactor medium was recirculated in the columnar reactor at a rate of 12 ml/h for an additional 5 days to allow the cells to distribute within the beads. The columnar reactor was then fed with fresh SO_2 solution at a rate of 12 ml/h, equivalent to a liquid residence time of 6.75 h. The liquid residence time was calculated based on the reactor volume minus bead volume devided by feed influent rate. The SO_2 solution was prepared by purging with a SO_2 gas mixture (5% SO_2, 5% CO_2, and balance N_2) at a rate of 10 ml/min in AD-MSS medium. The pH of the feed SO_2-AD-MSS solution was maintained at

6.80–6.90 using 6 *N* NaOH solution. The effluent from the reactor was pumped into a stripping column where it was purged with nitrogen to strip off H_2S. During the period of operation, the pH of the column effluent increased gradually to about 9.0 as the concentration of the sulfite in the column feed was increased above 7000 mg/l. This caused an upset in the reactor, resulting in an incomplete sulfite conversion. In order to maintain the pH of the reactor medium at 7.0 throughout the column, a recycling facility was added to the operation in place of stripping column as shown in Figure 2. With this arrangement, the column effluent was collected in a 250-ml glass bottle, its pH adjusted to 7.0 using 6 *N* phosphoric acid, purged continuously with N_2 to strip off H_2S, and recycled to the column at the rate of 48 ml/h. This procedure increased the total flow in the column to 60 ml/h. The effluent from the recycle bottle was discarded at a rate of 12 ml/h to equilize the SO_2 solution feed rate to the column. The recycle ratio (total liquid flow to the reactor divided by SO_2-AD-MSS solution flow rate) of the reactor was 5. At steady-state conditions, the column was fed with 10,000 mg/l of sulfite which was equivalent to 8.1 mmol/h·l. The column feed solution and the effluent were sampled daily to analyze sulfite, COD, organic acids, and pH. The recycle ratio was then altered to 2, maintaining a constant flow rate of feed at 12 ml/h to maximize the SO_2 productivity of the reactor. The inlet sulfite concentration was gradually increased to about 20,000 mg/l (16.4 mmol/h·l) and operated at this concentration for 50 d. Subsequently, the feed sulfite concentration was increased to about 25,000 mg/l (20 mmol/h·l) and operated for an additional 20 d.

At this time, portions of the biocatalyst beads were removed from the top and the bottom of the reactors for scanning electron microscopic (SEM) examination in order to study immobilization of the cells in teh pores of the beads. The beads were packaged under anaerobic conditions and sent to Conoco Co. (Punca City, OK) for SEM analysis.

Analytical methods

Hydrogen sulfide in the off-gas was analyzed using a gas chromatograph (Hewlett Packard HP 5890 Series II) equipped with a Teflon column (3 ft × 1/8 in.) packed with SuperQ (80 to 100 mesh) and a thermal conductivity detector. Temperatures of the column, injection port, and thermal conductivity detector were 50, 125, and 125 °C, respectively. Helium was used as the carrier gas at a flow rate of 25 ml/min. Sulfite was analyzed spectrophotometrically by the reaction of fuchsin and formaldehyde in sulfuric acid [21]. COD values were determined using Hach Chemical Co. (Loveland, CO) premeasured reagent vials. Organic acids were quantified by gas chromatograph (Hewlett Packard HP 5890 Series II) equipped with flame ionization detector. A 30-m HP–INNOWax column (19091N-133, Hewlett Packard) was used with a helium carrier flow of 1.5 ml/min through a ramped temperature profile from 120 °C (1 min) to 240 °C (1 min) at 10 °C per min. The injector and detector temperatures were at 250 °C, and 300 °C, respectively. For SEM analysis, the samples were prepared by slicing randomly selected beads with a razor, allowing them to dry in air, affixing them to stubs with carbon adhesive, and sputter-coating them with 12 nm of gold. They were then examined in a JEOL JSM-840A SEM at a working distance of 20 mm and an accelerating voltage of 20 kV.

Results and discussion

SRB flocs

Following the immobilization of the mixed SRB cultures by coculturing with anaerobic flocs, the reactor was operated in a continuous mode with an initial SO_2 feed rate of 0.1 mmol/(h·l) and an AD-MSS medium flow rate of 12 ml/h. The SO_2 feed rate was then gradually increased as the reactor medium showed no sulfite accumulation (Figure 3). Complete conversion of SO_2 was achieved at feed rates as high as 2.1 mmol SO_2/h·l. At this time, the AD-MSS medium flow rate was 15 ml/h·l. The feed and effluent CODs were 3500 and 1500 mg/l, respectively. The total organic acid concentrations in the feed and effluent were 1955 mg/l and 5 mg/l, respectively. The difference between the COD concentration and the total organic acid concentrations may be due to some mol-wt organic acids, which contribute to COD, but are not detected by our gas chromatograph technique [19]. Therefore, the stoichiometry of SO_2 conversion with respect to electron donor (AD-MSS medium) was 14.3 mg of COD per mmol of SO_2. The mixed liquor suspended solids (MLSS) of the reactor medium showed the biomass concentration to be 5000 mg/l. The average MLSS concentration in the effluent was 750 mg/l, which indicates that about 85% cell retention was obtained using the settling tube in the reactor. Use of the settling tube would eliminate the requirement of an additional gravity settler as pro-

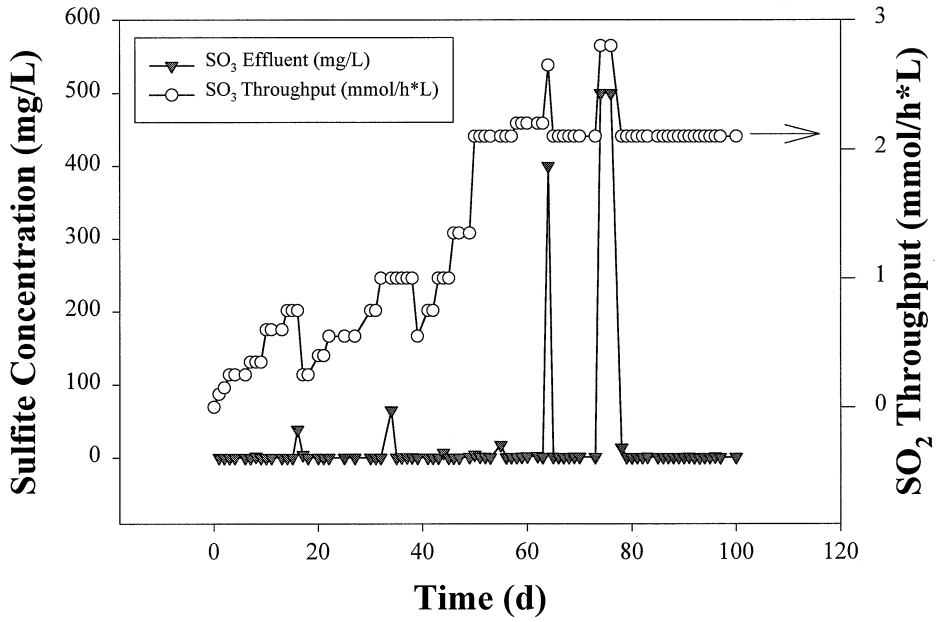

Figure 3. Sulfur dioxide feed rate and sulfite concentration during continuous operation of immobilized, mixed SRB cultures in a mixed reactor.

posed by Selvaraj and Sublette [17] and, thus, would reduce the capital and operating costs of the process. At a higher SO_2 feed rate, 2.8 mmol/h·l, very little SO_2 conversion (about 10%) was achieved as evidenced by sulfite accumulation in the reactor medium. However, the reactor recovered from the upset immediately by reducing the SO_2 feed rate to 2.1 mmol/h·l. Complete conversion of SO_2 was achieved at this feed rate.

κ-Carrageenan-PEI beads

The columnar reactor with κ-carrageenan-PEI cell beads was initiated with a sulfite feed of 0.19 mmol/h·l. The feed sulfite concentration was then increased gradually as no sulfite was detected in the column effluent (see Figure 4). Complete removal of sulfite was obtained at an inlet sulfite concentration of 2000 mg/l (1.7 mmol/h·l). During this time, the COD values for the feed and effluent were 3115 and 2450 mg/l, respectively. This indicates that utilization of the AD-MSS medium was 26.6 mg of COD per mmol of sulfite reduced. The total organic acid concentrations of the AD-MSS medium (the sum of all the organic acids detected in our GC technique) in the feed and effluent (1850 mg/l and 1220 mg/l, respectively) also indicated same utilization rate. However, when the feed sulfite concentration was increased above 1.7 mmol/h·l, disintegration of the cell beads in the column was observed as the bead height in the column was gradually reduced. As a result, the column effluent showed increasing concentrations of sulfite as shown in Figure 4, due to the loss of biomass along with the beads. The κ-carrageenan gel beads were not strong enough to be used in this type of operation at feed sulfite concentrations above 2000 mg/l (1.7 mmol/h·l). The abiotic control experiment revealed no sulfite reduction in the effluent, indicating that the sulfite reduction resulted solely from the biocatalyst in the beads.

BIO-SEP^{TM} beads

The sulfite concentrations of the feed and the effluent during the operation of the BIO-SEP reactor are shown in Figure 5. The sulfite concentration was gradually increased to 7000 mg/l (5.8 mmol/h·l) as very low concentrations (< 5 mg/l) of sulfite were detected in the column effluent. At inlet levels above 7000 mg/l, there was a gradual increase in the sulfite concentration in the effluent. At the same time, an increase in effluent pH was also observed. This increase in pH was probably due to the formation of hydroxyl ion in the reactor medium as a result of the reduction of sodium sulfite to H_2S and stripping of H_2S from aqueous phase, and oxidation of organic acids in the AD-MSS medium. Adjustment of the pH to 6.9 required 29.3 g of NaOH for 1 g-mol of sulfite feed. In order to main-

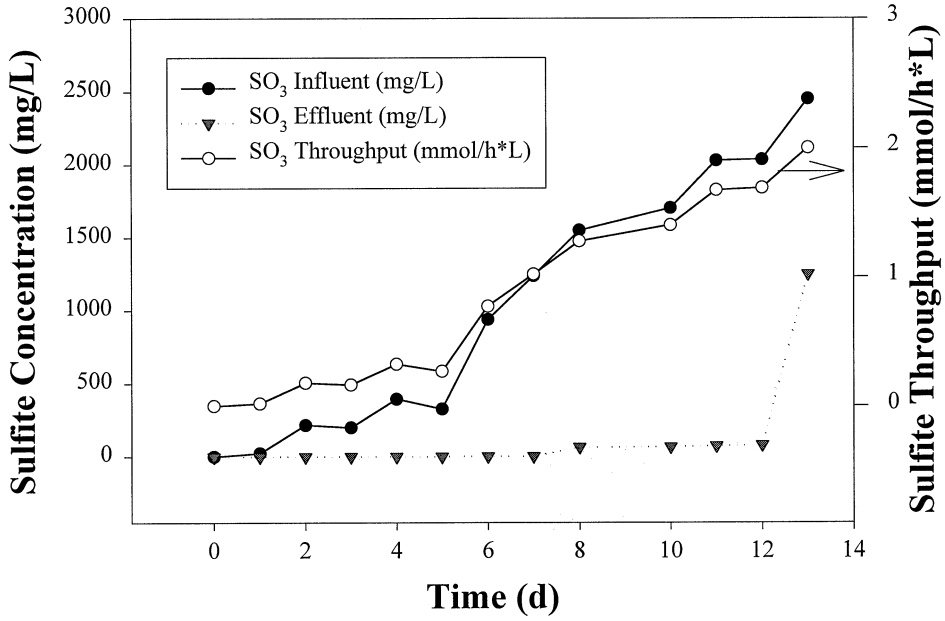

Figure 4. Sulfite conversion in a columnar reactor with cells immobilized in κ-carrageenan-PEI gelatin beads.

tain the constant pH at 7.0 in the column, an additional effluent recycling facility was installed to the columnar reactor as described above. With the recycling arrangement, the pH levels of the feed and the effluent were maintained at 6.9–7.0 using 6 *N* H_3PO_4. However, it is theoretically possible to recycle the NaOH formed in the column to adjust the sulfite feed. This would allow the process to be operated with no additional NaOH and H_3PO_4. Hoogovens Technical Services have demonstrated the regeneration of caustic scrubbing liquid after converting sulfite to sulfide and then to elemental sulfur in their BIO-FGD commercial process [8].

With this recycling arrangement, the sulfite feed was then gradually increased to 20,000 mg/l (16.5 mmol/h·l). Complete conversion of sulfite was achieved, as shown in Figure 5. When the sulfite throughput was increased above 16.5 mmol/h·l, the sulfite concentration in the effluent showed a gradual increase, which might be due to the inhibitory effect of H_2S on growth and activity of the mixed SRB. Reis et al. (1992) have shown that an H_2S concentration of 547 mg/l (16.1 mM) completely inhibits culture growth [22]. However, in our study, this inhibitory effect was found to be reversible when the sulfite feed rate was returned to 16.5 mmol/h·l. The reactor was operated in this mode for 50 d to monitor long-term operation. During this period, the feed and effluent COD concentrations of 4800 mg/l (2400 mg/l of total organic acids) and 3000 mg/l (575 mg/l of total organic acids), respectively, demonstrated that the stoichiometry with respect to electron donor was 7.25 mg of COD per mmol of sulfite reduced. The exact mechanism for the decreased carbon requirements by the biocatalyst immobilized in BIO-SEP beads is not completely understood. The MLSS of the BIO-SEP column effluent revealed a very low concentration of suspended solids (500–1000 mg/l) due to cell encapsulation in the beads and limited cell growth under immobilized conditions. This decreased suspended solids in the liquid effluent relative to other bioreactors would minimize capital and operational costs associated with excess biomass disposal. The control experiment with raw BIO-SEP beads (without cells) showed no reduction in the sulfite concentrations, suggesting that the sulfite reduction was due only to the biocatalyst in the BIO-SEP beads.

The SEM analysis of the beads (as shown in Figure 6) indicated immobilization of the biocatalyst was primarily in the pores of the beads rather than in the matrices. The major portions of pores were covered by cells with cocci-type morphology, along with some crystalline inorganic forms that might have precipitated from the sewage digest medium. However, the incomplete pore surface coverage suggests that further

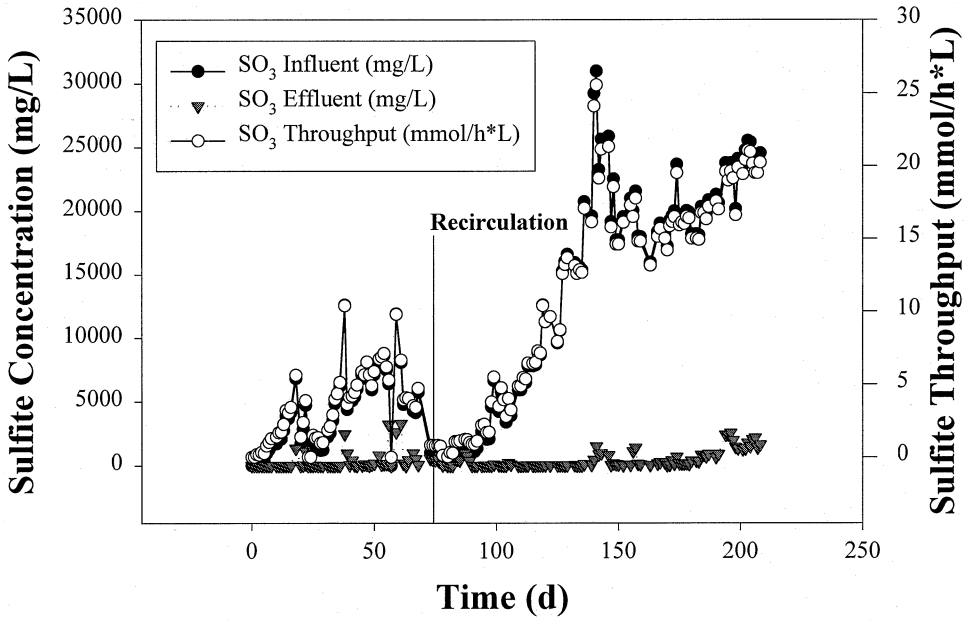

Figure 5. Sulfite conversion in a columnar reactor with cells immobilized in BIO-SEP^{TM} polymeric porous beads. By maintaining a constant pH of 7.0 within the reactor with recirculation arrangement, the productivity of the reactor is increased to 16.5 mmol/h·l with 100% conversion and 20 mmol/h·l with 95% conversion.

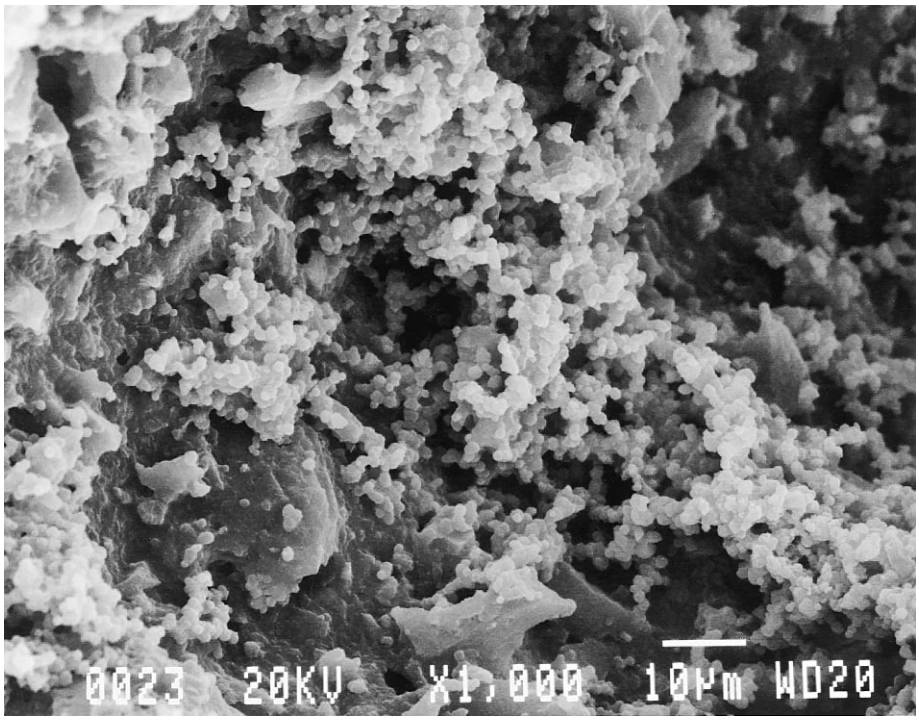

Figure 6. Scanning electron micrographs of mixed SRB colonies in the pores of a BIO-SEP^{TM} bead.

Table 1. Summary of results of various immobilized cell bioreactor studies

Reactor type	SO_2/SO_3^{2-} throughput (mmol/h·l)	Liquid residence time (h)/recycle ratio[a]	% Conversion
CSTR with SRB flocs	2.1	66.6/na	100
κ-Carrageenan-PEI cell beads	1.7	6.8/na	100
BIO-SEP beads (Dupont)	16.5	3.4/2	100
	20.0	3.4/2	95

[a] Residence time = working liquid volume of the reactor/sulfite feed influent rate. Recycle ratio = total liquid flow/sulfite influent rate.

increases in biocatalyst density, and hence bioreactor productivity, may be achieved.

Table 1 summarizes the maximal volumetric productivities achieved in the various reactor systems. Of the reactors investigated, the columnar type operated using biocatalist immobilized in BIO-SEP beads exhibited the highest SO_2 throughput. Sulfur dioxide conversion rates in this reactor were 16.5 mmol/h·l (with 100% conversion) and 20 mmol/h·l (with 95% conversion). This compares to others productivities of 16.3 mmol SO_2/h·l [23] with molasses as feed stock, 0.7 mmol SO_4^{2-}/h·l [15], and 13.0 mmol SO_4^{2-}/h·l [24] with CO_2 and H_2 as carbon and energy source, 4.3 mmol SO_4^{2-}/h·l [16] and 0.5 mmol SO_4^{2-}/h·l [25] with synthesis gas (CO and H_2) as feedstock. The highest productivities of 16.5 and 20 mmol/h·l in our work with sweage digest medium as feedstock were accomplished with less carbon utilization (7.25 mg of COD per mmol of sulfite) in the BIO-SEP reactor than in other bioreactors we studied.

Based upon reducing equivalents, SRB should require 48 mg of COD to reduce one mmol of sulfite to hydrogen sulfide. In this study as well as that reported by others [17], less COD (7.25 mg/mmol sulfite and 15 mg/mmol sulfite, respectively) was utilized. In our study, only influent and effluent sulfite concentrations were measured and it could be possible that there was also a partial reduction of sulfite resulting in thiosulfate intermediate. Thiosulfate itself contributes to measured COD at 65 mg COD/mmol. The reduction of sulfite to thiosulfate requires only 16 mg COD/mmol SO_3^{2-}. Thus, when the reactor was removing 16.5 mmol SO_3^{2-}/h·l, if the SRB were only reducing the sulfite to thiosulfate, 264 mg of COD/h would be utilized in the biological reduction and 539 mg of COD would be added to the system from the thiosulfate, resulting in a net increase in COD. It is thus seen that some incomplete conversion of the sulfite could account for the apparent underutilization of COD by the bacterial culture in these experiments. In subsequent experiments performed in our laboratory using synthesis gas as a carbon and energy source for sulfite reductions, we have been able to complete the mass balance on sulfur species. 100% of the sulfur in the influent sulfite was accounted for as H_2S in the liquid and gaseous effluent. In these studies, the SRB utilized the theoretically required amount of reducing equivalent from the synthesis gas mixture.

Conclusion

Immobilization of process culture in the biological reduction of sulfate and sulfite waste streams can have tremendous impact on the economic performance of a bioprocess due to the increased reactor productivity, decreased carbon utilization, and decreased biomass disposal costs afforded by such reactor systems. We have demonstrated that a mixed culture of sulfate-reducing bacteria, immobilized in a microporous, polymeric support and fed sewage digest medium, exhibited volumetric productivities 8-fold higher than gelatin immobilization support systems and 65-fold higher than free cell reactor systems [19]. A volumetric productivity of 16.5 mmol/h·l was achieved with 100% sulfite conversion. Sewage digest represents an inexpensive carbon and energy source, and the sewage solids requirement demonstrated in this study would be well within the range available at water treatment plants. Other carbon and energy sources, such as coal synthesis gas or ethanol, may be utilized in bioreactor systems dependent on their site availability. While this study investigated the biological reduction of SO_2 for flue gas applications, the reactor design and implications are directly applicable to a variety of sulfate and sulfite treatment applications, including the recycling

of FGD gypsum [3, 4], biogenic sulfide production for metals recovery in the mining industry [10, 11], and sulfur production from paper-mill waste streams and chemical industries.

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