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Dolomite limits acidification of a biofilter degrading dimethyl sulphide

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Abstract

The applicability of dolomite particles to control acidification in a *Hyphomicrobium* MS3 inoculated biofilter removing dimethyl sulphide (Me_2S) was studied. While direct inoculation of the dolomite particles with the liquid microbial culture was not successful, start-up of Me_2S-degradation in the biofilter was observed when the dolomite particles were mixed with 33% (wt/wt) of *Hyphomicrobium* MS3-inoculated compost or wood bark material. Under optimal conditions, an elimination capacity (EC) of 1680 g Me_2S m^{-3} d^{-1} was obtained for the compost/dolomite biofilter. Contrary to a wood bark or compost biofilter, no reduction in activity due to acidification was observed in these biofilters over a 235 day period because of the micro environment neutralisation of the microbial metabolite H_2SO_4 with the carbonate in the dolomite material. However, performance of the biofilter decreased when the moisture content of the mixed compost/dolomite material dropped below 15%. Next to this, nutrient limitation resulted in a gradual decrease of the EC and supplementation of a nitrogen source was a prerequisite to obtain a long-term high EC ($>$ 250 g Me_2S m^{-3} d^{-1}) for Me_2S. In relation to this nitrogen supplementation, it was observed that stable ECs for Me_2S were obtained when this nutrient was dosed to the biofilter at a Me_2S-C/NH_4Cl-N ratio of about 10.

Abbreviations: DW – dry weight, EC – elimination capacity, Me_2S – dimethyl sulphide, OL – organic loading rate, VS - volatile solids

Introduction

In a traditional biofilter, organic materials as e.g., compost and wood bark are used as the colonising surface for micro-organisms. Microbial inoculation of biofilters is rarely needed or advantageous because the ambiant microbiota rapidly colonises and adapts to the pollutants. In a previous report (Smet et al. 1996a), it was shown that the biofiltration of volatile organic sulphur compounds as e.g., Me_2S is an apparent and interesting exception since inoculation with *Hyphomicrobium* MS3 was necessary to obtain a high elimination capacity (e.g., 680 g Me_2S m^{-3} d^{-1} in an inoculated compost biofilter). However, due to accumulation of the metabolite sulphuric acid and the limited buffer capacity of organic substrata, long-term stability of wood bark and compost biofilters was shown to be poor ($<$ 30 days), since the activity of the culture Hyphomicrobium MS3 is strongly reduced at a pH below 5 (Smet et al. 1996a). Upon mixing small amounts of limestone ($CaCO_3$) particles in the compost material (5–24 kg m^{-3}), the long-term stability of the biofilter strongly increased, because of the stoichiometric neutralisation reaction between the $CaCO_3$ added and the metabolite H_2SO_4. No sulphate toxicity was observed in these limestone supplemented biofilters, since the toxic sulphate concentration for the culture *Hyphomicrobium* MS3 is 17 times higher than the maximum solubility of $CaSO_4$ (Smet et al. 1996b). For full-scale applications, however, mixing the colonising surface with this neutralising agent at regular time periods is not a very practical way to control the acidification.

Recently, Bonnin et al. (1994) obtained very high (1440–1680 g m^{-3} d^{-1}) ECs for the inorganic volatile compounds H_2S and NH_3 in a biofilter using a pure carbonate material (Mearl) as the substratum and operating at a surface loading rate of 500 m^3 m^{-2} h^{-1}. This sea mineral, consisting for more than 82% of $CaCO_3$, acted as a colonising surface, pH-neutralising agent and a source of inorganic carbon (carbonate, carbon dioxide), nitrogen and trace elements for the autotrophic H_2S- and NH_3-degrading microorganisms. The authors trickled a solution through the biofilter to provide moisture and additional nutrients and to wash out metabolites.

In this paper, the inorganic material dolomite was evaluated as a substratum and a tool to control acidification in *Hyphomicrobium* MS3-inoculated biofilters degrading Me_2S. Following pH adjustment, moisture and nutrient requirements in these biofilters were examined. With regard to the latter, nitrogen was singled out for study since it is an essential nutrient for microbial growth.

Materials and methods

Media

Mineral medium contained (in g L^{-1}): K_2HPO_4: 3.00; KH_2PO_4: 3.00; NH_4Cl: 3.00; $MgSO_4.7H_2O$: 0.50; $FeSO_4.7H_2O$: 0.01; final pH 7.0. For the supplementation of nitrogen as a single nutrient, NH_4Cl was used in different concentrations (0.30, 3.00, 30.00 g L^{-1}).

Reactor set-up

Three types of reactor were tested, containing pure dolomite, a dolomite/compost mixture (33% wt/wt) and a dolomite/wood bark mixture (33% wt/wt) as the substratum. Each reactor consisted of a Plexiglass column with an internal diameter of 0.046 m and an overall height of 0.5 m (Figure 1). The substratum was supported by a perforated Plexiglass plate and was filled up to a height of 0.30 m (reactor volume 0.5 $\times$ 10^{-3} m^3). Gas sampling points, fitted with Teflon-lined Mininert septa (Alltech Assoc.), were provided in the influent and effluent of the reactor. The air was humidified in a scrubber before entering the biofilter. The air flow rate was 1 L min^{-1}, corresponding to an apparent gas residence time in the biofilter of 30 s. Me_2S-concentrations up to 1340 ppmv were prepared using a dynamic diffusion system (Schoene and Steinhanses 1989). Tap water and nutrient solutions were

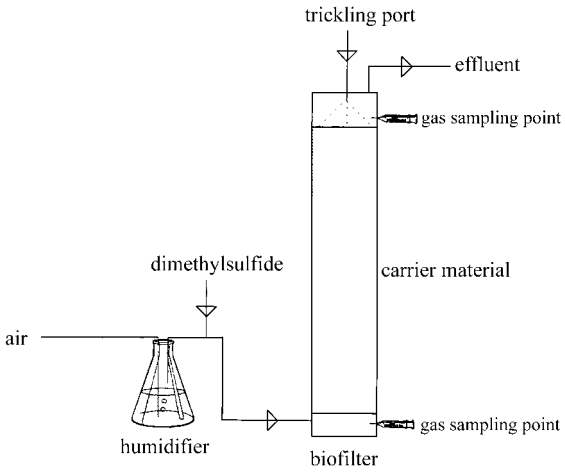

Figure 1. Reactor set-up of the lab-scale biofilter.

injected in periodic doses (50 mL) via the trickling port on top of the biofilter.

Carrier materials

The dolomite particles used consisted of (%wt): $CaCO_3$: 53.5–54.0; $MgCO_3$: 41.5–43.8; SiO_2: 0.5–1.2; Fe_2O_3: 0.3–0.7; Al_2O_3: 0.2–0.5. For the biofilter experiments, the dolomite sieve fraction between 1.40 and 4.76 mm was used. The compost used was produced from source separated municipal organic waste (the garden, fruit and vegetable fraction) by the so-called double process, i.e., the waste was treated in an anaerobic thermophilic digestor and subsequently subjected to an aerobic treatment (Gellens et al. 1995). The wood bark was obtained from the paper factory 'Stora Feldmuehle Langerbrugge nv' and contained mainly ground *Picea* tree bark. The nitrogen content of the compost and wood bark material used is 13.7 and 4.5 g kg^{-1} DW, respectively, while the C/N ratio of these materials is 15 and 119, respectively. Other characteristics of these materials were given in a previous report (Smet et al. 1996a).

Inoculum

Hyphomicrobium MS3 was enriched from soil samples. The enrichment procedure for this culture was described in a previous report (Smet et al. 1996a). The culture is degrading Me_2S according to:

$$CH_3SCH_3 + 5\ O_2 \rightarrow H_2SO_4 + 2\ CO_2 + H_2O$$

Inoculation of the biofilters was performed in batch mode by mixing the dolomite (dolomite biofilter), the

compost (dolomite/compost biofilter) and the wood bark (dolomite/wood bark biofilter) with 40 mL of a 1.5 g VS L^{-1} *Hyphomicrobium* MS3 culture (corresponding to an inoculum size of 120 g VS m^{-3} reactor) during a 10 minute period.

Analytical methods

Analysis of Me_2S was carried out using a Varian 3700 gas chromatograph, equipped with a FID. A 25 m DB1 bounded phase column (100% dimethyl polysiloxane; internal diameter 0.53 mm; film thickness 1.5:m) with He as a carrier gas was used at 30 °C. Using a Hamilton 1700 series syringe (Alltech Assoc.), 500 μl gas samples were injected. For the moisture content determination, a homogeneous sample of the substratum was taken by emptying the biofilter and mixing the material. The moisture content was calculated as the weight difference before and after drying ± 10 g material at 105 °C to constant weight (generally 24 hours). These samples were re-used as substratum after drying.

Results

Inoculation of the dolomite/compost biofilter

As was observed for the wood bark and compost biofilter (Smet et al. 1996a), very low (< 10 Me_2S m^{-3} d^{-1}) Me_2S-degradation was obtained in the pure dolomite biofilter when no inoculation was applied. However, also multiple inoculation (± 120 g VS m^{-3} reactor on day 1 and 13) of the pure dolomite biofilter with the liquid culture *Hyphomicrobium* MS3 was not successful (EC < 10 g Me_2S m^{-3} d^{-1} over a 17-day period tested) (data not shown). Upon mixing dolomite with *Hyphomicrobium* MS3-inoculated compost material, however, ECs up to 1680 g Me_2S m^{-3} d^{-1} were obtained after a 4-week period, corresponding to a removal efficiency of 85% at an organic loading rate (OL) of 1980 g m^{-3} d^{-1} (Figure 2). Due to practical problems with the Me_2S dosing system, the Me_2S-concentration in the influent of the biofilter was increased from 268 to 1340 ppmv (OL > 10000 g Me_2S m^{-3} d^{-1}) from day 32 to day 37. During this period, the EC dropped sharply from 1680 down to 200 g m^{-3} d^{-1}. As a possible explanation for this, both Me_2S-toxicity towards the inoculum *Hyphomicrobium* MS3 (Smet et al. 1996a) and the gradual decrease in moisture content of the carrier material can be stated. Indeed, the moisture content of the carrier material dropped from 19% (day 0) to 12% (day 40) during this experiment (Figure 2), suggesting that the humidification system performed insufficiently.

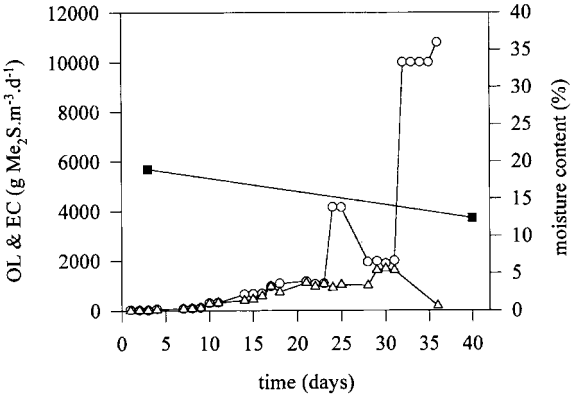

Figure 2. Organic loading rate (OL) (○) and elimination capacity (EC) (△) (g Me_2S m^{-3} d^{-1}) of the *Hyphomicrobium* MS3-inoculated dolomite/compost biofilter. The moisture content of the carrier material is represented as (■).

Effect of moisture content of the dolomite/compost material

From day 43 on, the organic loading rate was maintained at 2200–3500 g Me_2S m^{-3} d^{-1}. When 50 mL of tap water was trickled through the biofilter on days 45, 52 and 57, the moisture content of the carrier material increased from 12 up to 17%, 23% and 26%, respectively (Figure 3). No percolate was collected at the bottom of the filter during these experiments. A strong increase in EC was observed after day 45 (EC increased from 240 up to 750 g Me_2S m^{-3} d^{-1}), while almost no positive effect was observed after moistening the biofilter on days 52 and 57. Apparently, other parameters were limiting the biofiltration process when the moisture content of the dolomite/compost mixture exceeded 15%.

Trickling a complete mineral medium through the dolomite/compost biofilter

Upon addition of 50 mL of a complete mineral medium to the dolomite/compost biofilter on days 65, 71, 79 and 107 and 50 mL of tap water on day 92, about 12 to 20 mL of acidified (pH 4–5) percolate was collected at the bottom of the biofilter, while the moisture content increased up to 35%. Contrary to the addition of tap water on day 92, the supplementation of a nutrient solution resulted immediately in a strong increase in EC. However, a gradual drop in EC between two

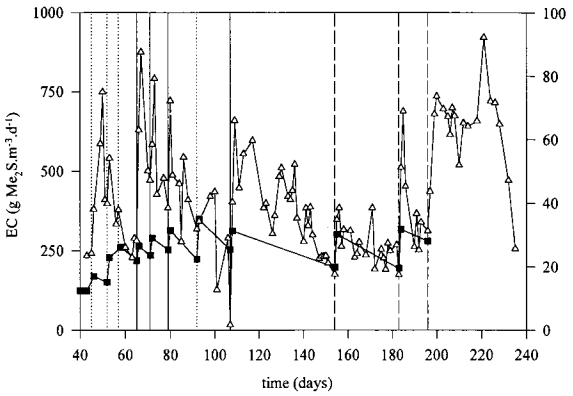

Figure 3. Elimination capacity (EC) (g Me_2S m^{-3} d^{-1}) of the dolomite/compost reactor (△) and moisture content of the carrier material (■) upon trickling tap water (dotted vertical lines on days 45, 52, 57 and 92), a complete mineral medium (full vertical lines on day 65, 71, 79 and 107) and a NH_4Cl-solution of 0.30 g L^{-1} (day 154), 3.00 g L^{-1} (day 183) and 30.00 g L^{-1}(day 196) (dashed vertical lines on day 154, 183 and 196) through the biofilter. The organic loading rate during this experiment was 2200-3500 g Me_2S m^{-3} d^{-1}.

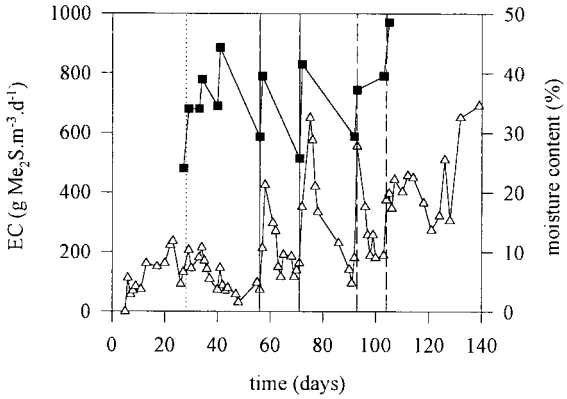

Figure 4. Elimination capacity (EC) (g Me_2S m^{-3} d^{-1}) of the dolomite/wood bark reactor (△) and pH-value of the carrier material (■) upon trickling tap water (dotted vertical line on day 28), a complete mineral medium solution (full vertical lines on day 56 and 71) or a NH_4Cl-solution of 3.00 g L^{-1} (day 93) or 30.00 g L^{-1} (day 104) (dashed vertical lines on day 93 and 104) through the biofilter. The organic loading rate during this experiment was 1000–3000 g Me_2S m^{-3} d^{-1}.

successive nutrient supplementations was observed, illustrating the occurrence of nutrient depletion.

Limiting nutrient in the dolomite/compost biofilter

Contrary to compost, dolomite is very poor in nitrogen content. However, nitrogen is an essential nutrient for microbial growth, since it makes up about 15% of the cell dry weight and is a major constituent of proteins and nucleic acids (Morgenroth et al. 1996). For this reason, nitrogen was used as a single nutrient in a next series of trickling experiments. On days 154, 183 and 196, 50 mL of a solution containing 0.30, 3.00 and 30.00 g L^{-1} NH_4Cl was brought on top of the biofilter (Figure 3). No peak EC for Me_2S was observed upon addition of the 0.30 g L^{-1} NH_4Cl solution (day 154). The concentration of 3.00 g L^{-1} NH_4Cl corresponded to the nitrogen concentration in the complete mineral medium. After addition of this 3.00 g L^{-1} NH_4Cl solution (day 183), a similar temporary increase in EC for Me_2S was observed as upon the complete mineral medium supplementation, suggesting nitrogen to be the limiting microbial nutrient in the biofilter. Upon trickling the 30.00 g L^{-1} NH_4Cl solution through the biofilter (day 196), a stable peak EC of 700 g m^{-3} d^{-1} was maintained over a 30-day period. In Table 1, it can be seen that high and stable ECs for Me_2S were obtained when NH_4Cl was dosed to the biofilter at a Me_2S–C/NH_4Cl–N ratio of about 10. The theoretical total amount of dolomite neutralised by the metabolite H_2SO_4over the 235-day dolomite/compost biofilter experiment was 76 g, being 23% of the dolomite material originally present in the biofilter.

The dolomite/wood bark biofilter

Upon mixing the dolomite material in the biofilter with *Hyphomicrobium* MS3-inoculated wood bark material, a maximum EC of 237 g Me_2S m^{-3} d^{-1} was obtained after a 3-week period (Figure 4). In accordance with the previous experiments, no effect was observed upon trickling tap water through the biofilter (day 28) when the moisture content of the carrier material exceeded 15%. Similar to the dolomite/compost biofilter, high (> 250 g Me_2S m^{-3} d^{-1}) and stable (> 35 days) ECs were obtained in the dolomite/wood bark biofilter when a solution of 30.00 g L^{-1} NH_4Cl was trickled through the biofilter. The theoretical total amount of dolomite neutralised by the metabolite H_2SO_4over the 139-day dolomite/wood bark biofilter experiment was 25 g, being 14% of the dolomite material originally present in the biofilter.

Discussion

Contrary to Bonnin et al. (1994) who successfully inoculated an inorganic Mearl biofilter with thiobacilli to optimise H_2S-removal, it was demonstrated in this work that inoculation of a pure dolomite biofilter

Table 1. Comparison between the NH_4Cl-N supplementation to the dolomite/compost biofilter and the cumulative Me_2S degradation between two successive nutrient additions

Period (days)	N-addition (g)	Me_2S-degradation (g)	C-degradation (g)	C/N ratio
155–183	0.004	4.37	1.69	430
184–196	0.04	2.46	0.95	24
197–235	0.4	12.45	4.81	12

Table 2. Literature overview of maximum elimination capacity (EC_{max}) (g Me_2S m^{-3} d^{-1}) obtained for Me_2S using different biotechnological waste gas systems (BF = biofilter; TF = trickling filter)

Reator	Substratum	Inoculation	EC_{max}	Reference
BF	Peat	Night soil sludge	77	Hirai et al. (1990)
BF	Peat	*Thiobacillus thioparus* DW44	97	Cho et al. (1991)
BF	Peat	*Hyphomicrobium* I55	114	Zhang et al. (1991)
TF	Polyurethane	*Hyphomicrobium* VS	329	Pol et al. (1994)
BF	Wood bark	No	10	Smet et al. (1996a)
BF	Wood bark	*Hyphomicrobium* MS3	35	Smet et al. (1996a)
BF	Compost	No	10	Smet et al. (1996a)
BF	Compost	*Hyphomicrobium* MS3	680	Smet et al. (1996a)
BF	Dolomite/wood bark	No	10	This work
BF	Dolomite/wood bark	*Hyphomicrobium* MS3	237	This work
BF	Dolomite/compost	No	10	This work
BF	Dolomite/compost	*Hyphomicrobium* MS3	1680	This work

with the Me_2S-degrading *Hyphomicrobium* MS3 culture was not successful. As a possible explanation for this failure, the absence of suitable protective colonising sites for *Hyphomicrobium* MS3 on these particles and the absence of microbial essential nutrients such as nitrogen can be suggested. It was shown in this work that organic materials like compost and wood bark can serve as an excellent carrier for the delivery of microbial inoculants into these biofilters (Figure 2).

As can be seen in Table 2, maximum ECs obtained in the *Hyphomicrobium* MS3-inoculated dolomite/compost and dolomite/wood bark biofilters were significantly higher in comparison with the corresponding *Hyphomicrobium* MS3-inoculated compost and wood bark biofilters (Smet et al. 1996a). More important, however, is the strong increase in performance stability of these inorganic biofilters. The reason for this is the combination of direct micro environment neutralisation of the metabolite sulphuric acid by the dolomite particles, together with the nutrient delivery of the organic materials used. Similar to that observed in the biofiltration tests with pure organic carrier materials, the compost/dolomite reactor performed better than the wood bark/dolomite reactor (Table 2) due to the higher nutrient content of the compost material.

When the biofilter performance was investigated over prolonged periods, both the moisture and nutrient content of the carrier materials were found to be the main parameters determining the removal capacity of the biofilter. In relation to the moisture content of the carrier materials used, it was observed that a minimum value of 15% is required for stable performance in these biofilters. When a complete mineral medium was trickled through the biofilter, peak ECs were obtained, suggesting that nutrient availability was limiting the long-term stability of the dolomite/compost and dolomite/wood bark biofilter (Figures 3 and 4). Moreover, nitrogen was found to be the limiting nutrient in these reactors. According to Morgenroth et al. (1996), not all nitrogen in bioreactors is recycled when the microorganism dies and lyses, because N_2 from denitrification or NH_3 can be lost to the atmosphere by stripping and NO_3^- can be leached out. In accordance with these authors, it was observed in this work that high and stable ECs for Me_2S can be obtained when nitrogen is dosed in a soluble form (NH_4Cl solu-

tion) at a C/N ratio of about 10. In order to limit the required trickling frequency over the biofilter and the concomitant production of a polluted waste water stream, further research will focus the addition of slow release nitrogen sources to these inorganic biofilters. According to Smith et al. (1996), nitrate should be preferred as the nitrogen source for biofilters and trickling filters in order to prevent clogging of the system. Indeed, growth yields for aerobic cultures are lower when using NO_3^-–N as a nitrogen source instead of NH_4^+–N.

As a conclusion, it can be stated that mixtures containing up to 66% by weight of a dolomite fraction can be used as an excellent substratum in biofilters degrading acidifying (Cl-, N- and S-containing) volatile compounds. Since the dolomite did not serve as a microbial attachment site for the *Hyphomicrobium* MS3, the dolomite had to be mixed with a fraction (33%) of an organic material. Following pH adjustment, the long term stability in these biofilters was shown to be improved by controlling the moisture content and by adding nitrogen at a Me_2S-C/NH_4Cl–N ratio of about 10.

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