

## LICENTIATE THESIS

## A Rockwool Biofilter for the Treatment of Restaurant Emissions

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### Licentiate thesis

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### **KUNSKAP**

Av Karin Boye

Alla de försiktiga med långa håvar träffar havets jätteskratt. Vänner, vad söker ni på stranden? Kunskap kan aldrig fångas, kan aldrig ägas.

Men om du rak som en droppe faller i havet att upplösas, färdig för all förvandling – Då ska du vakna med pärlemorhud och gröna ögon På ängen där havets hästar betar och vara kunskap.

### **PREFACE**

This licentiate thesis, a half fulfilment of a doctoral work, was carried out at the Division of Sanitary Engineering, Luleå University of Technology (LTU) between the years 1998 and 2000. The thesis consists of an introduction that summarises the work and three separate papers. The project was supported by Norrbottens Forskningsråd and Skandinavisk Ecotech AB (SEAB), who are gratefully acknowledged. Thanks to SEAB for initiating the research and providing the biofilter material used in this study. In addition, thanks to Stefan Spånberg, McDonald's, for assisting me with the full-scale biofilter and providing some of the equipment.

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Luleå in November 2000

Anneli Andersson Chan

### **ABSTRACT**

The use of biological air pollution control has shown its potential as an interesting treatment alternative for contaminated gas streams and extensive research and development has taken place over the past twenty years. Unlike conventional technologies, such as thermal and catalytic incineration or carbon adsorption, biofiltration allows effective pollution control at relatively low capital and operating costs, and without generation of secondary streams that may need a subsequent treatment. Biofiltration could be an appropriate method to treat restaurant waste gas whose emissions are often the cause of public complaints that could lead to expensive litigation or, in the worst case, eviction. However, since space is at a premium, there exists a need for a compact filter.

The objective of this licentiate thesis was to investigate the feasibility of a compact rockwool biofilter to treat emissions from a restaurant, to identify an appropriate microbial culture for the inoculation of the biofilter, and to evaluate different rockwool materials.

A full-scale biofilter installed at a fast-food hamburger restaurant was in operation for approximately one year. The restaurant staff observed a reduction of the odourous fatty smells and perceived that the filter effectively removed fat, making heat exchanging of the air possible, and leading to considerable energy savings. However, high deposits of grease caused problems with clogging, drying of the rockwool media, build-up of pressure drop, and the creation of anaerobic zones. The experiences from the full-scale biofilter initiated further studies on a pilot-scale biofilter, treating off-gases from a fryer. Fibre mats with pre-set structure replaced loose rockwool and a mechanical collector for grease aerosols was installed upstream of the biofilter that resulted in reduced pressure drops. Two specially designed rockwool mats, one hydrophobic and one hydrophilic, proved to possess good chemical and mechanical stability even when submerged in water. Bacteria from different environments were enriched in batch cultures, revealing that all cultures were able to use rape-seed oil, including its oxidation products, as their sole carbon and energy source. For maximum and lasting growth it was necessary to add a salt medium containing mainly phosphorus and nitrogen compounds, thus indicating the need for a nutrient control system in the biofilter. The rockwool material did not inhibit the growth of the cultures and seemed to have a certain buffering effect for acidic conditions. When inoculated in a pilot-scale biofilter it was shown that the mixed culture was able to immobilise and grow in the hydrophilic as well as hydrophobic rockwool media, with cell numbers ranging from 10<sup>4</sup> to 10<sup>10</sup> per gram dry media. Moisture content in the filter bed was, on average, 10 to 20% in the hydrophobic and 40 to 50% in the hydrophilic rockwool. The organic substances in the filter material (biomass, binder, and accumulated oxidation products from the oil) were evaluated as total volatile solids.

The off-gas from the fryer is a very complex mixture due to the constantly changing structure of the oil and typically contains a mixture of partially oxygenated hydrocarbons (i.e. carboxylic acids, aldehydes, ketones, alcohols, and esters). Also, significant differences in composition and concentrations were found between new and old rape-seed oil. The main odour causing compounds were suspected to be saturated and unsaturated aldehydes. Sampling of fatty acids and aldehydes before and after the biofilter indicated no significant reduction of total concentration. This was probably due to the short residence times in the filter (<10 seconds) in combination with low solubility of some of the components. It is also possible that more time is needed for adaptation of a proper bacterial community able to oxidize the pollutants.

### **SAMMANFATTNING**

Biologisk luftreningsteknik kan vara ett intressant behandlingsalternativ för förorenade gasströmmar och mycket forskning och utveckling har ägt rum inom området under de senaste tjugo åren. Till skillnad från traditionella tekniker, såsom termisk och katalytisk förbränning eller absorption i en aktiverad kolbädd, erbjuder biofilter en effektiv rening till relativt låga investerings- och driftskostnader, utan att generera sekundära föroreningsproblem. Filtrering genom en biologiskt aktiv bädd kan vara sätt att behandla restaurangemissioner som innehåller fetter från fritering och stekning med svåra luktproblem som följd. Dessa emissioner ger upphov till klagomål, som i sin tur kan leda till böter och i värsta fall vräkning av restaurangen. Dock måste det biologiska filtret vara relativt kompakt för att kunna byggas in i befintligt ventilationssystem.

Syftet med denna avhandling var att undersöka möjligheten at behandla restaurang emissioner i ett kompakt stenullsfilter, att identifiera en lämplig mikrobiell kultur för ympning av biofiltret, samt att utvärdera olika stenullsmaterials lämplighet som filtermedia.

Ett fullskalebiofilter installerades vid en hamburgerrestaurang och var i drift i ungefär ett år. Restaurangpersonalen observerade att mängden luktande föreningar minskade avsevärt och noterade att filtret effektivt avskiljde fett, vilket gjorde värmeväxling möjlig med energivinster som följd. De stora mängder fett som ansamlats i filtret skapade dock problem med igensättning, förhöjda tryckfall, uttorkning av stenullsmaterialet, samt anaeroba zoner. Erfarenheterna från fullskaleanläggningen ledde till vidare studier i ett pilotskale-biofilter, som behandlade emissioner från potatisfritering i rapsolja. Uppblåsta fibermattor med fast struktur ersatte den lösa stenullen och en mekanisk avskiljare för fetter installerades i kåpan ovanför fritösen. Detta resulterade i väsentligt reducerade tryckfall i biofiltret. Två speciellt framtagna stenullsmattor, en hydrofob och en hydrofil, visade sig vara kemiskt och mekaniskt stabila även under kontinuerlig bevattning. Bakterier från olika miljöer ympade och odlades i glasflaskor och det visade sig att alla testade kulturer kunde tillgodogöra sig rapsolja och dess oxidationsprodukter som enda kol- och energikälla. Tillsats av en saltlösning, innehållande bland annat fosfor- och kväveföreningar, krävdes för att få maximal tillväxt vilket visar att näring kan behöva tillsättas till biofiltret. Stenullsmaterialet hämmade inte bakterietillväxten och verkade ha en viss buffrande förmåga för sura förhållanden. Vid inympning i pilotskalebiofiltret visade det sig att den tillsatta blandkulturen immobiliserade och tillväxte i såväl det hydrofila som det hydrofoba stenullsmaterialet. Vid räkning av kolonier på agarplattor återfanns mellan 10<sup>4</sup> till 10<sup>10</sup> bakterieceller per gram torrt material. Fukthalten var i medeltal 10-20% i det hydrofoba och 40-50% i det hydrofila stenullsmaterialet. Organiskt material i filtermaterialet (biomassa, bindemedel samt ackumulerade oxidationsprodukter från oljan) utvärderades medelst glödgning.

Emissionerna från fritösen är en väldigt komplex blandning av partiellt oxiderade kolväten (tex. karboxylsyror, aldehyder, ketoner, alkoholer och estrar) eftersom oljan konstant ändrar struktur. Till exempel återfanns stora skillnader i innehåll och koncentrationer i emissionerna från fritering i ny och gammal rapsolja. De huvudsakliga ämnena som orsakar lukt misstänktes vara mättade och omättade aldehyder. Mätning av fettsyre- och aldehydkoncentrationer före och efter biofiltret påvisade ingen signifikant reduktion i filtret. Detta berodde troligen på de korta uppehållstiderna (<10 sekunder) i kombination med låg löslighet av några av komponenterna. Det är också möjligt att adaptionstiden måste vara längre för att hinna utveckla en bakteriekultur som klarar att effektivt oxidera föroreningarna.

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### **PAPERS**

- I. Andersson, A., Grennberg, K., *Isolation and characterization of a bacterial* population aimed for a biofilter treating waste-gases from a restaurant (submitted to Biotechnology and Bioengineering, 2000)
- II. Andersson, A., A study of a rockwool biofilter for the removal of odours, grease aerosols and VOCs in proceedings of the Air and Waste Management Association's 93<sup>rd</sup> Annual Meeting and Exhibition, Salt Lake City, Utah, USA, June 18-22, 2000
- III. Andersson, A., *Evaluation of rockwool biofilter media for the treatment of restaurant emissions* in proceedings of the 2000 USC-TRG Conference on Biofiltration (an air pollution control technology), University of Southern California, Los Angeles, California, USA, October 19-20, 2000

### 1. BACKGROUND

### 1.1 Biofiltration for air pollution control

During the past twenty years, the use of biological air pollution control has become a popular treatment alternative for contaminated gas streams. Unlike conventional technologies, such as thermal and catalytic incineration or carbon adsorption, biofiltration allows effective pollution control at relatively low capital and operating costs, and without generation of secondary streams that may need a subsequent treatment. Biofiltration has been considered economically advantageous in the treatment of large air flow waste streams, which contain low concentrations of volatile organic and inorganic compounds (VOCs and VICs), typically less than 1 g contaminant/m³ (Ottengraf, 1987; Leson & Winer, 1991; Kennes & Thalasso, 1998; Devinny, *et al.*, 1999). Organic compounds such as alcohols, aldehydes, ketones, and carboxylic acids, as well as inorganic compounds such as hydrogen sulphide and ammonia, demonstrate excellent biodegradability (Leson & Winer, 1991; Devinny *et al.*, 1999).

Biofiltration has been used for many years in Germany and the Netherlands (VDI Berichte, 1991; Leson & Winer, 1991), Japan (Ando, 1980), and, to a limited extent, also in the United States (Pomeroy, 1957; Bohn, 1975). More recent experiences can be found for example in Canada (Mohseni & Allen, 1996), New Zealand (Luo & Lindsey, 2000), Russia (Popov et al., 2000), and Asia (Hsu, et al., 2000; Hwang et al., 2000). Successful applications of this technology in Europe include the abatement of odourous organic and inorganic gases from a variety of industrial and public places, e.g. food processing (Koch et al., 1982; Don, 1985; Leson & Winer, 1991). Presently, over 600 biofilters and bio-scrubbers in Europe are believed to be active in deodorising and removing volatile organic compounds (VOCs) from waste gases (Leson & Winer, 1991; Fouhy, 1992). Motivated by new legislations, such as the 1990 Clean Air Act amendments in the U.S., significant fundamental and applied research on biofiltration has taken place during the last two decades. Also, a number of extensive reviews and studies about the development and technical aspects of biofiltration have been published in the past decade (Leson & Winer, 1991; Togna & Singh, 1994; Edwards & Nirmalakhandan, 1996; Swanson & Loehr, 1997; Wani et al., 1997; Kennes & Thalasso, 1998; D'Amato & DeHollander, 1999; Jorio & Heitz, 1999).

In a biofilter, contaminated streams are vented through a biologically active material where microorganisms, which have the ability to degrade organic/inorganic pollutants, are immobilized and form biofilms on the surface of the solids. As the contaminated stream passes through the filter bed, pollutants are transferred from the vapour phase to the biofilm where they are metabolised. The complete degradation of air contaminants yields CO<sub>2</sub>, water, and microbial biomass.

### 1.2 Factors affecting biofilter performance

The efficiency of a biofilter is dependent upon many parameters including biofilter packing medium, bed configuration, the contaminants loading rate, and dynamic mass loading. Also, operational parameters such as moisture content, temperature, pH, and microbial activity must be considered in order to optimise biofilter performance.

### 1.2.1 Design criteria and moisture control

The type of construction and installation of a biofilter for a given application will depend primarily on the availability of space relative to the required filter volume. Multistage systems could be advantageous if space constraints exist (Leson & Winer, 1991); they also offer a high degree of process flexibility and are advantageous when treating mixtures (Swanson & Loehr, 1997). The size required for a biofilter to remove air pollutants efficiently depends primarily on the loading rate and concentration of the compounds in the off-gas, and the rate of biodegradation per volume (Leson & Winer, 1991).

Maintaining an optimum moisture level is a key operational requirement for a biofilter. Moisture is necessary for the survival and metabolism of the microorganisms (Leson & Winer, 1991; Devinny *et al.*, 1999). Without providing adequate moisture, and especially if the raw gas is not saturated with water, the filter bed would quickly dry (Swanson & Loehr, 1997; van Lith, Leson & Michelsen, 1997). Overall, too little moisture may result in reduced biological activity and, therefore, a risk of breakthrough of incompletely treated gas. However, too much moisture can lead to compaction, clogging, and the formation of anaerobic regions (Corsi & Seed, 1995; Devinny *et al.*, 1999). In general, a moisture content between 40 and 60% by wet weight is recommended to obtain optimum biodegradation (Ottengraf, 1986; Leson & Winer, 1991; Wani *et al.*, 1997).

### 1.2.2 Biofilter bed material

The properties and characteristics of the support medium largely govern the overall effectiveness of a biofilter. The medium should have a high moisture retention capacity to prevent drying and a large surface area for high mass transfer and microbial attachment. It should provide mechanical support for the maintenance of the filter bed's internal structure, and a high porosity and/or void space to reduce head loss and ensure even distribution of incoming waste gas (Leson & Winer, 1991; Swanson & Loehr, 1997; Kennes & Thalasso, 1998; Devinny *et al.*, 1999). Compaction of the medium over time needs to be minimised in order to prevent increases in system pressure drops and/or channelling of untreated polluted gas through the biofilter. It is also desirable for the filter medium to have a significant pH buffering capacity to prevent acidification due to the build-up of acid laden waste gas streams (Leson & Winer, 1991). At high surface loads, the filter material will become more susceptible to dehydration and heat losses caused by insufficient raw gas conditioning (Leson & Winer, 1991). The composition of packing materials has been improved in recent years and various synthetic packing materials have been developed to retard the effects of ageing and maintain the bed porosity (Ottengraf, 1987; Devinny *et al.*, 1999).

Fibre based materials have been successfully used, for example, in biotrickling filters (Rydin et al., 1994; Wittorf et al., 1997; Ostlie-Dunn et al., 1998; Kozliak & Riley, 2000). The advantage of using fibres instead of granules is the higher surface-to-volume ratio that can be obtained, which improves the substrate mass transfer and provides more surface area for adsorption and microbial immobilisation. Rockwool fibre mats with pre-set structures and low densities are less subject to compacting and ageing and their use facilitates handling and improves flow distribution. In addition, the characteristics of the rockwool can be specifically designed, i.e. density, fibre length and thickness, amount of binder, hydrophobic/hydrophilic properties, etc, making it a very versatile filter medium.

### 1.2.3 Microbial community, temperature, pH, and inorganic nutrients

The presence of microorganisms capable of degrading VOCs is necessary for the biofiltration of waste gas streams. The growth and metabolic activity of microorganisms in a biofilter depend on many parameters including a suitable temperature and pH range. The temperature of the biofilter is mainly influenced by the temperature of the inlet air stream, and, to a limited extent, the exothermic biological reactions in the bed (Corsi & Seed, 1995). Low temperatures enhance sorption, but slows down the rate of microbial activity (Mc Nevin & Barford, 2000). Temperatures between 25 and 35°C have been suggested as suitable for biofilter performance, with 35°C often noted as the optimum temperature for aerobic microorganisms (Leson & Winer, 1991; Lee *et al.*, 1996; Swanson & Loehr, 1997). Biofilters are most frequently studied at mesophilic temperatures, even though there are some studies of thermophilic operations (Allen *et al.*, 2000; Cox & Deshusses, 2000). Microorganisms capable of degrading VOCs show optimum growth at pH values between 6 and 8 (Ottengraf, 1986; Leson & Winer, 1991; Shoda, 1991; van Lith *et al.*, 1997). This condition usually exists in a biofilter except when treating chemicals whose biodegradation results in acid end products such as H<sub>2</sub>S, and chlorinated compounds (Devinny *et al.*, 1999).

The microorganisms in biofilters consume contaminants for the energy and carbon they provide; however, they also need inorganic nutrients such as nitrogen, phosphorous, potassium, sulphur, and many others. Since synthetic media, like rockwool, does not contain an appropriate supply of nutrients, these must be added separately during operation. Little information exists on nutrient cycles and biofilter requirements at this time.

### 1.3 Lipid oxidation

Lipid oxidation is a very complex system of reactions. Basically, it is a radical reaction that produces hydroperoxides. These have no taste or smell, but are unstable and decompose into volatile and non-volatile substances. The majority of these decomposition products are aldehydes, but also formed are ketones and peroxides(Grosch, 1987; Moortgat *et al.*, 1992; Leissner *et al.*, 1993; Andersson, 1998). In general, volatile aldehydes have strong tastes and smells. Edible fats and oils both hydrolyse and cleave at the double bonds by oxidation when exposed to heat, air, and light (Petrucci, 1989).

The following external factors increase the degree of fat oxidation (Andersson, 1998):

- Temperature; every 10°C rise in temperature doubles the rate of oxidation
- ➤ Light; oxidation sensitivity increases with exposure to light
- > Oxygen concentration and the degree of unsaturation of the fatty acid
- Presence of metals or antioxidants

When water or steam is added to heated oil, volatile substances will take off in the emissions. The majority of these substances have higher vapor pressure (i.e. are more volatile) than the triglycerides that will for the most part stay in the oil (Leissner *et al.*, 1993).

### 1.3.1 Degradation of rape-seed oil

Lipids are biodegradable, abundant in nature, and can be excellent substrates for microbial energy-yielding metabolisms (Madigan *et al.*, 1997). Microorganisms utilize lipids after hydrolysis of the ester bonds; extracellular enzymes called lipases are responsible for the reactions. The results of the lipase actions are glycerol, mixtures of mono- and diglycerides, and fatty acids (Petrucci, 1989). Glycerol is converted to glyceraldehyde-3-phosphate (triose phosphate) and joins into the glycolysis. With the release of energy, fatty acids are oxidized to carbon dioxide and water in a series of reactions known as  $\beta$ -oxidation. In this process, oxidation occurs at the  $\beta$ -carbon atom of the fatty acid, followed by cleavage. This means that two-carbon pieces (acetic acid) are split off. The process requires the presence of coenzyme A (Petrucci, 1989).

### 1.3.2 Treatment of mixtures

The use of biofiltration has worked particularly well for processes that emit a steady gas stream containing one or two contaminants. However, in real case applications, multi-component pollution is more often the rule than the exception. One example is odorous, fatty gas emissions from restaurant facilities that typically contain a mixture of partially oxygenated hydrocarbons whose composition and concentration vary with time.

It is often difficult to anticipate biofilter treatment success of mixed pollutants (Devinny *et al.*, 1999). Performance will depend on contaminants characteristics such as solubility, adsorptivity, bond structure, potential biodegradability, and operating conditions. Competitive effects between pollutants can be important in both the mass-transfer and biodegradation steps of the biofiltration process (Mc Nevin & Barford, 2000). This is especially true for biodegradation where inhibition can occur due to preferential uptake (diauxy) of one substrate over another or by toxic interactions (Ergas *et al.*, 1995). An extreme case of negative interaction between pollutants was reported by van Langenhove & Smet (1996), who observed that a removal of an undefined mixture of aldehydes was reduced from 85 to 40% after the addition of 40 ppm SO<sub>2</sub> to the treated air stream.

Leson & Smith (1997) and Swanson & Loehr (1997) noted that greater time may be necessary to complete acclimation of biofilters treating complex mixtures; Devinny *et al.* (1999) noted that times needed could range from several minutes to as much as a year. When using a biofilter for the degradation of an aldehydes-containing waste gas, Don (1985) did not achieve any purification of the gas even after an operational period equal to six months. However, by starting up with a diluted waste gas and gradually increasing the concentration, a degradation capacity of 40 g C/m³h was attained after four months. When the filter was inoculated with active sludge from a a factory with aldehydes-containing wastewater, a similar degradation capacity was found after only two weeks of adaptation, thus showing the importance of a proper inoculum.

### 1.3.3 Grease aerosols

Cooking discharges contain substantial amounts of grease aerosols that can clog biofilters used in the treatment of food preparation exhaust (Devinny *et al.*, 1999). Large grease emissions can cause problems such as complaints from neighbours, bad working environments (i.e. slippery floors and odours), clogging of pipes and ventilation units, etc. Considerable energy savings could be made through heat exchange of the air if the grease aerosols were removed.

### 1.4 Evaluating a biofilter

### 1.4.1 Odour measurements

The amount of contaminant being removed in a biofilter is the primary measure of its effectiveness. The sophistication of contaminant monitoring systems varies widely. To monitor a treatment plant for odours, which vary and have very low concentrations, the human nose is often the best instrument. Human sensors allow for the detection of very low levels of some compounds (as low as parts per trillion in some cases) in a complex odour (Hodgins, 1995). Even small emissions can cause a nuisance even if they do not directly endanger health (Ottengraf, 1987). Quantitative measurements can be made by odour panels, or so called olfactometry (Schulz & van Harreveld, 1996; Walsh, 1996; Devinny et al., 1999). These groups of individuals sniff samples of air to determine whether an odour is present. They are provided with samples that have been diluted to various degrees, so that in some the odour is no longer perceptible. The concentrations of odour-causing contaminants are often measured in odour units per cubic meter (OU/m<sup>3</sup>), equal to the dilution factor required to reduce the concentration of the contaminant to its odour threshold. The olfactometric methods, however, are time consuming, expensive, labour intensive, and subject to large variations between analyses. An electronic nose, consisting of a non-specific sensor array, could offer a more objective means of measuring odours (Fenner & Stuetz, 1999).

### 1.4.2 Analytical techniques

The combination of Gas Chromatography and Mass Spectrometry (GC-MS) is the most common instrumental method for measuring contaminant concentrations in applications for air pollution control. GC-MS has an excellent sensitivity (about 0.2 ppb) and can ideally be used in the identification and quantification of odours (Walsh, 1996; Devinny *et al.*, 1999). Gas chromatography separates individual components according to their vapour pressures and solubility inside the GC column material. Mass spectrometry identifies the eluted components by their ionized molecular fragmentation patterns. GC-MS can be used to determine chemical concentrations and compositions of an odorous sample. The major limitation of this technique is that identification remains ambiguous or questionable as a result of the presence of unknown components at very low concentration levels (ppt). No indication is obtained as to the relevance of individual compounds in relation to the odour of the sample as a whole. Even if individual chemical concentrations and their odour threshold values are known, it is not possible to deduce the overall sample odour threshold or the odour character of the mixture of odours.

Grab samples can provide false results for the control efficiency since pollutant concentrations in the filter effluent do not react instantaneously to variations in the raw gas. The continuos off-gas monitoring for total organic carbon (TOC) with a flame ionization detector (FID) or photoionization detector (PID) addresses this problem, and is widely used for compliance testing. However, in the case of multi-contaminant off-gases, different FID/PID response factors and removal rates for different components can result in inaccurate figures for TOC and control efficiency (Leson & Winer, 1991). They also have limited capacity to measure low concentrations of contaminants.

### 2. OBJECTIVES

The objective of this licentiate thesis was to investigate the feasibility of a compact rockwool biofilter to treat air emissions from a restaurant that contained grease aerosols, VOC's, and odorous components. Identification of an appropriate microbial culture for the inoculation of the biofilter, as well as suitable environmental conditions for this culture was also carried out. Finally, different rockwool filter media was evaluated with respect to flow distribution, pressure drop, chemical and mechanical resistance, and aptness as immobilisation matrices for microorganisms.

### 3. METHODS

### 3.1 Batch laboratory experiments (Paper I)

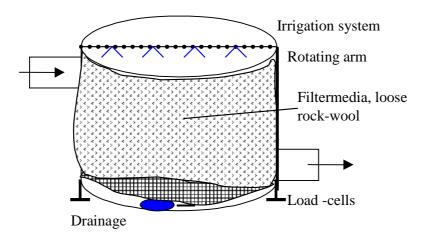
The isolation and characterization of a bacterial population suitable for inoculation of a rockwool biofilter were carried out in laboratory batch experiments in two phases (**Paper I**). Cultures from three various bacterial sources were studied under different chemical and physical conditions and compared to the growth of *Bacillus sp*, recommended for biofilter treatment of restaurant gases. The experimental set-up can be viewed in Figure 1. The effects of adding salt medium and/or rape-seed oil as a carbon and energy source were evaluated by means of viable count and pH monitoring. The effect of adding new rockwool was also assessed. These cultures were then used as inoculum in the pilot-scale biofilter (**Paper II and III**).



Figure 1. Experimental set-up for the laboratory batch experiments

### 3.2 Full-scale biofilter (Paper II)

Experiences from a full-scale biofilter (Figure 2), which was installed at a Swedish hamburger restaurant, are presented in **Paper II**. The operational parameters can be found in Table 1. Peat was originally used as filter material, but was replaced with an inert, loose fibre material (rockwool). The filter, which ran for approximately one year, was inoculated with a mono culture of *Bacillus sp*. Medium sampling was performed after 10 months of operation when moisture content, total volatile solids, pH, and viable count were established. Concentrations of fatty acids in the air before and after the biofilter were also measured.



**Figure 2.** Schematics of the full-scale biofilter

**Table 1**. Operational parameters for full- and pilot-scale biofilters

	Full-scale biofilter	Pilot-scale biofilter,	Pilot-scale biofilter,
		first test-run	second test-run
Filter area (m <sup>2</sup> )	3.2	0.36	0.36
Media depth (m)	0.40	3*0.3	3*0.3
Experimental length	1 year	28 days	15 days
Average flow (m <sup>3</sup> /h)	2700	400	140
Average residence time (s)	2	3	9
Average pressure drop (Pa/m)	4500	1700	250

### 3.3 Pilot-scale biofilter (Paper II and III)

The experiences from the full-scale biofilter initiated further studies on a pilot-scale filter (**Paper II and III**). The main design criterion for the pilot filter was a compact, multi-stage biofilter easy to place and handle in a restaurant environment. Therefore, the construction was changed from the full-scale filter with a circular area and vertical flow mode to a filter with a square area, composed of three filter units operated in a horizontal flow mode, see Figure 3. Two test runs were performed and the operational parameters for them are described in Table 1.

The pilot-scale biofilter was coupled to a potato fryer with rape-seed oil heated to 180°C. Sampling of the filter media was performed regularly to establish pH, moisture content, total volatile solids, and viable count. Sampling of the aldehydes and TVOC (Total Volatile Organic Compounds) were carried out to characterize the emissions, and sampling for aldehydes was performed before and after the biofilter at the end of each experimental period.

Filterbox

Filterbox

Filterbox

Filterbox

Filterbox

Fryer with rape-seed oil

Fryer with rape-seed oil

The three filter units, filled with hydrophobic and hydrophilic rock-wool fibre mats

Figure 3. Schematics of the pilot-scale biofilter

In **Paper III**, an evaluation of six different rockwool media, from three different manufacturers, is presented. Flow distribution and pressure drop were measured in the pilot-scale filter at a moisture content of 40%. The chemical and mechanical resistance, as well as the rockwool's aptness as immobilisation matrices for microorganisms, were visually evaluated after ten days agitation in a sludge suspension.

### 4. Major results and Discussion

### 4.1 Batch laboratory experiments (Paper I)

All cultures tested were able to use the rape-seed oil as their sole carbon and energy source. For maximum and lasting growth it was necessary to add a salt medium containing mainly phosphorus and nitrogen compounds. The rockwool biofilter material did not inhibit the growth of the bacteria and seemed to provide a certain buffering capacity for acidic conditions. An exponential growth phase during a period of 3 to 8 days with an increase of cell numbers by a factor of 10<sup>3</sup>-10<sup>5</sup>, with generation times of 9 to 33 hours, was followed by slower growth. After a stationary phase of 25 to 40 days, the bacterial number started to decrease. Metabolic activities of the growing microbial population could have changed the nature of the environment to the point where it became unfavourable. This may have been brought about by the decreasing pH, by the depletion of nutrients and/or oxygen, or by the accumulation of toxic metabolites. It was obvious when the colony morphologies were studied at each plate count that the number of different bacterial species had decreased with time and that a natural selection of bacteria able to survive in the batch environment had developed.

The addition and degradation of rape-seed oil caused pH to decrease in the batch cultures, occasionally down as low as pH 3. However, this did not seem to influence the number of cells, which indicates a high bacterial tolerance to acid conditions. When oil was provided in excess, the bacterial populations may have multiplied by hydrolysing the rape-seed oil and preferentially using the glycerol part of the oil as their carbon and energy source, but may have left the fatty acids unmetabolised. This could then explain the decreasing pH in the

flasks. The fatty acids could have become growth inhibitory or toxic if the concentrations were too high. As long as rape-seed oil was supplied in abundance, the microbial population was never "forced" to use the fatty acids as their carbon and energy source; this would mean that only the glycerol part, or about 5% of the carbon in the rape-seed oil, was utilized.

A simplified calculation of the maximal number of bacteria that theoretically could be attained with the carbon present in the system and no limitations (such as lack of oxygen or temperature or pH inhibition) was compared to the highest number of cells experimentally obtained with the viable count method. It was assumed that about 1/3 of the substrate carbon was incorporated into cells (and thus available for growth) and about 2/3 were converted to CO<sub>2</sub> (Mathur, 1991). This calculation showed that only about 7% of the carbon available for growth were used. If this would be the case, it might become difficult to obtain complete degradation of the oil and its oxidation products in a biofilter where large amounts of grease are deposited (surplus of carbon).

The amount of oil was decreased in phase two of the batch experiments, to assure that the principal carbon source would be the limiting substrate for the bacteria. The calculation then showed that about 55% of the carbon available for growth were used in these flasks, which also were in reasonable agreement with results obtained when measuring COD in the batch suspensions (**Paper I**). This would thus indicate enhanced carbon utilization; the bacteria were "forced" to use the fatty acids as well. An ideal metabolic carbon utilization is obviously never possible to obtain, but the calculations gave an idea of how well the bacteria used the carbon and energy substrate available for growth. One should also bear in mind that the viable count method might underestimate the real number of bacteria since only cultivable, viable cells are detected on the agar plates used, which might constitute only a part of the total population.

The appearance of the batch suspensions changed during the experimental period from a transparent liquid with oil floating on the surfaces to a thick, yellow-white suspension after 15-20 days in flasks where oil was provided in excess. This could be due to the bacterial production of surface-active substances, which transformed the oil into an emulsion, and made the oil readily available for the microorganisms. In flasks where oil were the limiting substrate, the same phenomena occurred during the first 10-15 days, but around day 35 the suspensions turned back to transparent yellow liquids with white particles in suspension. This could be a indication of fungus growth favoured by the acid conditions prevailing in these flasks or be a sign of the formation of micelles. Fatty acids tend to form micelles – colloidal particles that may aggregate and precipitate from solutions during environmental changes such as changes in pH, temperature, and salt concentrations (Keenan & Sabelnikov, 2000).

One of the mixed cultures was further enriched and compared to a culture of *Bacillus sp*, with a few simple biochemical tests (Madigan *et al.*, 1997). Both were found to be aerobic rods. The *Bacillus sp* culture was gram-positive with the ability to form endospores, while the bacterium from the mixed culture was gram-negative and did not have the ability to form endospores. Both were mesophiles and grew well in the temperature range of 21 to 37°C. Since all cultures were able to use the rape-seed oil they were considered suitable for inoculation of a biofilter that treats waste gases from a frying process with rape-seed oil. However, it is important to bear in mind that conditions in batch laboratory flasks and in a biofilter differ greatly.

### 4.2 Full-scale biofilter (Paper II)

The restaurant staff was generally satisfied with the filter performance, the filter removed fat effectively, which made heat exchanging of the air possible, and led to energy savings. Reductions of the odourous fatty smells were also observed. However, with time, the decrease in flow caused problems in the kitchen since odorous and fatty emissions remained there. Sampling of the filter media after 10 months of operation revealed that the moisture content and the bacterial numbers were very low.

The conditions in the filter seemed to be very heterogeneous; a thick layer of grease on top of the filter material obstructed irrigation directly and also indirectly by disturbing the function of the load cells in the bottom of the filter. These were programmed to start the rotating irrigation arm at a certain weight loss (due to evaporation and drying), but this rarely occurred since grease accumulated in the filter, thus increasing the total weight. Sampling of the filter media at different levels showed that the moisture content was very low (less than 5%), except in one sample (70%), and that the grease accumulated in the filter media exceeded 50% (calculated from the total volatile solids) in some samples. Figure 4 show a new rockwool fibre, and one that have been coated with biofilm and fatty oxidation products. pH in the filter media was around 4, and viable count on agar plates showed less than 1\*10² bacteria/g dry media in the first two samples and 3\*10<sup>7</sup> bacteria/g dry media in the sample with the higher moisture content. No bacteria from the *Bacillus sp* culture originally inoculated in the filter were found. Results from parallel air samplings of fatty acids before and after the biofilter on two occasions showed a large spread both in composition and total content (**Paper II**), and no significant reduction of fatty acid concentration could be shown.

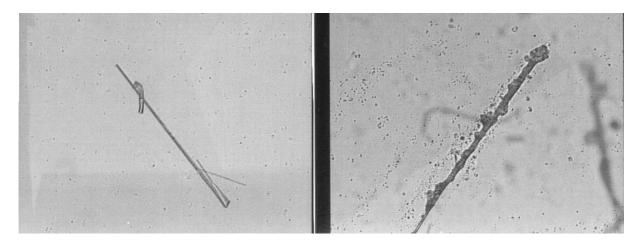


Figure 4. New rockwool fibre (left) and fiber after one year of operation (right). 400 times enlargment

### 4.3 Pilot-scale biofilter (Paper II and III)

Sampling of the rockwool media revealed that moisture content varied between samples, but overall the humidification system seemed to work satisfactorily. As expected, the hydrophilic rockwool was able to hold water better than the hydrophobic, also resulting in a higher number of bacteria in the hydrophilic media samples. The numbers of bacteria found in this study were comparable to those found previously (Medina *et al.*, 1995; Cardenas-Gonzalez *et al.*, 1998; Acuña *et al.*, 1999).

By studying the morphology of the colonies, changes in the bacterial populations could be discovered. After less than a week, the specialist culture of *Bacillus sp* was significantly depleted, such that they were undetectable on agar plates. The mixed culture enriched with rape-seed oil were the dominating culture over the entire biofilter.

The total volatile solids gave an estimate of how much organic material (including binder, biomass, and accumulated oxidation products from the oil) was present in the filter material during the experimental period. The rockwool initially contained about 1.8% (hydrophobic) and 0.6% (hydrophilic) of binder, a phenol formaldehyde resin. A higher sorption of fatty oxidation products was noticed in the hydrophobic material, about fivefold the amount sorbed in the hydrophilic material.

pH in media samples and drainage was found to be relatively stable at 7 (±1) during both experimental periods, indicating that the material could possess a certain buffering capacity. Temperatures in the filter below 0°C were detected on a few occasions when the outdoor temperature was below -25°C. However, no decrease in bacterial numbers was detected. Nevertheless, this showed that considerations must be taken to the placement of the filter in a full-scale application in cold climates. Bed temperatures between 10 and 40°C are acceptable for the mesophilic micro-organisms most frequently present in a biofilter, but one should strive to keep the off-gas temperature close to their optimum range for biological activity (30-35°C) (Leson & Winer, 1991; van Lith *et al.*, 1997).

The mechanical collector for grease aerosols, composed of one metal and one textile fibre filter installed upstream of the biofilter, proved to be efficient. Very small depositions of grease and fatty oxidation products in the channel as well as in the rockwool filter media could be detected. However, the collector needs to be cleaned or changed on a regular basis, since the accumulation of grease contributes to increased pressure drops in the system.

The composition and structure of the rape-seed oil is constantly changing when exposed to heat, light, oxygen, and potatoes, which makes a clear definition of the emissions virtually impossible. In addition, significant differences in composition and concentrations were found between new and old rape-seed oil (Paper II). Overall, very low concentrations of TVOCs, aldehydes, and fatty acids were detected (<20 mg/m<sup>3</sup>), making the sampling difficult. However, these low concentrations still caused rather strong odours, indicating that some of the components have very low threshold values. Air samples taken before and after the biofilter at the end of the experimental runs indicated that no reduction of total aldehyde concentrations was achieved in the biofilter (Paper II and III). A few compounds, i.e. hexanal, were reduced by 10-35%, but the total concentration of aldehydes increased. The most probable explanation for this is too short residence times in combination with poor water solubility for some of the aldehydes. Contaminants must move from the air phase into the biofilm in order to be biodegraded; the time provided for mass transfer was probably too short. Residence times will probably have to be substantially increased to obtain a significant reduction. Typical residence times needed for commercial applications range from 25 seconds to several minutes (Leson & Winer, 1991; Devinny et al., 1999). To achieve this, a larger surface area of the filter will most likely be needed, which in turn poses a challenge since restaurant space is limited. Several filters in parallell could be one way to approach this problem.

Since the experimental runs were fairly short (28 and 15 days), there is also a possibility that the proper bacterial culture did not have enough time to adapt and become active in degrading the aldehydes. The viable count method does not provide any information about the degradation activity of the microorganisms, but merely gives an indication of the number of cells that can grow under the prevailing conditions. The bacteria in the biofilter, previously enriched with rape-seed oil, could have survived merely with the various oxidation products that accumulated in the filter, and may have had little interest in the passing air-borne aldehydes. Physical adsorption on to the filer material may have been exhausted due to saturation and might even have caused a release of aldehydes from the filter. It is also possible that more time is needed for acclimation of a proper bacterial community able to oxidize the pollutants. Typical residence times needed for commercial applications range from 25 seconds to several minutes, thus raising the need for a larger surface area of the filter, posing a challenge since space is limited. However, a solution with several filters in parallel could be one way to solve this problem.

### 5. CONCLUSIONS

Operation of a full-scale rockwool biofilter at a fast-food restaurant indicated that odourous fatty smells could be reduced and the removal of grease aerosols made heat exchanging of the air possible, which led to energy savings. However, to obtain an environment favourable for biological degradation and to avoid clogging, a large part of the grease aerosols must be removed prior to the biofilter.

In a pilot-scale biofilter, the implementation of a mechanical collector for grease aerosols and an improved design for moisture and nutrient control proved successful. The bacterial inoculum used was able to immobilize and grow in the rockwool media with cell numbers ranging from  $10^4$  to  $10^{10}$  per gram dry media.

However, the residence times employed in this study (<10 s) were not enough to obtain a significant reduction of neither aldehyde nor fatty acid concentrations in the air stream. It can be concluded that the off-gas from the fryer is very complex, containing a mixture of hydrophobic and hydrophilic compounds.

Finally, this study showed that rockwool could be a suitable biofilter medium. In two specially designed rockwool mats, low pressure drops were developed. Further, they were easy to handle, prevented compacting, proved to possess good chemical and mechanical stability even when submerged in water, and were found to be suitable for microbial immobilization.

### 6. OUTLOOK

This work has provided some answers and generated lot of new questions, as academic work often does. Biofiltration is a complex process and it is a challenge to provide the microorganisms with the right environment to enhance the degradation of pollutants. More work is needed to establish at what residence times it is possible to obtain an effective treatment. To do that, it is necessary to understand how the pollutants are captured (mass-transfer) and subsequently biodegraded and which of these two phenomena that is the limiting step. A short residence time equals a compact and more economical biofilter, which would be of interest in many applications.

The analytical difficulties to measure low concentrations of a complex mixture of contaminants also present a challenge. Is there a way to perform cheap and reliable measurements to evaluate the biofilter performance? The characteristics of complex odours cannot be derived reliably from the individual chemical characteristics and concentrations of the compounds present in a gas mixture. The human nose is often the best instrument to assess odours, so far superior to any analytical instrument. However, dynamic olfactometry measurements are costly and tedious. The development of electronic noses could in the future become an interesting measuring alternative (Hodgins & Simmonds, 1995); Fenner & Stuetz, 1999).

To assess the rockwool media properly, long-term testing will be necessary to get an idea of e.g. pressure drop development and chemical and physical stability. Refinement of the moisture and nutrient control system for the biofilter might be necessary. More research and development is needed before the rockwool biofilter can be employed successfully in full-scale applications to treat restaurant emissions.

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### **PAPER I**

# Isolation and characterization of a bacterial population aimed for a biofilter treating waste-gases from a restaurant

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submitted to Biotechnology and Bioengineering

### Isolation and characterization of a bacterial population aimed for a biofilter treating waste-gases from a restaurant

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### **ABSTRACT**

Biofiltration units are microbial ecosystems for the treatment of low-concentration, biodegradable waste gases. The aim of this work was to isolate and characterize an appropriate microbial culture for inoculation of a rockwool biofilter that treats waste gases from a fast food restaurant using rape-seed oil in its frying process. Batch cultures from three various bacterial sources were studied under different chemical and physical conditions and compared to the growth of Bacillus. All cultures were able to degrade rape-seed oil and its oxidation products, with a simultaneous pH decrease in the batch cultures. It was necessary to add a salt medium containing e.g. phosphorus and nitrogen to obtain maximum growth. An exponential growth phase during a period of 3 to 8 days, with generation times of 9 to 33 hours, was followed by slower growth. After a stationary phase of 25 to 40 days, the bacterial number started to decrease probably due to substrate and/or oxygen depletion, or unfavourable pH. The rockwool biofilter material did not inhibit the growth of the bacterial cultures and seemed to have a buffering capacity, which could prevent acidification of a future biofilter. The isolated bacteria from the mixed culture were found to be mesophilic, aerobic, gram-negative rods.

### **KEY WORDS**

Biofilter; Rape-seed oil; Rockwool filter; Inocula; Viable count

### INTRODUCTION

Microbial reactions have been used extensively to treat wastewater and solid waste throughout the twentieth century, but it has only been since the 1950s that such techniques have been used to treat waste gases (Pomerov, 1957). During the last decades, biofiltration for air pollution control has been established as a reliable, cost-effective technology for controlling low-concentration biodegradable waste gases from a wide range of industries and public sectors. Organic compounds such as alcohols, aldehydes, ketones, and carboxylic acids, as well as inorganic compounds such as hydrogen sulphide and ammonia, demonstrate excellent biodegradability (Devinny, et al., 1999). The microorganisms may grow in a biofilm either on the surface of a porous medium or suspended in the water phase surrounding the medium particles. The filter bed medium often consists of an organic material like compost, soil, or peat, having the advantage of containing a large diversity of microorganisms and nutrients. However, problems with degradation of the material, compacting, and the development of large pressure drops have led to a more extensive use of synthetic materials in different packing mixtures. Fibrous materials, e.g. rockwool, have several advantages over other materials in that they are light, flexible, low in pressure drop, less microbially degradable, and easy to handle (Shoda, 1991).

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### Inoculation of a biofilter

A healthy population of microorganisms is fundamental for a successful biofilter operation. Inoculation of the filter bed is necessary if a synthetic filter material is used, as in the case of rockwool. Inoculation can also be an effective way to shorten the start-up or adaptation time of the biofilter and/or enhance the degradation rate of pollutants, or to resume normal conditions quickly if microbial activity deteriorates during operation (Shoda, 1991, Wani, et al., 1997). However, enriched pure cultures from a laboratory environment may have difficulties surviving in field conditions. The inoculation species have to be able to grow at the pH and temperature prevalent in the biofilter, to use the nutrients available, and to grow fast enough to survive competitive organisms and avoid predators. Natural selection will play its role and the more efficient organisms will become dominant. Mixed cultures often originating from wastewater treatment plants or from similar origin have been used as inoculum in many cases (Deshusses, et al., 1995; Ergas, et al., 1995; Morgenroth, et al., 1996). This type of general inocula has the advantage of containing a vast variety of rugged organisms with a wide degradative range and an ability to also work in a fluctuating environment. However, there may be some concern regarding the risk of pathogens, especially when the biofilter is to be situated in a restaurant environment. For such applications, inoculation of a laboratory grown defined mixed culture would, therefore, be preferable. The choice and preparation of a proper inoculum is an important matter for research, especially in the use of biofilters in the food industry.

### Degradation of rape-seed oil

Lipids are biodegradable, abundant in nature, and can be excellent substrates for microbial energy-yielding metabolisms (Madigan, et al., 1997). Microorganisms utilize lipids after hydrolysis of the ester bonds, and extracellular enzymes called lipases are responsible for the reactions. The results of the lipase actions are glycerol, mixtures of mono- and diglycerides, and fatty acids (Petrucci, 1989). Glycerol is converted to glyceraldehyde-3-phosphate (triose phosphate) and joins into the glycolysis. With the release of energy, fatty acids are oxidized to carbon dioxide and water in a series of reactions known as  $\beta$ -oxidation. In this process, oxidation occurs at the  $\beta$ -carbon atom of the fatty acid, followed by cleavage. This means that two-carbon pieces (acetic acid) are split off. The process requires the presence of coenzyme A (Petrucci, 1989).

### **Objective** and scope

The objective of the study was to isolate an appropriate microbial culture for the inoculation of a biofilter that treats waste gases from a fast-food restaurant who uses rape-seed oil in its frying process, as well as identifying appropriate environmental conditions for this culture. The study was conducted in two experimental phases. In the first phase the growth of three different bacterial populations was compared, originating from activated sewage sludge, horse manure, and rockwool filter medium from an existing biofilter. The effects on the bacterial growth by adding a salt medium and/or rape-seed oil as a carbon and energy source, and the presence of new rockwool were also evaluated. In the second phase, the growth of one mixed culture from a rockwool biofilter medium was compared to a mono culture of *Bacillus*, using two different carbon- and energy substrates.

### MATERIALS AND METHODS

### **Experimental design**

The growth of different bacterial cultures was studied in batch laboratory experiments performed in glass flasks. All the experiments were carried out at room temperature (21°C +/-2°C), and the flasks were placed on a shaking table (140 rpm, Lab-Shaker, Adolf-Küner AG Basel, Switzerland) for continuous oxygen supply.

### Phase one

Samples of microorganisms were taken from three different locations: a pilot rockwool biofilter treating exhaust gas from a fast-food restaurant (RW), activated sludge from a wastewater treatment plant (AS), and horse manure (HM). The original water content of the three samples was 23% for RW, 98% for AS and 81% for HM. Approximately 5 ml of AS or 5 g of HM or RW was added to 100 ml flasks containing 50 ml of 0.9% NaCl (pH 6.75). The flasks were left overnight on a shaking table and were, thereafter, used to inoculate the suspensions of phase one, which consisted of 26-100 ml flasks. The complete experimental design is shown in Table I. The salt medium used is specified in Table II (first phase). The rape-seed oil was taken from a fryer at the restaurant where the pilot biofilter was situated (Table III, first phase). Excess oil was added to simulate the conditions found in the pilot rockwool biofilter mentioned earlier, where large amounts of grease were deposited.

**Table I.** Experimental design, first phase. 26-100 ml flasks were monitored for a total of 77 days.

Flask no	Inoculum (1)	Liquid (2)	Rape- seed oil	New rockwool	Flask no	Inoculum (1)	Liquid (2)	Rape- seed oil	New rockwool
1	RW	NaCl	No	No	15	No	Salt medium	No	Yes
2	AS	NaCl	No	No	16	RW	Salt medium	No	Yes
3	HM	NaCl	No	No	17	AS	Salt medium	No	Yes
					18	HM	Salt medium	No	Yes
4	RW	Salt medium	No	No					
5	AS	Salt medium	No	No	19	No	NaCl	Yes	Yes
6	HM	Salt medium	No	No	20	RW	NaCl	Yes	Yes
					21	AS	NaCl	Yes	Yes
7	No	NaCl	No	Yes	22	HM	NaCl	Yes	Yes
8	RW	NaCl	No	Yes					
9	AS	NaCl	No	Yes	23	No	Salt medium	Yes	No
10	HM	NaCl	No	Yes	24	RW	Salt medium	Yes	No
					25	AS	Salt medium	Yes	No
11	No	NaCl	Yes	No	26	HM	Salt medium	Yes	No
12	RW	NaCl	Yes	No					
13	AS	NaCl	Yes	No					
14	HM	NaCl	Yes	No					
$^{(1)}$ 0.1 $^{n}$	nl (.	<sup>2)</sup> 20 ml	<sup>(3)</sup> 1 g	<sup>(4)</sup> 0.5	g				

**Table II.** Salt medium used in the two phases of experiments.

Constituent	First phase (g/l)	Second phase (g/l)
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	0,5	1,0
$NaNO_3$	0,5	0,5
$KH_2PO_4$	1,0	1,0
$NaH_2PO_4 * H_2O$	0,5	0,5
$Mg SO_4 * 7 H_2O$	0,42	0,42
$CaCl_2 * 2 H_2O$	0,026	0,026
NaOH	8	8
рН	6,8	7,5

**Table III.** Carbon and energy substrate used in the two phases of experiments.

Constituent	First phase (g/l)	Second phase (g/l)
Fryer oil (rape-seed oil),	50	2
taken from the restaurant fryer with pilot biofilter Filter bed fat,	0	2.
extracted with hexane from pilot biofilter	Ů	-

A scale-up experiment was performed on day 34 using inocula from flasks numbered 24 (RW), 25 (AS), and 26 (HM). 100 ml of salt medium (Table II, first phase) and 5 g of fryer oil (Table III, first phase) were added, together with inocula, to 500 ml flasks to get a start concentration of approximately 1\*10<sup>5</sup> CFU/ml (colony-forming units per ml). This experiment was performed in duplicates of RW, AS, and HM; six flasks were prepared and monitored for a total of 100 days.

### Phase two

The composition of the salt medium was changed in the second phase of the experiments to contain more nitrogen in the form of (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> (Table II, second phase), while the amount of carbon and energy substrate was decreased to 2 g/l (Table III, second phase). The experimental design consisted of 14-100 ml flasks and is shown in Table IV. Flasks 1 to 12 were triplicates and flasks 13 and 14 were duplicates with fryer oil added in excess similar to the experimental conditions in phase one (50 g rape-seed oil/l). 20 ml of salt medium (Table II, second phase) was added, together with inocula from the 500 ml flasks in phase one, to all flasks to obtain start concentrations of approximately 1\*10<sup>5</sup> CFU/ml. The carbon and energy substrate was added in two different forms to verify how they would affect pH and if the bacteria could degrade them equally well. The rape-seed oil, taken directly from the restaurant fryer (from now on called fryer oil), was compared to grease extracted with hexane from the filter bed in the pilot biofilter (from now on called *filter bed fat*). Two different inocula were also compared in this study: the RW mixed culture in the 500 ml flask from phase one (from now on called mixed culture), and a mono culture of Bacillus (from now on called mono culture). The latter one was chosen because many bacilli produce extracellular hydrolytic enzymes that break down e.g. lipids, permitting the organisms to use these products as carbon sources and electron donors (Madigan et al., 1997).

**Table IV.** Experimental design, second phase. 14 shake flasks (à 100 ml) were monitored for a total of 45 days.

Flask no	Inoculum	Carbon- and energy substrate
1-3	Mono culture	Fryer oil
4-6	Mono culture	Filter bed fat
7-9	Mixed culture	Fryer oil
10-12	Mixed culture	Filter bed fat
13-14	Mixed culture	Fryer oil in excess

### Measurements and analyses

### Viable count

Bacterial cell counts were performed with the viable count method to evaluate growth. Samples of 0.1 ml were, after a series of dilutions in 0.9% NaCl, spread over the surfaces of nutrient agar plates (granulated agar, 12 g/l, no 1.01614, Merck, Germany, mixed with nutrient broth, 8 g/l, no 1.05443, Merck, Germany). The plates were then incubated for three days at room temperature (21°C +/-2°C) and the number of colonies were counted. Counts between 20 and 300 colonies per plate were considered significant, and dilutions from 10 to  $10^8$  were performed to obtain the appropriate number of colonies. The results are expressed as CFU/ml (colony-forming units per ml).

### pH

The pH values in the flasks were measured using a pH electrode dipped into the flasks and rinsed with 70% ethanol before and between measurements to avoid contamination.

### COD and NH<sub>4</sub>-N

Chemical Oxygen Demand ( $COD_{Cr}$ ) were determined according to Swedish standard SS 02 81 42 and the ammoniacal nitrogen ( $NH_4$ -N) was measured by using an autoanalyser TRAACS 800 (Bran+Lubbe). The  $COD_{Cr}$  and  $NH_4$ -N content in each flask from the second phase were determined at the end of the experimental period, after filtration to remove large particles.

### Microbial characterization

Besides Gram-staining and test for the ability of form endospores, a few biochemical tests were performed to characterize the cultures (Madigan et al., 1997): catalase, carbohydrate fermentation, oxidation/fermentation test, nitrate reduction, citrate utilisation, indole test, hydrogen sulphide production, and test for motility.

### **RESULTS AND DISCUSSION**

#### Phase one

Viable count and pH measurement results from the initial 26 flasks are presented in Table V. The experimental design can be found in Table I. It was concluded that the rockwool material did not inhibit the growth of the bacterial cultures (compare flasks 1-3 with 8-10, flasks 4-6 with 16-18, and flasks 12-14 with 20-22) and that the presence of new rockwool seemed to have a pH rising effect. The pH decreased more in flasks where rape-seed oil had been added (compare flasks 1-3 with 12-14 and 20-22, and flasks 4-6 with 24-26), but the presence of rockwool seemed to tone down this effect (flasks 20-22). This could indicate some buffering capacity of the rockwool filter media, which could prevent acidification of a future biofilter. In flasks 7, 11, 15, 19, and 23, where no inoculum had been added, no growth was detected except in flask number 15. In flasks where inoculum, but neither carbon source nor salt medium was added (flasks 1-3 and 8-10), only nutrients supplied by the inoculum were available to the bacteria. This could explain the initial growth followed by a subsequent decrease of cell numbers.

**Table V.** Viable count and pH measurements from bacterial suspensions in 100 ml flasks, first phase.

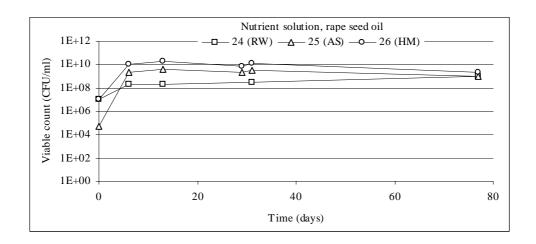
Day:	0	6	13	29	31	77	0	17	29	72
Flask	CFU/ml	CFU/ml	CFU/ml	CFU/ml	CFU/ml	CFU/ml	pН	pН	pН	pН
no	_	_	_			_				
1	$1*10^{7}$	$1*10^{8}$	$6*10^{7}$			$1*10^{7}$	6,7	5,9		7,3
2	$5*10^4$	$3*10^{6}$	$5*10^{6}$			$1*10^3$	6,7	5,9		7,2
3	$1*10^{7}$	$1*10^{7}$	$6*10^6$				6,7	6,0		7,3
	7	7	7	7		6				
4	1*10 <sup>7</sup>	2*10 <sup>7</sup>	2*107	1*10		9*10 <sup>6</sup>	6,8	6,9		6,5
5	$5*10^4$	6*10 <sup>4</sup>	nd				6,8	6,9		4,7
6	$1*10^{7}$	$2*10^6$	$1*10^{6}$				6,8	6,9		6,9
7	nd	nd	nd				6,7	7,1		8,0
8	1*10 <sup>7</sup>	1*10 <sup>8</sup>	6*10 <sup>7</sup>			$1*10^6$	6,7	7,1		7,6
9	5*10 <sup>4</sup>	6*10 <sup>6</sup>	6*10 <sup>6</sup>			nd	6,7	7,3		7,0
10	$1*10^{7}$	1*10 <sup>7</sup>	$2*10^7$			2*10 <sup>5</sup>	6,7	7,5		8,7
10	1*10	1*10	2*10			2*10	0,7	7,0		0,7
11	nd	nd	nd				6,7	6,3	5,1	5,6
12	$1*10^{7}$	$9*10^{7}$	$7*10^{7}$	$7*10^{6}$		$1*10^{6}$	6,7	7,0	5,0	4,8
13	5*10 <sup>4</sup>	$1*10^{3}$	nd	$1*10^{2}$		nd	6,7	6,4	4,9	4,7
14	$1*10^{7}$	$4*10^{7}$	8*10 <sup>7</sup>	$2*10^{7}$		$2*10^6$	6,7	5,3	5.0	4,8
							,	,	,	ĺ
15	nd	$7*10^{5}$	$2*10^{7}$				6,8	6,3		6,1
16	$1*10^{7}$	$3*10^{7}$	9*10 <sup>7</sup>			$4*10^{7}$	6,8	6,9		6,9
17	$5*10^4$	$1*10^{6}$	nd			nd	6,8	7,0		5,0
18	$1*10^{7}$	$7*10^{6}$	$1*10^{7}$			nd	6,8	7,0		6,0
19	nd	nd	nd			7	6,7	7,3		6,0
20	$1*10^{7}$	$2*10^{8}$	$6*10^{7}$			$1*10^{7}$	6,7	6,7		5,9
21	$5*10^{4}$	$4*10^{7}_{-}$	$7*10^{7}$			$1*10^6$	6,7	6,6		6,0
22	$1*10^{7}$	$4*10^{7}$	$1*10^{8}$			$8*10^6$	6,7	6,9		6,1
22	1						<i>(</i> 0	7.0	<i>5</i> 2	15
23	nd	nd	nd		2*108	1 * 1 0 9	6,8	7,8	5,3	4,5
24	1*10 <sup>7</sup>	2*10 <sup>8</sup>	2*10 <sup>8</sup>	2*109	3*10 <sup>8</sup>	1*109	6,8	6,9	6,3	5,2
25	5*10 <sup>4</sup>	2*10 <sup>9</sup>	4*10 <sup>9</sup>	2*10 <sup>9</sup>	3*10 <sup>9</sup>	1*109	6,8	5,9	5,6	6,3
26	$1*10^{7}$	$1*10^{10}$	2*10 <sup>10</sup>	7*10 <sup>9</sup>	1.3*10 <sup>10</sup>	2*10 <sup>9</sup>	6,8	6,0	6,0	6,2

nd: non detectable

It was obvious when the colony morphologies were studied at each plate count that the number of different bacterial species had decreased with time and that a natural selection of bacteria able to survive in the batch environment had developed. During the course of the experiment, notes were taken of how the suspensions changed with time and at the end (after 77 days), the following was observed: flasks 1-10 and 15-18: transparent liquids, no visible signs of growth; flasks 12-14: slightly turbid suspensions, oil floating on the surfaces; flasks 20-22: white and turbid liquids with visible growth in the suspensions, yeast and mould grew on the surfaces. Thick and "milky" homogenous suspensions were observed in flasks 24-26. This could be due to the bacterial production of surface-active substances, which transform the oil into an emulsion, and makes the oil readily available for the microorganisms.

### Growth and pH measurements in flasks 24 (RW), 25 (AS), and 26 (HM)

It was evident that the presence of both the salt medium and rape-seed oil contributed to the highest growth of bacteria, as was the case in flasks 24, 25, and 26. The growth in these suspensions is presented in Figure 1. This indicates that a salt medium must be added in a future rockwool biofilter, in addition to the carbon and energy substrate provided by the gas stream, to offer essential nutrients e.g. nitrogen and phosphorous for the bacteria.



**Figure 1.** Growth of bacterial cells in 100 ml flasks 24, 25, and 26 from phase one.

A difference in growth between the three different bacterial populations (RW, AS, and HM) was observed. The HM flask (no 26) contained the highest amount of bacteria (2\*10<sup>10</sup> CFU/ml on day 13), followed by the AS flask (no 25). A rapid exponential growth occurred in these two flasks the first 6 days with an increase of cell numbers by a factor of 10<sup>3</sup> (HM, no 26) and 10<sup>5</sup> (AS, no 25). Generation or doubling times calculated for the two cultures during the exponential phase (day 0-6) was 14 hours in flask 26 and 9 hours for flask 25. These numbers are probably overestimating the real generation times since an initial lag-phase may be included, but the values could serve well for comparative purposes. After 6-10 days, the growth reached a stationary phase. The pH (Table V) in flasks 25 and 26 had decreased approximately one unit on days 17 and 29, but had increased again on day 72.

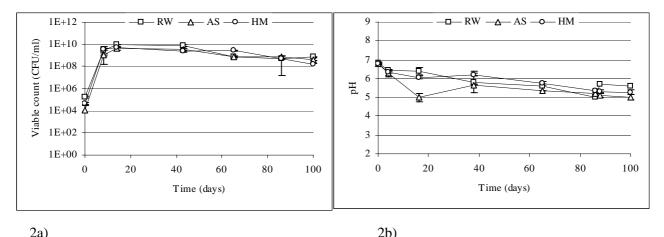
The growth rate was considerably slower in flask 24 (RW) than in flasks 25 and 26; the generation time calculated from start to day 6 was about 33 hours. However, the cell numbers continued to increase throughout the experimental period, parallel to a pH decrease (from 6.8 to 5.2 in 72 days). On day 77 the three cultures (flasks 24, 25, and 26) all contained approximately 10<sup>9</sup> CFU/ml.

When comparing the three bacterial cultures, it is important to remember that they originated from completely different environmental conditions and could have brought different nutrients to the system. The rockwool inoculum (RW) came from a very selective environment in a biofilter with low water content and substrate merely from a frying process. The activated sludge (AS) and horse manure (HM) were probably composed of a greater variety of microorganisms that had been living in a richer environment, and these inocula probably brought more nutrients to the cultures.

### Upscale of RW, AS and HM cultures: growth and pH measurements

The upscale gave further insight into the growth of the three different microbial populations. In Figure 2, the growth (2a) and pH (2b) for the three cultures are presented as mean values (of two) with standard deviations. Unfortunately, one of the RW flasks broke on day 20 and as a result only one value was available for RW after that day. A rapid exponential growth occurred in all flasks the first 8 days with generation times ranging from 11 to 14 hours when cell numbers increased by a factor of 10<sup>5</sup>. The exponential growth phase was followed by slower growth and a stationary phase after which cell numbers started to decrease (Figure 2a). A pH decrease was observed in all flasks and the pH went from 7 to between 5 and 6 at the end of the experimental period (figure 2b).

Apart from the "pH dip" in the two AS flasks on day 16, the six cultures seemed to behave identically and be well adapted to use the rape-seed oil as their sole carbon and energy source. By studying the colony morphologies it was concluded that the number of different bacterial species was four or five, with similar colonies in the three different enrichment cultures. The appearance of the batch suspensions changed during the experimental period from a transparent liquid with oil floating on the surfaces to a thick, yellow-white suspension after 15-20 days that remained in this form for the rest of the experimental period.



**Figure 2.** Growth and pH in 500 ml flasks from phase one. Mean values (of two) with standard deviations.

### **Growth limitations in phase one**

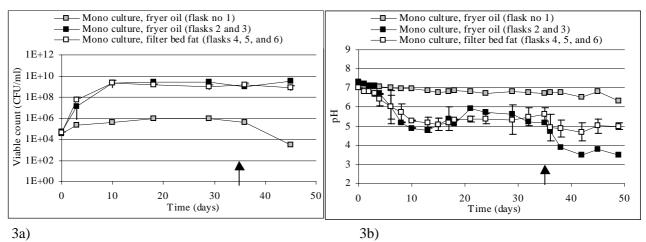
Metabolic activities of the growing microbial population could have changed the nature of the environment to the point where it became unfavourable. This may have been brought about by the decreasing pH, by the depletion of nutrients and/or oxygen, or by the accumulation of toxic metabolites. The bacterial populations may have multiplied by hydrolysing the rapeseed oil and preferentially using the glycerol part of the oil as their carbon and energy source, but may have left the fatty acids unmetabolised. This could then explain the decreasing pH in the flasks. The fatty acids could have become growth inhibitory or toxic if the concentrations were too high. As long as rape-seed oil was supplied in abundance, the microbial population was never "forced" to use the fatty acids as their carbon and energy source; which would mean that only about 5% of the carbon in the rape-seed oil was utilized. If this would be the case, it might become difficult to obtain complete degradation of the oil in a biofilter with large amounts of grease collected. Therefore, it was decided to decrease the amount of oil in phase two, to assure that this principal carbon source would be the limiting substrate for the bacteria. Another possible limitation might have been the lack of a nitrogen source in the system. When calculating the C:N:P ratio in the added salt medium 100:0.5:1.2 is obtained, which could be compared to a recommended ratio of approximately 100:5:1 (Biddlestone and Gray, 1985). Therefore, it was decided that the amount of nitrogen in the salt medium should be increased in phase two and in the form of ammonium rather than nitrates since the conversion of ammonia is less energy consuming for the bacteria (Stanier et al., 1976).

### Phase two

The growth and pH curves for the two cultures, the mono culture (*Bacillus*) in Figure 3 and the mixed culture (*RW*) in Figure 4, are presented as mean values (of three) with standard deviations. The growth in flask no 1 (mono culture, fryer oil) differed significantly from the growth of other two in that same group (flasks 2 and 3), probably due to poor condition of the inoculum, hence, the results for flask no 1 are presented separately (Figure 3).

### Growth and pH measurements of the mono culture (Bacillus)

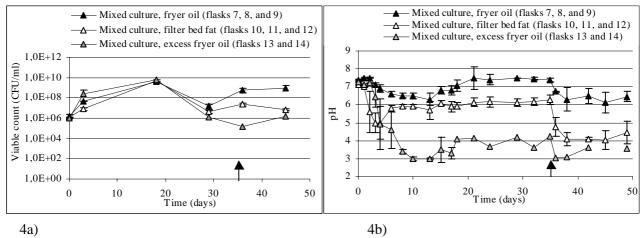
The mono culture showed an exponential growth the first 3 days (Figure 3a) with an increase of cell numbers by a factor of 10<sup>3</sup>, except for flask no 1. Generation times calculated for these three days were 26 hours for flask 1, 9 hours for flasks 2-3, and 7 hours for flasks 4-6. Cell numbers continued to increase between day 3 and 10 in flasks 2-6, followed by an approximate 20-day long stationary phase. pH (Figure 3b) decreased about 0.5 unit in flasks 2-6 during the exponential growth phase (day 0-3); however, between days 3 and 10, pH dropped drastically about two units from 7 to 5, probably due to the accumulation of secondary metabolites. Yet, this did not seem to influence the number of cells, which suggests that the bacteria were rather tolerant to pH changes. Thereafter, pH was fairly stable between 5 and 5.5 during the stationary phase. pH in flask no 1 stayed rather constant around 7 throughout the experimental period, indicating low metabolic activity of these bacteria. No significant differences could be established between the two carbon and energy sources, fryer oil and filter bed fat, as it seemed that the mono culture could degrade both equally well. To see if the stationary growth was due to a lack of carbon substrate, 0.04 g of fryer oil or filter bed fat was added to the flasks on day 35 (indicated by an arrow in Figure 3a and b). This addition caused a nominal increase of cell numbers in the flasks with fryer oil (2-3), but had no visible effect in the flasks with filter bed fat (4-6). The addition of fryer oil caused pH to drop 1-1.5 units to 3.5 in flasks 2 and 3, while the addition of filter bed fat had a more modest effect in flasks 4-6, where pH declined about 0.5 unit to 4.7 (Figure 3b). The appearance of the suspensions in the flasks changed during the experimental period from transparent liquids with oil floating on the surfaces to thick, yellow-white suspensions after 10-15 days and then back to transparent yellow liquids with white particles in suspension around day 35. When examining these particles in a microscope, it was concluded that it was most likely fungi probably favoured by the acid conditions prevailing in these flasks.



**Figure 3.** Growth (3a) and pH (3b) in 100 ml flasks from phase two. Mean values with standard deviations. Comparison of two carbon and energy substrates: fryer oil and filter bed fat for the mono culture (*Bacillus*). An arrow indicates the addition of 0.04 g fryer oil or filter bed fat.

### Growth and pH measurements of the mixed culture (RW)

The initial cell number of the mixed culture being almost  $10^2$  times higher than the initial cell number of the mono culture could explain that the increase of cell numbers was lower in flasks 7-12, as compared to the increase in flasks 2-6 (compare figure 3a with Figure 4a). However, the final maximum number of bacteria obtained was about the same for the two cultures (around  $10^9$  CFU/ml), probably set by the supply of nutrients. The increase of cell numbers in flasks 13 and 14 during the first 3 days was of the same order of magnitude as in flasks 2-6 (a factor of  $10^3$ ). Generation times for the exponential growth from days 0 to 3 were 14 hours for flasks 7-9, 26 hours for flasks 10-12, and 9 hours for flasks 13-14. Unfortunately, the dilutions on day 10 failed to give a countable number of cells, therefore, no results were obtained for the period between days 3 and 18. pH (Figure 4b) decreased about one unit in flasks 7-9 and 10-12 from start to day 10, while the drop of pH was more drastic in flasks 13 and 14 where pH decreased from 7.3 to 3.0 during the first 10 days. This did not seem to influence the number of cells, which again points to a high bacterial tolerance to acid conditions.

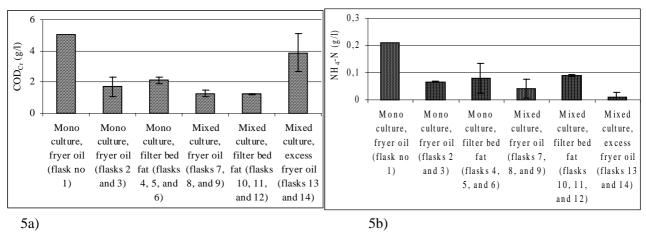


**Figure 4.** Growth (4a) and pH (4b) in 100 ml flasks from phase two. Mean values with standard deviations. Comparison of two carbon and energy substrates: fryer oil and filter bed fat for the mixed culture (*RW*). An arrow indicates the addition of 0.04 g fryer oil or filter bed fat, or 20 ml extra salt medium in the case of *mono culture*, *excess fryer oil*.

Cell numbers decreased between days 18 and 29, for that reason, fryer oil, filter bed fat, or salt medium was added on day 35 (indicated by an arrow in Figure 4a and b). This led to a drastic drop in pH (Figure 4b) of about two units to pH 4 in the filter bed fat flasks (10-12) and a more modest decrease of about one unit in the fryer oil flasks (7-9 and 13-14). The addition of extra nutrients seemed to stimulate the growth in all flasks initially, but then the cell numbers either stabilized (flasks 7-9 and 13-14) or decreased (flasks 10-12), possibly due to growth inhibition in connection with the pH changes. It seemed that the mixed culture preferred the fryer oil substrate to the filter bed fat substrate, the initial growth being slightly faster in flasks 7-9 and 13-14 compared to that in flasks 10-12. The addition of extra fryer oil also generated an increase of bacterial cells in flasks 7-9 and 13-14, which was not the case when extra filter bed fat was added to flasks 10-12. This could probably be due to the initial hydrolysation of the fryer oil, which generated easy degradable glycerol, compared to the possibly more complex secondary oxidation products in the filter bed fat. Also, the mixed culture had been enriched on fryer oil (in phase one) and, therefore, could have developed a preference towards this carbon and energy substrate. All the batch suspensions were transparent white-yellow with white particles in suspension throughout the experimental period, which could be the sign of fungi growing in the suspensions.

### COD<sub>Cr</sub> and NH<sub>4</sub>-N analyses

The COD<sub>Cr</sub> and NH<sub>4</sub>-N content in each of the 14 flasks were determined at the end of the experimental period in order to get an idea of the organic degradation and the oxidation reactions that had occurred in the flasks, and to also give an indication if one of the two carbon substrates were more easily oxidized than the other. Results from these analyses are presented in Figure 5, as mean values with standard deviations.



**Figure 5**. COD<sub>Cr</sub> (5a) and NH<sub>4</sub>-N (5b) analyses performed on day 50, 100 ml batch flasks from second phase. Mean values with standard deviations.

The values in Figure 5a and b can be compared to the COD values of the added carbon substrates and to the amounts of NH<sub>4</sub>-N added to the flasks during the experiment. To flasks 1-12, a total of 4 g/l of fryer oil or filter bed fat, corresponding to an approximate COD value of 11.5 g/l, and 0.21 g NH<sub>4</sub>-N g/l were added. To flasks 13 and 14, 50 g/l of rape-seed oil, corresponding to a COD value of approximately 144 g/l, and 0.42 g NH<sub>4</sub>-N g/l were added. In Table VI, an estimation of how much COD and ammonia-nitrogen that remained in the flasks after 50 days are given. The values are calculated as the measured COD<sub>Cr</sub> respective NH<sub>4</sub>-N value divided by the added amounts of COD and NH<sub>4</sub>-N.

**Table VI.** Estimation of the remaining COD and ammonia-nitrogen (in % of the added amounts) in the 100 ml flasks from second phase. Values presented are mean values with standard deviations.

Flask no	COD (%)	Ammonia-nitrogen (%)
1 (mono culture, fryer oil)	44	99
2-3 (mono culture, fryer oil)	$15 \pm 5$	$31 \pm 1$
4-6 (mono culture, filter bed fat)	$18 \pm 2$	$37 \pm 26$
7-9 (mixed culture, fryer oil)	$11 \pm 2$	$20 \pm 16$
10-12 (mixed culture, filter bed fat)	$11 \pm 0.5$	$42 \pm 1$
13-14 (mixed culture, excess fryer oil)	$3\pm1$	$3\pm4$

Flask no 1 (mono culture, fryer oil) stands out from the others because its activity had been significantly lower since almost all of the ammonia-nitrogen remained. This is in accordance to what was earlier suspected that somehow the inoculum in this flask had not been able to multiply and degrade in the same manner as in the other flasks. The disappearance of more than half of the COD in this flask, despite low bacterial activity, indicates that the analytical filter used in the  $COD_{Cr}$  analysis caught a large part of the oil. This must thus be taken into consideration when interpreting the COD estimates. In flasks 13 and 14, with an excess of fryer oil, the  $COD_{Cr}$  value was higher than in the other flasks (Figure 5) which is logical since more oil was initially added to these two flasks. The degradation had been high in these flasks; almost all of the ammonia-nitrogen and a large part of the COD were consumed after

50 days (Table VI). The COD values in flasks with fryer oil did not significantly differ from those with filter bed fat; thus, confirming that no large difference could be seen of the microbial ability to use the two carbon substrates. The mixed culture (*RW*) seemed to have been slightly more efficient than the mono culture (*Bacillus*) since the remaining COD and NH<sub>4</sub>-N were lower in flasks 7-12.

### **Growth limitations in phase two**

It is probable that the growth was limited by something other than lack of nutrients, at least in flasks 2-12 since both ammonia-nitrogen and COD remained after 50 days. Waste products could have built up to an inhibitory level, causing pH to decrease, and thereby ceasing growth by an inhibition of important lipase enzymes. However, cell numbers remained high even when pH dropped to about 3, indicating that the bacteria were tolerant also to acid conditions. When trying to correlate the pH and viable count number obtained, low correlations were found (data not shown).

### Estimation of the carbon utilization in phase one and two

A simplified calculation of the maximal number of bacteria (per litre) that theoretically could be attained with the carbon present in the system and no limitations could be compared to the highest number of cells experimentally obtained with the viable count method in phase one and two. The following assumptions were made:

- Rape-seed oil (C<sub>57</sub>H<sub>101</sub>O<sub>6</sub>) has a mole weight of 881 g/mole
- In aerobic respiration, about 1/3 of the substrate carbon is incorporated into cells (and thus available for growth) and about 2/3 are converted to CO<sub>2</sub> (Mathur, 1991)
- The weight of one bacteria is approximately  $10^{-12}$  g (wet weight), the bacteria consists of 90% water and the carbon content of a bacteria (on a dry-weight basis) is 50% (Madigan et al., 1997). One bacteria would then contain approximately  $5*10^{-14}$  g carbon.

Phase one: 50 g rape-seed oil/l equals approximately 39 g C/l. If 33% of this is used for new cells (13 g C/l), then the maximal theoretical number (no limitations) of bacteria would be 3\*10<sup>14</sup> bacteria/l. The highest measured experimental value obtained in phase one was 2\*10<sup>13</sup> CFU/l (flask 26, HM). In other words, about 7% of the available carbon was used for growth.

Phase two: In phase two, flasks 13 and 14 received the same amount of carbon as in phase one, the highest measured experimental value in these flasks was  $6*10^{12}$  CFU/l. This would mean that only 2% of the available carbon were used for growth. This is in contrast with COD results presented in Table VI, where a large part of the carbon seemed to be consumed after the experimental period. For the rest of the flasks in phase two, 2 g rape-seed oil/l was added (day 0-35) which equals 1.6 g C/l. If 33% of this is used for new cells (0.5 g C/l), then the maximal theoretical number of bacteria would be  $1*10^{13}$  bacteria/l. The highest measured experimental value obtained in phase two was  $5.5*10^{12}$  CFU/l (flask 10-12).

In other words, about 55% of the available carbon were used for growth in these flasks, which is in reasonable agreement with results obtained in Table VI. An ideal metabolic carbon utilization is obviously never possible to obtain, but the calculations could give an idea of how well the bacteria have been able to use the carbon and energy substrate. When the rape-seed oil had been provided in excess, in phase one and in flasks 13-14 in phase two, it seemed as though the bacteria only used a small part of the available carbon. This could indicate that the bacteria used mainly the glycerol and left the fatty acids unmetabolized. When the carbon substrate was supplied in limited quantities, as was the case in phase two, the carbon utilization seemed to improve. The viable count method might underestimate the real number

of bacteria since only culturable, viable cells are detected on the agar plates used. These bacteria might constitute only a part of the total population.

### Characterization of bacteria

A few simple biochemical tests (Table VII) were performed to characterize the most abundant bacterium from the mixed culture (*RW*) and the mono culture (*Bacillus*). Both these two were found to be aerobic rods. The mono culture was gram-positive with the ability to form endospores, while the bacterium from the mixed culture was gram-negative and did not have the ability to form endospores. Both were mesophiles; the bacteria from the mixed culture grew better at 32°C, compared to 21°C and 37°C, while the *Bacillus* culture grew best at 37°C.

**Table VII.** Biochemical test results for the most abundant bacterium in the mixed culture (*RW*) and mono culture (*Bacillus*).

Tests	Mixed culture	Mono culture
Catalase	+	+
Carbohydrate fermentation	-	-
Oxidation/fermentation	-	-
Nitrate reduction	+	-
Citrate utilisation	+	-
Indole	-	-
Hydrogen sulphide production	-	-
Motility	-	-

### **Limitations of the study**

The aim of the study was principally comparative and aimed to find out if the different cultures of bacteria chosen were able to use rape-seed oil and its oxidation products as their sole carbon and energy source. It is important to point out that the conditions for the bacteria in the laboratory batch flasks are much different from the conditions prevailing in a biofilter. The microorganisms in the flasks are suspended and very mobile and the supply of water and substrate is almost unlimited. In a biofilter, on the other hand, the microorganisms will be immobilized on the filter medium with a minimum motility. Furthermore, equilibrium is created in the flasks, where oxygen is first solved in the liquid and thereafter available to the bacteria. This process is more complicated in a biofilter where the microorganisms are probably influenced by the strong airflow. The oxygen will be passing through a thin water film first, and then penetrating into the biofilm. Finally, the risk of product inhibition by volatile metabolites could be greater in the flasks if secondary degradation products are accumulated in the liquid. In a biofilter, the secondary oxidation products could build up to inhibitory levels in the immediate vicinity of the bacteria. However, volatile secondary oxidation products could follow the gas out and an accumulation of these products in the filter could thereby be avoided. Moreover, during the lifetime of a biofilter there will be a development of a microbial flora specialised in the secondary products; hence the final degradation products will be carbon dioxide and water.

#### CONCLUSIONS

The preparation of a proper inoculum is important for a successful biofilter operation and the results of this study gave insight into the basic relationships concerning bacterial growth and generated knowledge for future inoculation of rockwool biofilters for the fast-food industry using rape-seed oil in its frying process. Bacteria from three different environments (from activated sewage sludge, horse manure, and rockwool filter medium from an existing biofilter) were enriched in batch cultures, and it was found that all three cultures were able to use the rape-seed oil as their sole carbon and energy source. For maximum and lasting growth it was necessary to add a salt medium containing e.g. phosphorus and nitrogen. An exponential growth phase of 3 to 8 days was followed by slower growth and a stationary phase of about 40 days. The rockwool material did not inhibit the growth of the cultures and seemed to have a buffering effect, which could prevent acidification of a future biofilter. The addition and degradation of rape-seed oil caused pH to decrease in the batch cultures, but this did not influence the number of cells which suggest that the cultures were tolerant to acid conditions. One of the mixed cultures (from the rockwool filter medium) was further enriched and compared to a mono culture of Bacillus. Biochemical tests showed that the most abundant bacterium from the mixed culture were mesophilic, aerobic, gram-negative rods. Their ability to use the rape-seed oil, as well as its oxidation products, were found to be good. They were, therefore, both considered suitable for inoculation of a biofilter that treats waste gases from a frying process with rape-seed oil. However, since conditions in batch laboratory flasks and in a biofilter differ greatly, this has to be verified in further experiments. Future work implies inoculation of pilot and full-scale biofilters and studies of the microbial establishment.

#### **ACKNOWLEDGEMENT**

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# **PAPER II**

# A study of a rockwool biofilter for the removal of odours, grease aerosols and VOCs

Andersson, A.

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# A study of a rockwool biofilter for the removal of odours, grease aerosols and VOCs

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# **ABSTRACT**

Emissions from food manufacturing tend to contain a mixture of partially oxygenated hydrocarbons whose composition and concentration vary with time. Little research has been carried out on the capacity of biofilters to deal with such effluents. The objective of this study was to investigate the feasibility of a compact rockwool biofilter for a fast-food restaurant to remove grease aerosols, VOCs, and odorous components. This article presents experiences from a full-scale biofilter as well as tests run in a pilot scale multi-stage biofilter.

The full-scale biofilter was in operation for approximately one year. The restaurant staff were generally happy with the filter performance; they perceived that the fatty odours were reduced notably and that the grease aerosols were effectively removed. However, sampling of the filter media revealed that the moisture content and the bacterial numbers were very low. Problems encountered in the full-scale application initiated further pilot-scale studies. Synthetic rockwool fibre mats used as filter material seemed to be an appropriate habitat for microorganisms as the inoculated mixed culture was found in relatively high numbers. A mechanical collector for grease aerosols, installed upstream of the biofilter, proved to be efficient. Moisture content in the filter material varied because of high velocities of air through the filter, which involved drying of the material. After three weeks of operation, samples of aldehydes were taken in the inlet and outlet of the biofilter. No reduction could be established, possibly due to the short residence times. Future work will demand investigations into the design of the filter, the composition of the off-gases, the activities of the bacteria in the filter, and a search for the best method for evaluating the biofilter performance. It can be concluded that the off-gas from a restaurant is a very complex mixture due to the constantly changing structure of the oil.

KEY WORDS

Biofilter, rockwool, food industry, grease, odors, VOC, aldehydes, fatty acids

### **INTRODUCTION**

The food industry is often the target of public complaints due to the odorous gas emissions, especially when situated in densely populated areas, and could lead to expensive litigation or in the worst case eviction. Waste gases that contain volatile organic compounds (VOCs) are facing increasingly stringent environmental regulations all over the world, rising the need for efficient treatment methods. Besides being odorous, VOCs are responsible for the production of pollutants known as photochemical oxidants, principally ozone. These compounds could be toxic to humans, damage crops, and are implicated in the formation of acid rain <sup>1</sup>.

During the last decades, biofiltration for air pollution control has been established as a reliable, cost-effective technology for controlling low-concentration biodegradable waste gases. Biofiltration is not presently a well recognized waste air treatment technique in Sweden but could be applied in a wide range of industries and public sectors including the food industry <sup>2,3</sup>. To treat the off-gases from a restaurant, other abatement techniques like incineration often prove economically impossible. Adsorption on activated carbon or aqueous absorption could be problematic because of the high moisture content of the gas and the low solubility of some of the compounds. Emissions from food and drink manufacturing tend to contain a mixture of partially oxygenated hydrocarbons (i.e. carboxylic acids, aldehydes, ketones, alcohols, and esters) whose composition and concentration vary with time. This poses a special challenge for a biofilter, as it has to work with intermittent loads and deal with changes in the concentration of individual contaminants. Little research has been carried out on the capacity of biofilters to deal with such effluents.

The idea behind a biofilter is to let micro-organisms degrade pollutants from the air and use these substances as their primary carbon and energy source. The key to a successful biofilter operation is to create a healthy ecosystem in the filter, by controlling parameters like moisture content, pH, temperature, access to oxygen, and nutrients. The choice of filter material is fundamental and in recent years various synthetic packing materials have been developed to retard the ageing effects and maintain the bed porosity. Since synthetic materials do not contain micro-organisms or nutrients, these must be added. Rockwool seems to be a suitable biofilter medium since it is cheap, develops low pressure drops, has good mechanical properties, and offers a seemly habitat for micro-organisms <sup>4,5</sup>. It also possesses the advantage of having either hydrophilic or hydrophobic properties, which means good water holding capacity and the ability to sorb hydrophobic compounds. This could be useful in the food industry since the emissions could contain large amounts of grease aerosols. For the restaurant, this often causes problems such as complaints from neighbours, bad working environment in the kitchen (i.e. slippery floors and bad smell), clogging of pipes and ventilation units etc. Considerable energy savings could be made through heat exchange of the air if the grease aerosols were removed from the air. Large depositions of grease could also cause problems in a biofilter with the creation of anaerobic zones and difficulties in maintaining the medium moisture content.

#### Objective and scope

The objective of this study was to investigate the feasibility of a compact rockwool biofilter for a fast-food restaurant to remove grease aerosols, VOCs, and odorous components. The article presents experiences from a full-scale biofilter as well as tests run in a pilot scale multi-stage biofilter.

# **EXPERIMENTAL METHODS**

# **Experimental set-up**

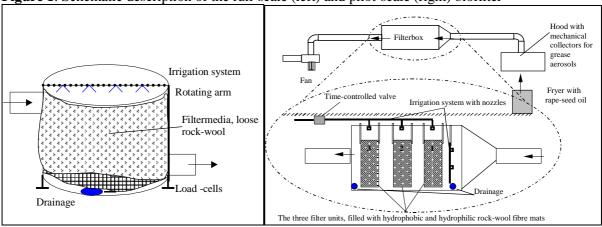
#### Full-scale filter

A Swedish hamburger restaurant installed a full-scale biofilter in 1996, for which operational parameters and a schematic overview are provided in Table 1 and Figure 1. Peat was originally used as filter material, but high grease levels in the air and a degradation of the filter medium caused particles in the outlet to block the subsequent heat exchanger. Also, high pressure-drops rapidly developed. In 1997, the filter medium was replaced with an inert, loose fibre material (rockwool) with a large surface area. The filter, which ran for approximately one year, was inoculated with a specialist culture of *Bacillus*.

**Table 1**. Operational parameters for full- and pilot-scale biofilters

	Full-scale biofilter	Pilot-scale biofilter
Flow (m <sup>3</sup> /h)	3500-1800	400
Filter area (m <sup>2</sup> )	3.2	0.36
Media depth (m)	0.40	3*0.3

Figure 1. Schematic description of the full-scale (left) and pilot-scale (right) biofilter



#### The pilot-scale biofilter

The experiences from the full-scale biofilter initiated further studies on a pilot-scale filter at the University of Luleå, Sweden, for which the operational parameters are described in Table 1. The main design criterion for the pilot filter was a compact, multi-level biofilter, easy to place and handle in a restaurant environment. Therefore, the construction was changed from the full-scale vertical filter with circular area and down flow mode to a horizontal filter with a square area, composed of three filter units operated in a side flow mode, see Figure 1. The experiences from the full-scale filter also led to some changes in design of the irrigation system and in the choice of filter material. A timer-based irrigation system with spray nozzles in the inlet (to saturate the off-gases) and at the top of the biofilter (for direct irrigation) was introduced. Drainage collected at the bottom of the filter was measured and controlled and, in combination with weighing the filter units regularly, the irrigation was adjusted. Fibre mats with pre-set structure and lower densities replaced the loose rockwool used in the full-scale filter to improve flow distribution, facilitate handling, and reduce pressure drops. Rockwool mats with hydrophobic properties were used in thin layers in between thicker hydrophilic mats to improve the distribution of water and hydrophobic compounds in the filter. A mechanical collector for grease aerosols was installed upstream of the biofilter, right above the fryer. The experimental period lasted for 28 days, from November 23 to December 22, 1999. The fryer was operated 7-8 h/day, 5 days/week when potatoes were fried in vegetable oil (rape-seed) heated to 180°C. The designed flow in the pilot-scale filter (400 m³/h) was not quite enough to remove all the emissions from the fryer, so the surrounding air in the room also had a fatty smell. Therefore, the fan and the irrigation were constantly left on, except for one occasion (on day 27). The fibre mats were inoculated with both a mixed culture selected from a wastewater treatment plant, previously enriched with rape-seed oil as the only carbon source, and a specialist culture of *Bacillus* previously used in the full-scale application. A massive sampling programme was performed during the four weeks of test runs.

# Sampling and analyses

# Flows and pressure drops

Air velocities in the channel before and after the biofilter were determined using a *SwemaAir 30* (SWEMA, Farsta, Sweden), and the flow was calculated from the average velocity. Pressure drops over the biofilter were measured with a *SwemaMan 2000* (SWEMA, Farsta, Sweden).

# **Temperature**

The temperature of the gas-stream to and from the biofilter was measured manually with a mercury thermometer. In the pilot-scale, temperatures were registered twice an hour with *Orion Tinytalk 1* loggers (Gemini data loggers, INTAB, Sweden), placed in the surrounding room, in the lid of the biofilter construction, and in the middle of each filter unit.

#### Nutrient solution added to the biofilter

At the start of the pilot-scale biofilter, a nutrient solution (see below) was spread, together with the bacteria, over the filter material to attain around 40% moisture content. 500 ml of nutrient solution (no bacteria) was also added one time during the operation of the filter (on day 23).

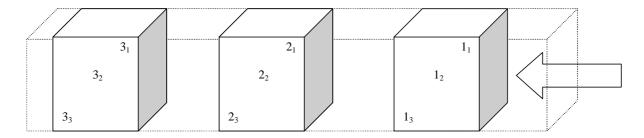
# Nutrient solution

 $0.25 g (NH_4)_2SO_4$ ,  $0.25 g NaNO_3$ ,  $0.013 g CaCl_2 * 2 H_2O$ ,  $0.21 g MgSO_4 * 7 H_2O$ ,  $0.5 g KH_2PO_4$ ,  $0.5 g NaH_2PO_4 * H_2O$ ,  $500 ml H_2O$  and 20 ml 0.2 M NaOH.

# Medium sampling

Sampling of the filter media was performed after 10 months from the full-scale biofilter and regularly from the pilot-scale filter (eight occasions during the experimental period). Filter media from the pilot-filter were taken from nine different sampling points, see Figure 2. Moisture content and ignition residue was determined according to the Swedish standard SS 02 81 13. Microbial cell enumeration was performed using an extraction technique and plating method (viable count). Approximately 1-2 grams, dry weight, of filter media were added to a dilution bottle containing 50 ml of sterile 0.9% NaCl, and then left for one hour on a shaking table (140 rpm, Lab-Shaker, Adolf-Küner AG Basel, Switzerland). A series of dilutions (in sterile 0.9% NaCl) were prepared from this and a volume of 0.1 ml sample was spread over the surface of a nutrient agar plate. The plate was then incubated for 2 days at room temperature (21°C +/-2°C) and the number of colonies were counted. Counts between 20 and 300 colonies per plate were considered significant. The results are expressed as CFU/gram dry filter media (CFU: colony-forming units).

**Figure 2**. Sampling points in the filter units (pilot-scale biofilter).  $1_1$  top and front of the first filter unit (hydrophobic media),  $1_2$  in the middle (hydrophilic media), and  $1_3$  in the back and lower part (hydrophilic media).  $2_1$ ,  $2_2$ ,  $2_3$  and  $3_1$ ,  $3_2$ ,  $3_3$  were all hydrophilic media



#### pH

pH of the collected drainage, as well as in the dilution bottles with media and NaCl (from viable count) were measured using a pH electrode dipped into the bottles, rinsing with 70% ethanol between measurements to avoid contamination.

# Gas sampling

Measurements with a photoionization detector (PID) in the off-gases of the pilot-scale filter failed due to the low concentrations (below the detection of the instrument). Air samples were taken with low-flow pumps and sorbent sample tubes, in the channel before and after the biofilter.

#### **Fatty acids**

Charcoal and XAD-2 tubes were used for the determination of fatty acids, stored at  $-18^{\circ}$ C until analysis. They were derived into alkyl esters <sup>6</sup> and analysed using a gas chromatograph (HP 5890) coupled to a flame ionization detector (FID). A standard solution composed of eleven alkyl esters was used (Mixture FO 7, 90-1107, LOT NO: 6071:2, Larodan Fine Chemicals).

#### **TVOC**

For identification of VOCs, tenax sorbent tubes were used and analysed using a gas chromatograph (HP 6890) with a mass-selective detector and a Perkin Elmer ATD 400, coupled to a mass spectrometer.

#### Aldehydes

For identification of aldehydes, Waters Sep Pak XPosure\* was used and analysed with a TPS HPLC (high-performance liquid chromatograph) with a DAD SM 5000 detector. The aldehyde analyses have been performed with a standard solution consisting of: formaldehyde, acetaldehyde, acetone, acroleine, propanale, crotonaldehyde, butanale, bensaldehyde, pentanale, hexanale, heptanale, octanale, nonanale and decanale.

#### RESULTS AND DISCUSSION

# **Experiences from the full-scale biofilter**

The gas temperature at the inlet and outlet of the biofilter was 31°C (±1°C). The initial flow through the rockwool filter was 3500 m³/h, thus creating an average surface loading of about 1000 m³/m²\*h and an average empty bed residence time (EBCT) of about 11 seconds. The initial pressure drop was approximately 1400 Pa (3500 Pa/m). After 10 months of operation the flow had decreased to 1800 m³/h, and the pressure drop was 2200 Pa (5500 Pa/m). These values may be compared to values obtained when peat was used as filter material: then, the flow was down to 575 m³/h and the pressure drop was 2900 Pa (7250 Pa/m) after 10 months of operation. The staff perceived that the filter effectively removed fat, which made heat exchanging of the air possible, and led to energy savings. Reductions of the odourous fatty smells were also observed and the restaurant was generally happy with the filter performance. However, with time, the decrease in flow caused problems in the kitchen since odorous and fatty emissions remained there. This led to a more detailed examination of the rockwool filter media after 10 months.

The conditions in the filter seemed to be very heterogeneous; a thick layer of grease on top of the filter material obstructed irrigation directly and also indirectly by disturbing the function of the load cells in the bottom of the filter. These were programmed to start the rotating irrigation arm at a certain weight loss (due to evaporation and drying), but this rarely occurred since grease accumulated in the filter, thus increasing the total weight. Sampling of the filter media at different levels showed that the moisture content was very low (less than 5%), except in one sample (70%), and that the grease accumulated exceeded 50% (calculated from the ignition residue) in some samples. pH in the filter media was around 4, and viable count on agar plates showed less than  $1*10^2$  bacteria in the first two samples and  $1*10^7$  bacteria in the third sample (the one with higher moisture content). No bacteria from the Bacillus culture originally inoculated in the filter were found. Results of parallel samplings of fatty acids in the air before and after the biofilter on two occasions showed a large spread both in composition (number of different acids found) and total content, see Table 2. The total fatty acid content was significantly higher at the first sampling occasion when the rape-seed oil in the fryers was older, compared to the second one when the oil had just been changed. No significant reduction of fatty acids over the biofilter could be shown.

**Table 2**. Concentrations of fatty acids found in the gas-stream before and after the full-scale biofilter, average values with standard deviations. First sampling was performed after seven months of operation, the second after ten months

Sampling occasion	Before filter	After filter	
	(mg fatty acids/m <sup>3</sup> )	(mg fatty acids/m <sup>3</sup> )	
1: Old rape-seed oil	$19.1 \pm 13.0$	$12.0 \pm 5.5$	
2: New rape-seed oil	$3.7 \pm 1.6$	$3.1 \pm 1.0$	

# **Experiences from the pilot-scale biofilter**

# Flows, pressure drops and temperatures

The problems encountered in the full-scale biofilter initiated further studies on a pilot-scale filter. The flow before and after the biofilter was measured twice a day during operation, and the pressure drop over the filter units several times per day. As can be seen from Table 3, the flow was significantly higher in the channel after the filter, indicating some leakage into the filter construction. The flow seemed to decrease slightly over time, possibly due to an increasing pressure drop. The average flow through the filter was approximately 390 m<sup>3</sup>/h, thus creating an average surface loading of about 1000 m<sup>3</sup>/m<sup>2</sup>\*h and an average EBCT of about three seconds. The overall pressure drop was relatively stable over the entire period, approximately 1700 Pa/m, which shows that rockwool fibre mats developed lower pressure drops than the loose rockwool used in the full-scale filter.

**Table 3**. Average flows before and after the pilot-scale biofilter and pressure drops over the filter units; average values with standard deviations

Parameter	
Flow in channel before filter	$341 \pm 52 \text{ m}^3/\text{h}$
Flow in channel after filter	$434 \pm 64 \text{ m}^3/\text{h}$
Pressure drop over unit 1	$535 \pm 98 \text{ Pa}$
Pressure drop over unit 2	$619 \pm 83 \text{ Pa}$
Pressure drop over unit 3	$561 \pm 154  \text{Pa}$

The room temperature, where the pilot-filter was situated, varied between 14°C and 23°C (day and night variations), with an average value of 18°C. The temperature of the air passing through the biofilter (logger placed in the lid of the filter construction) fluctuated between 4 and 28°C, with an average value of 15°C. This difference was due to the activity of the fryer and also because of a draft of cold air from the outlet. The temperature in the middle of the filter units varied between 1.5°C and 35°C, and most of the variations could be related to the frequency of the irrigation and the activity of the fryer. In general, the temperature peak was noted in the afternoon after 5-6 hours of operation, with the lowest temperatures being detected early in the morning (around 05:00) and during the week-ends.

The fan and irrigation was turned off for 24 hours on one occasion (day 27), and temperatures below 0°C were detected in the middle of filter units 2 and 3. Also, on two occasions (day 22 and 25), parts of the filter media in the lower part of filter unit 3 froze when the outdoor temperature was below -25°C. However, no decrease in bacterial numbers could be detected because of this. Nevertheless, this shows that considerations must be taken to the placement of the filter in a full-scale application in cold climates. Bed temperatures between 10 and 40°C are acceptable for the mesophilic micro-organisms most frequently present in a biofilter, but one should strive to keep the off-gas temperature close to their optimum range for biological activity (30-35°C) <sup>7,2</sup>.

#### Sampling of the filter media

Sampling of filter media was performed on eight occasions; results are presented in Table 4, where viable count of the inoculum and the drainage water are also presented for comparison. Measurements of the pH were performed on all these occasions (in drainage as well as in media samples mixed in NaCl) and the pH was found to be relatively stable around 7 ( $\pm 1$ ) on all occasions. This is advantageous since most micro-organisms capable of degrading VOCs show optimum growth at pH values between 6.5 and 7.5  $^{7}$ .

**Table 4.** Filter media sampling in filter unit 1, 2, and 3 (on days 2, 3, 7, 14, 17, 22, 25, and 28). Average values of moisture content, ignition residue, and viable count are presented with standard deviations. Sample point  $1_1$  consisted of hydrophobic media, while sample point  $1_2$  to  $3_3$  consisted of hydrophilic rockwool media

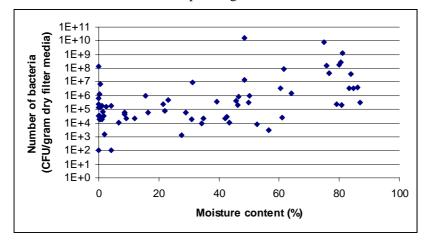
Sampling point	Moisture content (%)	Ignition residue (%)	Viable count		
			(CFU/gram dry media)		
$1_1$	$17 \pm 16$	$4.8 \pm 3.8$	$3.5 E+4 \pm 4.4 E+4$		
$1_2$	$11 \pm 16$	$1.0 \pm 0.3$	$9.2 E+5 \pm 2.3 E+6$		
1 <sub>3</sub>	$68 \pm 17$	$0.5 \pm 0.3$	$2.7 E+9 \pm 6.6 E+9$		
$2_1$	$40 \pm 16$	$0.8 \pm 0.3$	$1.3 \text{ E+5} \pm 1.6 \text{ E+5}$		
$2_2$	$7 \pm 9$	$0.9 \pm 0.1$	$2.4 \text{ E+5} \pm 3.5 \text{ E+5}$		
$2_3$	$76 \pm 9$	$0.3 \pm 0.2$	$1.3 E+9 \pm 3.1 E+9$		
31	$44 \pm 12$	$1.0 \pm 0.9$	$2.7 \text{ E+5} \pm 3.3 \text{ E+5}$		
$3_2$	$28 \pm 21$	$0.6 \pm 0.2$	$1.8 \text{ E+7} \pm 4.7 \text{ E+7}$		
$3_3$	$78 \pm 8$	$0.2 \pm 0.1$	$3.0 E + 8 \pm 4.7 E + 8$		
Drainage			$9.4 \text{ E+5 CFU} / \text{ml} \pm 7.8 \text{ E+5}$		
Inoculum			1.1 E+9 CFU / ml ± 7.8 E+8		

The average moisture content over the period was 40%. However, the variations within the filter were large (from 0.1 to 85%), as shown in Figure 3. This could be due to the high velocities of air passing through the filter; if the air is not humidified to 100% when it enters the filter, it will dry out the material. The best way to solve this would be to install a separate humidification chamber before the biofilter reactor, and to increase the biofilter area to lower the velocities through the filter. In fact, it was filter unit 3 that seemed to have the most stable conditions throughout the experimental period most likely because of considerably lower air velocities passing through. A relatively large number of bacteria were found in all the samples, well comparable to numbers previously found in biofilters <sup>8,9</sup>.

The highest number of bacteria was found in the front and lower end of filter unit one, but numbers of the same order of magnitude were found in the lower ends of units 2 and 3. This indicates that the substrates from the gas were well distributed over the whole filter and not mainly accessible for the micro-organisms in the first filter unit. By studying the morphology of the colonies, changes in bacterial populations could be discovered. After less than a week, the specialist culture of *Bacillus* was significantly depleted, such that they were undetectable on agar plates. The mixed culture enriched with rape-seed oil (originally from a waste-water treatment plant) were the dominating culture over the entire biofilter.

Since the sampling provided a large number of data, efforts were made to find a correlation between moisture content and number of bacteria (Figure 3). However, no significant correlation ( $r^2$ =29%) could be established, probably because of the constantly changing conditions in the filter. A sample taken at one time may have had a higher moisture content the day before and since the bacteria has a certain tolerance to altering conditions, the effect will be shown at a later point. Such a reaction delay is evident by looking at the high numbers of bacteria found at 0 to 5% moisture content – very few bacteria could survive in such an environment for longer periods, but for shorter periods it may be acceptable.

Figure 3. Number of bacteria and corresponding moisture content in filter media samples

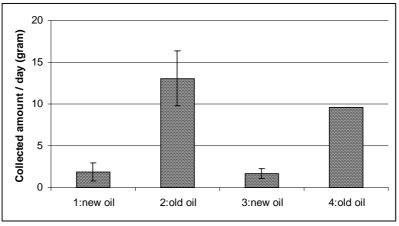


Ignition residues could be used to evaluate the content of organic matter accumulated in the filter material. The rockwool initially contains about 1% of binder, which also is organic and partly degradable. The average ignition residue for the whole biofilter, obtained over the entire experimental period was 2%, considerably lower than values found in the full-scale filter media where values up to 50% were found.

# Evaluation of the mechanical collector for grease aerosols

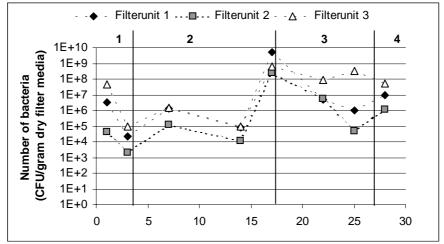
Two mechanical collectors for the removal of grease aerosols were placed directly above the fryer, one traditional kitchen filter in steel (about 5 cm in thickness) and one textile fibre filter. These filters were weighed at the beginning and the end of each day of operation (precision: 0.01 gram). The experimental period was divided in four, with regards to the age of the oil used in the fryer. During the first (day 1-6) and third (day 18-27) periods, new rape-seed oil was used, while older rape seed oil was used during the second (day 7-17) and fourth (day 28) periods. Since period four only consisted of one day of operation no standard deviation could be established. Significant differences in collected amounts were noticed, as can be seen in Figure 4. A similar result was obtained if the collected amount was related to the load (amount of potatoes fried) instead of time (not shown). The two mechanical collectors seemed to be efficient and only small deposits could be detected in the channels to the biofilter, measured on pieces of tape. After 28 days these pieces had collected a thin layer of grease, corresponding to a weight increase in the order of 0.01 gram (2%).

**Figure 4**. Accumulated amounts of grease aerosols on mechanical collectors: average day values with standard deviations



The number of bacteria also seemed to fluctuate in accordance to these four periods, in particular filter unit number one, see Figure 5. This may suggest that the amount of organic off-gas constituents was higher when the rape-seed oil used in the fryer was older. The decrease in numbers during the first period could probably be explained with a certain adaptation period needed for the bacteria. Such an acclimation could range from minutes to days and even weeks, depending on the microbial culture and the substrate to be degraded <sup>3, 10</sup> where longer times may be necessary to complete acclimation in biofilters treating complex mixtures <sup>2</sup>.

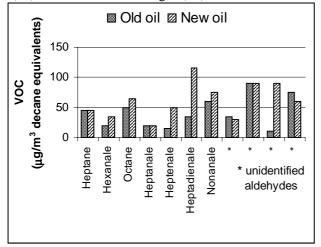
**Figure 5.** Number of bacteria in the biofilter during the four experimental periods (period 1 and 3: new oil, period 2 and 4: old oil). The values presented are average values for each filter unit (calculated from the three sampling points) on eight sampling occasions (days 2, 3, 7, 14, 17, 22, 25, and 28)



# Air sampling of TVOCs and aldehydes

At the start of the experimental period, parallel air samples of TVOCs and aldehydes of the fryer emissions were taken. The TVOC measurement was used as a general screening to identify the major compounds and their concentrations. Sampling was performed when frying both with old and new rape-seed oil. Results clearly showed that aldehydes were the dominating compounds, as can be seen in Figure 6a. Similar results have earlier been found by Andersson et al. <sup>11</sup>(oxidation of rape-seed oil) and Moortag et al. (deep-frying chicken) <sup>12</sup>. The identification of each aldehyde proved to be difficult, since the concentrations were low and the number of different compounds were vast. Therefore, only hexanale and nonanale could be identified with certainty. The amount of ketones was low, which is good since these compounds could interfere with the aldehyde analyses. Since the aldehydes dominated the TVOC analyses, it was decided that further sampling for evaluation of the biofilter was performed by aldehyde analyses.

**Figure 6**. VOC sampling of the emission gas from the fryer. Two samplings were performed: gas emitted when new and old rape-seed oil was used. Identified dominating compounds shown to the left (6a) and TVOCs to the right (6b)



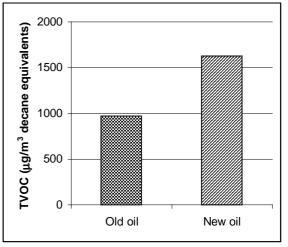
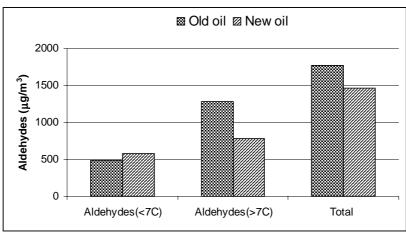


Figure 6b also indicates that when frying with new rape-seed oil, as compared to frying in old rape-seed oil, more VOCs were emitted the air. However, the analysis of the parallel aldehyde sampling gave a somewhat different result, which can be seen in Figure 7. The total amount of aldehydes were actually higher in the emissions from frying with old rape-seed oil, although the difference was less pronounced than the TVOC results. One should bear in mind that these two analyses were performed with different methods and different standards, and comparisons should be made with caution. A difference in result was also found depending on the molecular size of the aldehydes. Aldehydes with less than seven carbon-atoms were slightly more numerous in the emissions from new oil, whereas the aldehydes with seven carbon-atoms or more were found in lower quantities. It is likely that the most volatile compounds are emitted as soon as new rape-seed oil is heated, but that the more complex compounds take longer before they volatilise and may be dominating as the oil gets older. The composition and structure of the oil is constantly changing when exposed to heat, light, oxygen, and potatoes, which makes a clear definition of the gas stream virtually impossible.

**Figure 7**. Aldehyde sampling of the emission gas from the fryer. Two samplings were performed: gas emitted when old and new rape-seed oil was used in the fryer



To evaluate the performance of the biofilter, three samplings of aldehydes were performed before and after the biofilter after about three weeks of operation. All measurements were performed with new rape-seed oil in the fryer (third period) on days 22, 23, and 25. The results are presented in Figure 8, where it can be seen that the total aldehyde concentration was slightly higher after the biofilter, although the values are of the same order of magnitude. The aldehydes with less than seven carbon-atoms were slightly reduced on two occasions, whereas the aldehydes with seven carbon-atoms or more increased in concentration at all occasions (not shown).

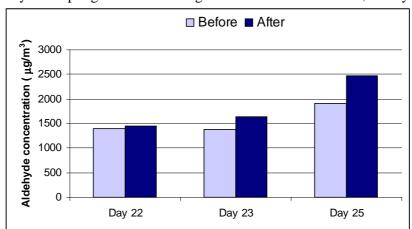


Figure 8. Aldehyde sampling of the emission gas before and after biofilter, on day 22, 23, 25

These results indicate that aldehydes were not reduced in the biofilter, but rather released. This may suggest that the heterogeneous bacteria oxidised carbon components such as alkanes or alcohols into aldehydes in the filter. However, many factors could have influenced the result, which makes it difficult to interpret. The leakage into the filter construction could have affected the results due to the high concentrations of aldehydes in surrounding air (because of the diffusion from the fryer). The bacteria found in the filter with the viable count method may not have been very active or did not have time to degrade the aldehydes, simply because the residence time was too short. It is also possible that three weeks was too short for the proper bacterial culture to establish and become active. All the same, the results are surprising and demand further investigations in the design of the filter, the composition of the off-gases, the activities of the bacteria in the filter and the best method for evaluating the biofilter performance.

#### CONCLUSIONS

A full-scale biofilter installed at a fast-food hamburger restaurant was in operation for approximately one year. Overall, the staff was happy with the filter performance; they perceived that the fatty odours were reduced notably and that the grease aerosols were effectively removed. However, sampling of fatty acids in the air before and after the biofilter was performed on two occasions, but no significant reduction could be established. Sampling of the filter media after 10 months revealed that the moisture content and the bacterial numbers were very low. Questions arose if the filter was really working as a biofilter or mainly as a mechanical collector for fat and grease aerosols and some volatile odorous components. The high deposition of these hydrophobic compounds in the filter bed lowered the pH and hindered the transfer of water, substrate, and oxygen. It also interfered with the irrigation system by inhibiting the load cells to function properly.

High pressure drops also developed over the loose rockwool filter material, thus lowering the outflow from the kitchen and restaurant.

Further studies were performed on a pilot-scale biofilter, treating off-gases from a fryer. Fibre mats with pre-set structure replaced the loose rockwool and the pressure drop was reduced. The material seemed to be an appropriate habitat for micro-organisms. The inoculated mixed culture, previously enriched with rape-seed oil, was found in relatively high numbers (10<sup>5</sup>-10<sup>10</sup>/gram dry media) and dominated over the specialist culture of *Bacillus* that was significantly depleted after one week of operation. The mechanical collector for grease aerosols, composed of one metal filter and one fibre filter installed upstream of the biofilter, proved to be efficient. Very small depositions in the channel and in the filter media could be detected and the pH in the biofilter was stable around 7. Moisture content in the filter material ranged from 0,1 to 85%, with an average of 40%. The timer-based control system for irrigation worked satisfactory, but a separate humidification chamber before the biofilter would be a way of improving the moisture control. The high velocities through the filter imply a risk for fast drying out of the material and provide short residence times. No reduction of aldehydes occurred in the biofilter, probably due to the short residence time or that the proper bacterial culture did not have time to establish in the filter. Interference in the form of leakage or high concentrations of aldehydes in surrounding air could also have influenced the result.

It can be concluded that the off-gas from the fryer is a very complex mixture due to the constantly changing structure of the oil. Significant differences in composition were found between new and old rape-seed oil. This makes an evaluation of the biofilter difficult and demands further investigations for the proper design of this type of biofilter.

#### Future work

Further studies in the pilot-scale filter will be performed, with a lower flow in order to decrease the face velocities and increase the residence time. Improvement of the moisture control is needed, as well as further investigations of the emissions composition and the best evaluation method for the biofilter.

#### **ACKNOWLEDGEMENTS**

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# **PAPER III**

# **Evaluation of rockwool biofilter media for the treatment of restaurant emissions**

Andersson, A

In proceedings of the 2000 USC-TRG Conference on Biofiltration (an air pollution control technonology), University of Southern California, Los Angeles, California, USA
October 19-20, 2000

# **Evaluation of rockwool biofilter media for the treatment** of restaurant emissions

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#### **ABSTRACT**

Due to odorous, fatty gas emissions, restaurants are often targets of public complaints that could lead to expensive litigation or, in the worst case, eviction. Biofiltration could be an appropriate method to treat restaurant waste gas, which contains a mixture of partially oxygenated hydrocarbons with aldehydes among the main odour causing compounds. However, when space is at a minimum the large areas normally required for biofiltration could be a problem. When trying to develop a compact biofilter for this application, the choice of biofilter media becomes crucial. The objective of this study was to evaluate different rockwool media, with respect to flow distribution, pressure drop, chemical and mechanical resistance, and aptness as immobilisation matrices for microorganisms. Two of the rockwool media, one hydrophobic and one hydrophilic, were then used in two test runs in a pilot scale biofilter coupled to a potato fryer with rapeseed oil. The average surface loading was 1,000 m<sup>3</sup>/m<sup>2</sup>\*h (residence time of about 3 seconds) during the first test run, and 400 m<sup>3</sup>/m<sup>2</sup>\*h (residence times about 9 seconds) during the second test run. Evaluation of the biomass showed that a mixed culture was able to immobilise and grow in both media. Moisture content ranged from 10 to 20% in the hydrophobic and from 40 to 50% in the hydrophilic rockwool. The organic substance in the filter material (biomass, binder, and accumulated oxidation products from the oil) was evaluated as total volatile solids. Aldehyde sampling at the end of the pilot scale test runs indicated no reduction of total aldehyde concentrations in the biofilter, probably due to the low solubility of some of the components in combination with short residence times.

#### **KEY WORDS**

Biofilter, rockwool, grease, fast food restaurants, odours, aldehydes

# **INTRODUCTION**

During the last decades, biofiltration for air pollution control has been established as a reliable, cost-effective technology for controlling low-concentration, biodegradable waste gases. It is a technique that could be used to treat various volatile compounds in a wide range of industries and public sectors (Leson & Winer, 1991; Devinny *et al.*, 1999), e.g. the food industry. Because of their odorous and fatty gas emissions, restaurants are often the targets of public complaints that could lead to expensive litigation or, in the worst case, eviction. These emissions tend to contain a mixture of partially oxygenated hydrocarbons (i.e. carboxylic acids, aldehydes, ketones, alcohols, and esters) whose composition and concentration vary with time. The main odour causing compounds are suspected to be the saturated and unsaturated aldehydes (Moortgat *et al.*, 1992). A TVOC (Total Volatile Organic Carbon) measurement, performed to identify the major gas compounds from the fryer used in this study, showed that aldehydes were the dominating compounds (Andersson, 2000). Andersson, 1998, found similar results when studying oxidation of rapeseed oil. Biofiltration could be an appropriate method to treat the waste gas from a restaurant facility. Other abatement techniques such as aqueous absorption or adsorption on activated carbon would not

be effective because of the relatively low solubility of some aldehydes and high moisture content of the emitted gas. As well, burning the off gas would be economically impossible. The large areas normally required for biofiltration could be a problem because of the restricted available space in a restaurant environment. When trying to develop a compact biofilter for these types of applications, extra care must be taken in the choice of biofilter media. The properties and characteristics of the support medium largely govern the overall effectiveness of a biofilter. The medium serves as living space for the microorganisms, humidity reservoir, and mechanical support for the maintenance of the internal structure of the filter bed.

A good packing material should have a high water retention capacity without becoming saturated, low bulk density, structural integrity, capacity to buffer acidification of the filter material, and the ability to buffer high concentrations of the contaminants (Devinny *et al.*, 1999). Filter media have traditionally consisted of organic material such as soil or compost, but in recent years the composition of packing materials has undergone notable improvements to retard the ageing effects and maintain the bed porosity.

Fibre based materials have been successfully used, for example, in biotrickling filters (Rydin et al., 1994; Wittorf et al., 1997; Ostlie-Dunn et al., 1998). The advantage of using fibres instead of granules is the higher surface-to-volume ratio that can be obtained, which improves the substrate mass transfer and provides more surface area for adsorption and microbial immobilisation. Rockwool fibre mats with pre-set structures and low densities are less subject to compacting and ageing and their use facilitates handling and improves flow distribution. In addition, the characteristics of the rockwool can be specifically designed, i.e. density, fibre length and thickness, amount of binder, hydrophobic/hydrophilic properties, etc, making it a very versatile filter medium. Since the rockwool does contain neither microorganisms nor nutrients, these must be added separately.

#### Objective and scope

The objective of this study was to evaluate six different rockwool media for their aptness to be used in a biofilter for the treatment of restaurant emissions. Parameters examined included flow distribution, pressure drop, chemical and mechanical resistance, and aptness as immobilisation matrices for microorganisms. Two media were selected and tested in a pilot scale biofilter in two test runs.

# MATERIAL AND METHODS

# Experimental set-up

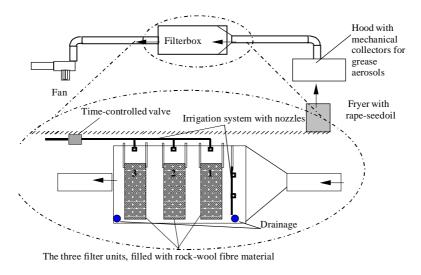
The six different types of rockwool materials used are presented in Table 1. Materials 1, 2, and 3 (from manufacturer A) were specifically designed to be either hydrophobic or hydrophilic, whereas materials 4 (manufacturer B), and 5 and 6 (manufacturer C) were regular insulation mats with mainly hydrophobic characteristics.

**Table 1.** Description of the different rockwool materials used in the study.

Sample number	Rockwool description	Mat size (m)	Density (kg/m <sup>3</sup> )
1	A, loose hydrophilic	-	80
2	A, hydrophilic preset mat	0.6*0.6*0.03	33
3	A, hydrophobic preset mat	0.6*0.6*0.15	27
4	B, preset mat	0.6*0.6*0.15	40
5	C, preset mat	0.6*0.6*0.10	80
6	C, preset mat	0.6*0.6*0.10	100

A pilot scale biofilter set-up (Figure 1) was initially used to evaluate flow and pressure drop through the six different rockwool media, each having a moisture content of 40% (no bacteria). Only the first filter unit was used at this stage. Air velocities through the filter unit were determined using a *SwemaAir 30* (SWEMA, Farsta, Sweden), and the pressure drop over the filter unit was measured with a *SwemaMan 2000* (SWEMA, Farsta, Sweden).

After evaluating the flow characteristics of the materials, small samples of each material (0.02\*0.03\*0.08 m) were placed in glass jars with a sludge suspension from an activated sludge unit treating municipal wastewater. The samples were continuously agitated for ten days on a shaking table (100 rpm, Lab-Shaker, Adolf-Küner AG Basel, Switzerland). The chemical and mechanical resistances of the materials were thereafter examined, and by looking at the surface as well as through a microscope the immobilisation of microorganisms was inspected.



**Figure 1.** Schematics of the pilot scale biofilter.

#### Pilot scale test runs

During the pilot scale test runs, only two of the rockwool materials were used (nos. 2 and 3). The biofilter was coupled to a potato fryer with rapeseed oil (Figure 1) which was heated to 180°C and whose emissions contained a mixture of partially oxygenated hydrocarbons with both hydrophobic and hydrophilic characteristics. The pilot scale was composed of three filter units that operated in a side flow mode, each filter unit having a depth of 0.3 m. with a square area of 0.6\*0.6 m. A timer-based irrigation system with spray nozzles in the inlet (to saturate the off-gases) and at the top of the biofilter (for direct irrigation) was utilised. Drainage was collected at the bottom of the filter and a mechanical collector for grease aerosols was installed upstream of the biofilter, right above the fryer. The first filter unit was filled with both hydrophilic rockwool mats (no. 2) and thin layers of hydrophobic mats (no. 3), to improve the distribution of both water and hydrophobic compounds in the filter. The second and third filter units were filled with hydrophilic media (no. 2) only. The fibre mats were inoculated with a mixed bacterial culture selected from a wastewater treatment plant, previously enriched with rapeseed oil, and a culture of Bacillus. A nutrient solution was spread over the filter, resulting in approximately 40% moisture content at the start-up of the filter. The nutrient solution used had the following composition:  $0.5 g (NH_4)_2SO_4$ , 0.25 gNaNO<sub>3</sub>, 0.013 g CaCl<sub>2</sub> \* 2 H<sub>2</sub>O, 0.21 g MgSO<sub>4</sub> \* 7 H<sub>2</sub>O, 0.5 g KH<sub>2</sub>PO<sub>4</sub>, 0.25 g NaH<sub>2</sub>PO<sub>4</sub> \* H<sub>2</sub>O, 500 ml H<sub>2</sub>O, and 20 ml 0.2 M NaOH.

The first pilot-scale test runs lasted twenty-eight days, with two units of the fryer operating 7-8 h/day, 5 days/week. After a shut-down of about four weeks, a second set of pilot-scale tests were run for fifteen days at lower surface loadings, increased residence times, and lower mass loadings (only one unit of the fryer). Inoculation and nutrient addition was performed again for the second test run.

#### Medium sampling

From numerous points, sampling of the filter media was performed regularly from the pilot-scale filter during the experimental periods. Moisture content and total volatile solids were determined according to the Swedish standard SS 02 81 13. Microbial cell enumeration was performed using an extraction technique and plating method (viable count). Approximately 1-2 grams, dry weight, of filter media were added to a dilution bottle containing 50 ml of sterile 0.9% NaCl, and then left for one hour on a shaking table (140 rpm, Lab-Shaker, Adolf-Küner AG Basel, Switzerland). A series of dilutions (in sterile 0.9% NaCl) were prepared and a volume of 0.1 ml sample was spread over the surface of a nutrient agar plate. The plate was then incubated for two days at room temperature (21°C±2°C) and the number of colonies were counted. Counts between 20 and 300 colonies per plate were considered significant. The results are expressed as CFU/gram dry filter media (CFU: colony-forming units). pH of the collected drainage as well as in the dilution bottles with media and NaCl (from viable count) were measured using a pH electrode.

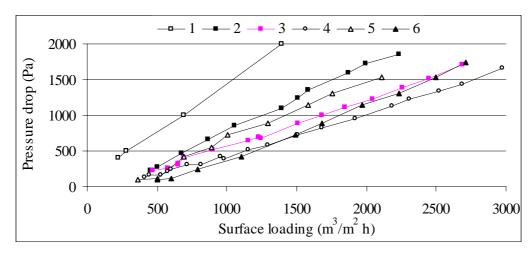
# Gas sampling

Since aldehydes were suspected to be the main odour causing compounds in the emission, the biofilter was evaluated by aldehyde sampling at the end of each experimental period. The identification of each aldehyde proved to be difficult because of their low concentrations and the vast number of different compounds. Samples were taken from the channel before and after the biofilter with low-flow pumps and sorbent sample tubes (Waters Sep Pak XPosure\*). For identification of aldehydes, a TPS HPLC (high-performance liquid chromatograph) was used with a DAD SM 5000 detector, using a standard solution consisting of: formaldehyde, acetaldehyde, acetone, acroleine, propanal, crotonaldehyde, butanal, bensaldehyde, pentanal, hexanal, heptanal, octanal, nonanal, and decanal.

# **RESULTS AND DISCUSSION**

#### Pressure drop vs. surface loading

Results from the initial pressure drop-loading tests are presented in Figure 2. A linear relationship between pressure drop and surface loading was found for all these materials ( $r^2 \ge 0.99$ ), even at very high gas velocities. These results deviated from previous experiments conduced by Van Langenhove *et al.*, 1986; Allen & Yang, 1991; and Sabo *et al.*, 1993. Their experiments using organic materials (i.e. wood bark, compost, peat, porous burned clay, wood chips, and coconut fibres) revealed that the pressure drop first increased in direct proportion to the rising air velocity, but then deviated from linearity at higher air velocities forming a semi-parabolic curve. Thus, the behaviour of pressure drop seems strongly dependent on the type of material used.



**Figure 2.** Pressure drop over one filter unit (at 40% moisture content) versus surface loading for the six different rockwool materials.

The loose rockwool (no. 1) had a tendency to compact, which induced an uneven flow through the filter unit and caused a notable pressure drop over the filter unit even at low surface loadings. Earlier experiences from a full scale experiment with this type of rockwool (Andersson, 2000) also showed that the pressure drop increased significantly over time. Material no. 1 was, therefore, excluded from further testing. Materials 2 to 6 (all with preset fibre mats) seemed more capable of favouring an even air distribution through the filter and to develop lower pressure drops. No evident relationship between pressure drop and density of the rockwool was established. Materials nos. 4 and 6 showed the lowest pressure drops with their densities differing substantially (40 kg/m³ as compared to 100 kg/m³).

# Resistance and immobilisation

The effects of the rockwool media samples being continually agitated in an activated sludge liquid were evaluated visually and their results are presented in Table 2. The chemical and mechanical resistance of materials 1 to 3 was found to be relatively good, as well as their aptness as immobilisation matrices for microorganisms. Materials 4 to 6 lost their mechanical structure and fell apart when submerged in water. Therefore, they were excluded from further testing.

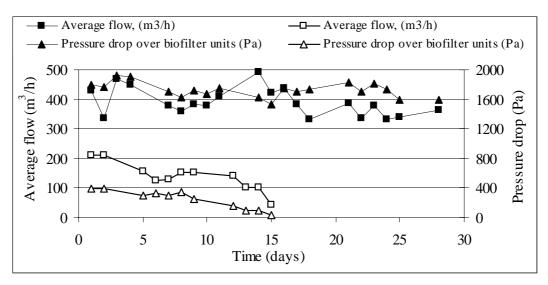
**Table 2.** Results of continuos irrigation with activated sludge over the different rockwool materials.

Sample number	Mechanical and	Immobilisation of		
	chemical resistance	microorganisms		
1	++	+++		
2	+++	+++		
3	+++	++		
4	+	+		
5	+	++		
6	+	++		

+++ Very good, ++ Average, + Poor

#### Pilot scale test runs

For our following pilot scale test runs, only filter material numbers 2 and 3 were used since these two seemed to be the most appropriate for the application. The operational flow and resulting pressure drop for the two pilot-scale test runs are presented in Figure 3.



**Figure 3.** Average flow and pressure drop through the biofilter versus time; filled symbols represent the first pilot scale test run, while empty symbols represent the second pilot scale test run.

During the first test run, the average surface loading was 1,000 m³/m²\*h, resulting in an empty bed residence time of approximately 3 seconds. The pressure drop during this time was approximately 1,700 Pa over the biofilter. During the second test run, the average surface loading was 400 m³/m²\*h, resulting in an empty bed residence time of approximately 9 seconds. The pressure drop was then approximately 250 Pa over the biofilter units. A decrease of flow was noticed after roughly ten days, with a simultaneous reduction of pressure drop over the biofilter units. This was attributed to an accumulation of grease in the mechanical collector upstream of the biofilter, with an increase of the overall pressure drop in the system. Consequently, this filter will have to be cleaned or changed on a regular basis.

# Medium sampling

In Table 3, results from medium sampling of the first filter unit are presented for the two filter materials (nos. 2 and 3) from the first and second pilot scale test run. Moisture content varied between samples, but overall the humidification system seemed to work satisfactorily. As expected, the hydrophilic rockwool was able to hold water better than the hydrophobic, resulting in a higher number of bacteria in these samples. The numbers of bacteria found in this study were comparable to those found by researchers in other biofilter applications (Medina *et al.*, 1995; Cardenas-Gonzales *et al.*, 1998; Acuña *et al.*, 1999).

**Table 3**. Media sampling of hydrophilic (no. 2) and hydrophobic (no. 3) rockwool in the first filter unit. Average values of moisture content, total volatile solids, and viable count are presented with standard deviations for samples from the first and second pilot scale test run.

	Moisture content		Total volatile solids		Viable count		
	(%)		(%)		(CFU/gram dry media)		
	First	Second	First	Second	First	Second	
Hydrophilic (no 2)	40 ± 17	51 ± 12	$1.0 \pm 0.3$	$0.8 \pm 0.2$	$1.2*10^9 \pm 4.3*10^9$	$2.9*10^7 \pm 4.7*10^7$	
Hydrophobic (no 3)	17 ± 16	$10 \pm 13$	$4.8 \pm 3.8$	$3.8 \pm 1.3$	$3.5*10^4 \pm 4.4*10^4$	$1.2*10^5 \pm 1.3*10^5$	

The total volatile solids gave an estimate of how much organic material (including binder, biomass, and accumulated oxidation products from the oil) was present in the filter material during the experimental period. This measure could be used to anticipate clogging of the filter, caused either by excessive biomass growth or by the accumulation of fatty oxidation products. There were no indications of clogging in the biofilter probably thanks to the mechanical collector installed upstream of the biofilter that effectively removed grease aerosols, and also because of the relatively short experimental periods. The total volatile solids in new rockwool material samples were determined for the two media in order to get an estimation of the amount of binder (a phenol formaldehyde resin) initially present in the material. The quantity of binder dictates whether the rockwool will have hydrophilic or hydrophobic properties. New hydrophobic rockwool (no. 3) was found to have about three times as much binder as compared to the hydrophilic (no. 2): 1.8% binder as compared to 0.6% ( $\pm 0.1\%$ ).

In Table 4, a rough estimation has been made of what the total volatile solids consisted of and in what proportions. The calculations were made under the assumption that the whole filter unit was filled with either hydrophilic or hydrophobic rockwool. One filter unit with dry rockwool material weighed 4.5 kg at the start. The amount of biomass was estimated from the numbers of bacteria found by viable count, with the assumption that one bacteria weighs approximately  $5*10^{-13}$  g (a sphere with the diameter of 1  $\mu$ m and the density of 1,000 kg/m<sup>3</sup>).

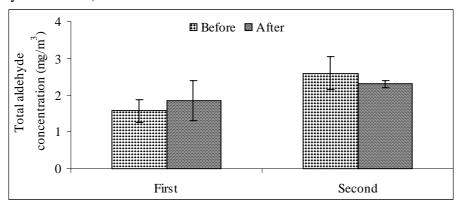
**Table 4**. Average total volatile solids in the first filter unit, based on numbers from Table 3, and a rough estimation of their composition. Data from the first and second pilot scale test run.

	Average total volatile solids (g)		Bi	` '		oxidation acts (%)	Bioma	Biomass (%)	
	First	Second	Firs	t Second	First	Second	First	Second	
Hydrophilic (no. 2)	45	36	60	75	34	24.8	6	0.2	
Hydrophobic (no. 3)	216	171	37	5 47.3	62.4	52.6	0.00004	0.00016	

As can be seen from Table 4, the total volatile solids was almost five times higher in the hydrophobic material as compared to the hydrophilic. For both materials, higher values were found during the first pilot scale run, probably owing to the higher mass loading rates (two fryer units) and, consequently, a greater accumulation of fatty oxidation products. The amount of biomass was negligible in the hydrophobic media, and constituted only a minor part of the hydrophilic media. However, this calculation may underestimate the number of bacteria. Since the viable plate technique only counts those microbes that can grow on the specific nutrient agar used, a large percentage of the present microorganisms in the biofilter may not be included. pH in media samples and drainage was found to be relatively stable at 7 (±1) during both experimental periods, indicating that the material could possess a certain buffering capacity. This is advantageous since most microorganisms capable of degrading VOCs show optimum growth at pH values between 6.5 and 7.5 (van Lith *et al.*, 1997).

#### Air sampling of aldehydes

Aldehyde samples taken before and after the biofilter are presented in Figure 4, as average values with standard deviations. Three samples were taken at the end of the first pilot scale run (on days 22, 23, and 25) and two samples were taken at the end of the second pilot scale run (on days 14 and 15).



**Figure 4.** Total aldehyde concentration in the air before and after the biofilter, samples taken at the end of the first and the second pilot scale test run. Average values with standard deviations.

Results indicate that no reduction of total aldehyde concentrations was achieved in the biofilter. Certain compounds, i.e. hexanal, were reduced by 10-35%, while others increased in concentration. The most probable explanation for this is poor water solubility for some of the contaminants in combination with too short residence times. Results were slightly improved during the second test run, but residence times will probably have to be substantially increased to obtain a significant reduction. Typical residence times needed for commercial applications range from 25 seconds to several minutes (Leson & Winer, 1991; Devinny et al., 1999). To achieve this, a larger surface area of the filter will most likely be needed, which in turn poses a challenge since restaurant space is limited. However, a solution with several filters in parallel would be one way to solve this problem. On-line sampling of concentrations before and after the biofilter would be preferable to sorbent samples. Since the biofilters were run for a fairly short time, there is also a possibility that the proper bacterial culture did not have enough time to establish and become active in degrading the aldehydes. The microbial community in the filter, earlier enriched with rapeseed oil, could have survived with the oxidation of fatty acids that accumulated in the filter, and may have had little interest in the passing aldehydes. Physical adsorption may have been exhausted due to saturation and might have caused a release of aldehydes from the filter.

#### CONCLUSION

This study shows that rockwool could be a suitable biofilter media for the treatment of restaurant emissions. Rockwool mats with pre-set structures developed considerably lower pressure drops with improved gas flow distribution when compared to loose rockwool. Two specially designed rockwool mats, one hydrophobic and one hydrophilic, proved to possess good chemical and mechanical stability even when submerged in water. Two test runs were performed using these two rockwool media, and results showed that a mixed culture was able to immobilise and grow in the filter media. Moisture content ranged from 10 to 20% in the hydrophobic and from 40 to 50% in the hydrophilic rockwool during the experimental period with a higher number of cultivable cells found in the latter one. A higher content of total volatile solids (including binder, biomass, and accumulated fatty oxidation products) was present in the hydrophobic filter material. There were no indications of clogging in the biofilter possibly because of the mechanical collector, which effectively removed grease aerosols, or the relatively short experimental periods. pH remained stable at around 7 in the drainage as well as in both materials throughout the experimental periods, thus indicating that the material possessed a certain buffering capacity. Aldehyde sampling at the end of the pilot scale test runs indicated no significant reduction of total aldehyde concentrations in the biofilter, most likely due to poor water solubility for some of the contaminants in combination with too short residence times. Future work involves test runs at increased residence times as well as long-term testing of the media in a biofilter.

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