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On-line Monitoring of Dissolved Gases Using Microporous Membrane Inlet and Mass Spectrometry

B. S. Ferreira, F. van Keulen and M. M. R. da Fonseca*

Centro de Engenharia Biológica e Química, Instituto Superior Técnico
Av. Rovisco Pais, 1049-001 Lisboa, Portugal

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Summary

Mass spectrometry has been widely used for the monitoring of fermentation processes, since it allows the on-line analysis of gas streams and of dissolved gases or volatile components in fermentation broths. In the present study, an interface modul consisting of a microporous hydrophobic hollow fibre membrane was proposed. This module, installed in an external loop, worked as a gas contactor between a stream of recirculating liquid and a carrier gas which was analysed by the mass spectrometer. The feasability of such system was demonstrated and optimisation procedures in order to achieve satisfactory analysis sensitivity and response times were discussed.

Key words: fermentation monitoring, mass spectrometry, microporous hydrophobic hollow fibres

Introduction

Mass spectrometry is used for the on-line analysis of gas streams and of dissolved gases or volatile components in fermentation broths. The potential advantages of mass spectrometry over other analytical techniques are outlined in the literature (*1*).

In mass spectrometry, samples are introduced into high vacuum chamber ($\approx 10^{-3}$ Pa) where they vaporise and ionise. The ionised molecules are then focused into the mass analyser and are separated according to their mass to charge ratio (m/z). One of the most critical features in reaching maximum precision and accuracy is the inlet system and associated processes involved in the sample transfer from the fermenter to the high vacuum of the mass spectrometer (MS), as it was recognised by several authors (*1*).

The most popular inlet systems for mass spectrometric analysis of gas phases are capillary and membrane inlets. Capillary inlets sample a fraction of the gas to be analysed and reduce the pressure of the sample to the high vacuum under which the mass spectrometer works. Membrane inlets provide a barrier, through which the analysed molecules must diffuse, between the sample and the vacuum.

Most inlet systems, for the analysis of dissolved gases or volatiles in a liquid phase, consist of pervaporation membranes like those used in sampling from a gas phase. Some are located directly in the fermenter, while others are in contact with a stream recirculating from the fermenter.

Simultaneous analysis of both gas and liquid phases is invaluable in the fermentation industry for the monitoring of gaseous and liquid process streams. However, this is only achieved with a few systems. One of the difficulties is that the total pressure and water content in the ionisation chamber differ considerably from phase to phase. Furthermore, few mass spectrometers have valve systems that allow to alternate between two inlets directly connected to the high vacuum region.

* Corresponding author; E-mail: p1088@alfa.ist.utl.pt

The interface proposed in this paper is a microporous hydrophobic hollow fibre membrane module that acts as a contactor between a stream of recirculated liquid and a carrier gas to be analysed by the MS.

With such an interface, a liquid phase can be analysed indirectly through the analysis of a gas phase of related composition, *i.e.* the carrier gas flowing inside the fibre lumen. Thus, fermentation monitoring can be performed through alternate analysis of two gaseous streams at atmospheric pressure (one from the fermenter headspace and the other from the membrane module). This is possible with simple spectrometers with valve manifolds before the capillary inlet, allowing the sequential analysis of several gas streams.

The performance of the module was assessed using a water-air system. The response to changes in the dissolved oxygen concentration was compared to the response obtained with an oxygen electrode.

The effect of the carrier gas composition and flow rate, and of the recirculating liquid flow rate on the response of the MS was also evaluated.

Materials and Methods

Implementation of the interface

The interface module was made of a bundle of hollow fibres in a glass shell. The carrier gas flows inside the lumen of the fibres of the membrane module, stripping the components to be analysed from the liquid stream that flows through the shell side of the hollow fibre module (Fig. 1). The concentration of the components in the carrier gas, at the module outlet, is related to their concentration in the liquid phase. Since the fibres are hydrophobic, the gas-liquid interface remains immobilised at the external fibre surface and the pores do not wet. Thus, the transferred molecules diffuse freely in the pores and the resistance of the membrane to mass transfer between phases is negligible (*2*,*3*), in contrast to what happens with silicone membranes used in pervaporation systems, in which the transferred molecules must diffuse into the membrane material.

Microporous polypropylene hollow fibres with inner and outer diameters of 0.330 and 0.630 mm, respectively, were from Akzo/Enka (Plasmaphan®, USA). The

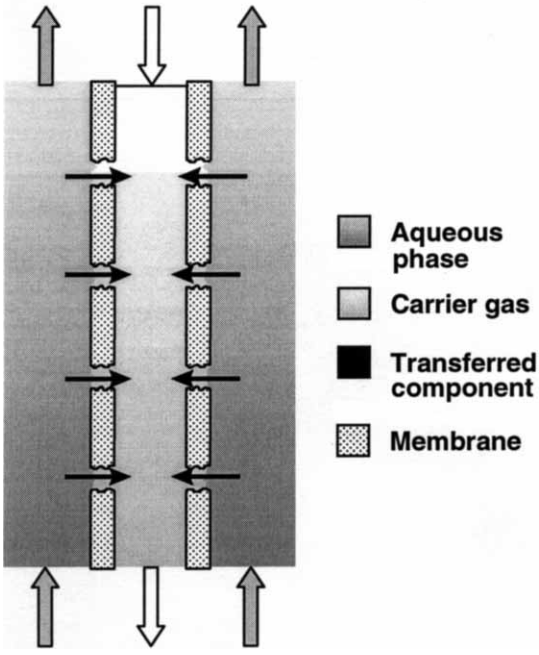

Fig. 1. Operating scheme of the microporus hydrophobic hollow fibre module

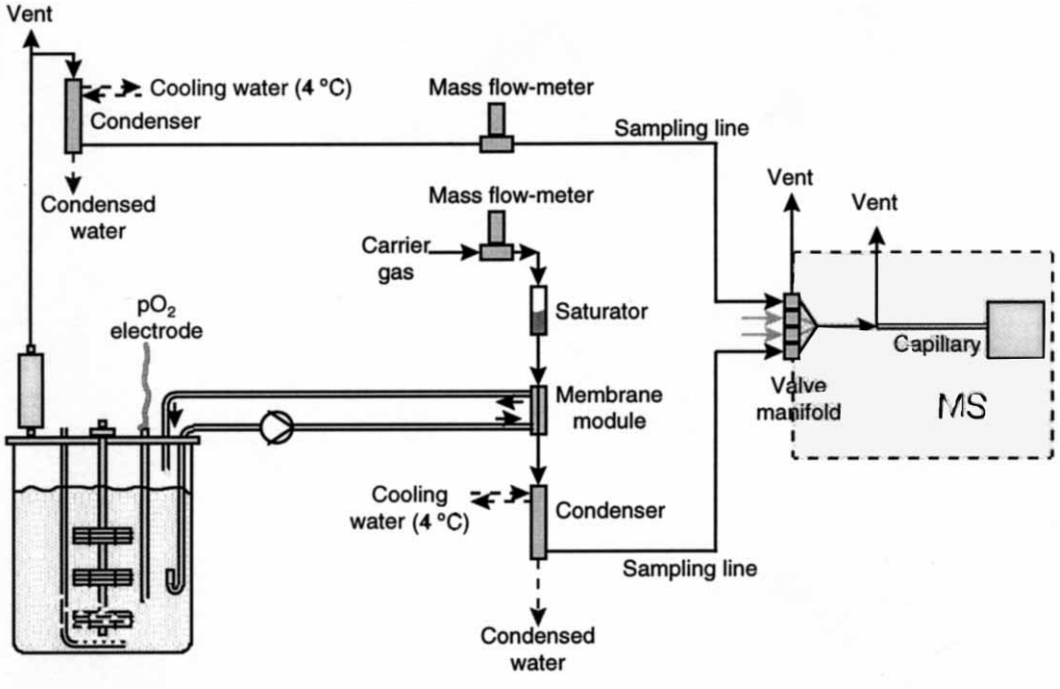

Fig. 2. Experimental set-up for testing module and for the optimisation of the operating conditions

pore diameter was 0.2 μm and the surface porosity was 69 %. The hollow fibre module was constructed by inserting 40 fibres into a glass shell (1.3 cm in internal diameter and 14 cm long) and gluing them on both ends of the shell with an epoxy resin Epo-Kwick (Buehler, USA). The available fibre length for mass transfer was 7 cm and the total surface area for transfer was 55.4 cm^2.

The membrane module was set up in an external loop where the liquid was recirculated (Fig. 2). The advantages of taking the sample in a recirculating stream are two-fold: (*i*) the measurements are not dependent on the mass transfer conditions inside the fermenter and (*ii*) the gas bubbles that would interfere with the measurements can be removed prior to the stream passing the module. Furthermore, it is easier to avoid the fouling of the membranes by choosing an adequate flow rate of the recirculation stream and by pulsing the flow periodically.

The carrier gas was pure nitrogen or a mixture of pure nitrogen with air, so that the gas stream leaving the module had a composition similar to that of typical inlet air or exhaust gas streams from a fermentation. The module was operated in counter-current to maximise the average mass transfer driving force (*5*).

Mass spectrometer

A quadrupole capillary inlet mass spectrometer (Spectra International/Leda Mass Ltd., UK) equipped with a Faraday cup and a multi-valve inlet system allowing sequential analysis of up to 16 sampling lines was used. The MS was controlled by a conventional personal computer with software provided by Leda Mass Ltd. For the monitoring of oxygen, a mass to charge ratio of 32, corresponding to its most frequently occurring fragment, was chosen.

Table 1 shows the compositions of the gaseous mixtures used for the mass spectrometer calibration. They were determined by the extreme vertices design which focuses the calibration to the region of interest within the experimental space (*4*).

Table 1. Composition of the calibration mixtures

	Mixture				
	1	2	3	4	5
	Molar fraction /%				
N_2	97.2	92.9	87.9	82.8	77.7
O_2	2.8	7.1	12.1	17.2	22.3

System set-up

A 2 litre fermenter containing deionised water (Biostat M from B. Braun, Germany) was sparged with 100 mL/min SPT air or nitrogen. All experiments were performed at 25 °C and at 350 rpm. Water was recirculated from the fermenter to the module by means of a gear pump (model MV-Z, Ismatec, Switzerland).

Dissolved oxygen measurements were carried out with an oxygen electrode (Ingold, Switzerland) with a first-order time constant τ_E = 0.1 s. Zero and 100 % calibration of the electrode were performed at 25 °C and 350 rpm by sparging 200 mL/min nitrogen and air, respectively, until stable signals were obtained.

Prior to entering the module, the carrier gas was saturated with water. At the module outlet the carrier gas was dried in a condenser (cooled with water at 4 °C) before being sampled to the MS.

Gas flow rates were controlled by thermal mass flow controllers (Bronkhorst Hi-Tech, Holland).

Results and Discussion

The proposed interface system was first tested by comparing its response to changes in the dissolved oxygen concentration with the response given by an oxygen electrode (Fig. 3). Six different conditions were tested sequentially according to Table 2.

During stage *i*) there was a stable electrode reading, but the signal obtained by the analysis of the carrier gas decreased, stabilising at value of about 0.31 μPa. This decrease can be ascribed to changes in composition of

Table 2. Conditions used in the comparison between the performance of the proposed system and that of an oxygen electrode; the carrier gas flowing through the fibres was pure nitrogen, the water recirculation rate was 200 mL/min, the sparged gas flow rate was 100 mL/min (SPT) and the fermenter stirring speed was 350 rpm

Condition	Fermenter sparging gas	Carrier gas flow rate mL/min SPT
i)	air	100
ii)	nitrogen	100
iii)	air	100
iv)	air	50
v)	nitrogen	50
vi)	air	50

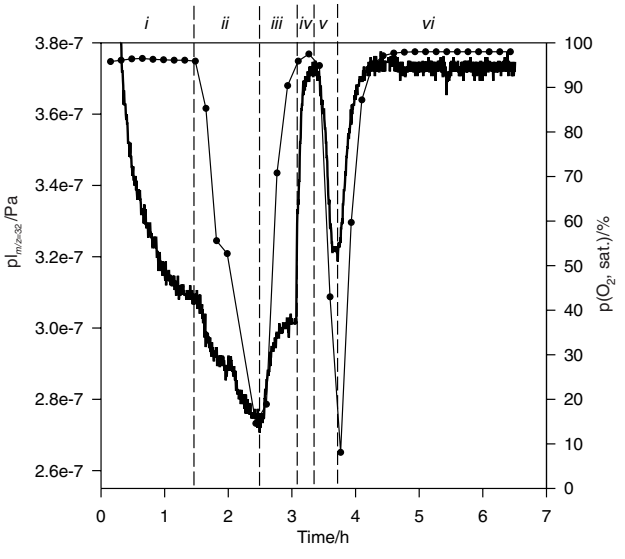

Fig. 3. MS measurements of dissolved oxygen (–) compared with measurements obtained with an oxygen electrode (●)

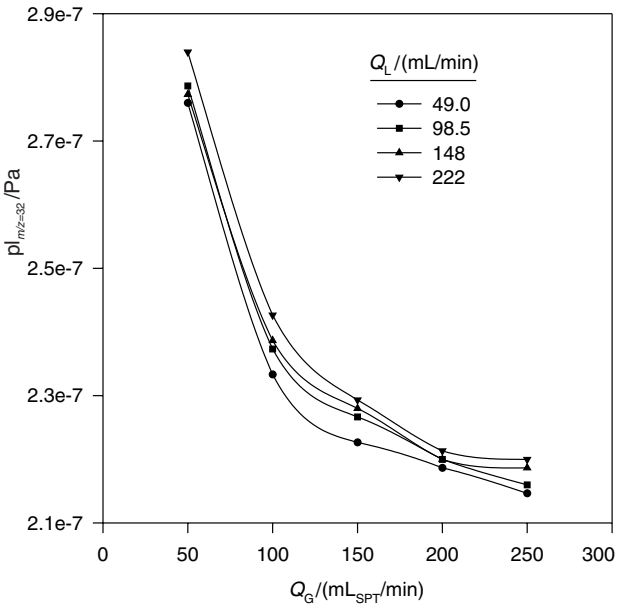

Fig. 4. Effect of the flow rates of the carrier gas (nitrogen), Q_G, and of the recirculated water saturated with oxygen, Q_L, on the MS response at a mass to charge ratio m/z=32

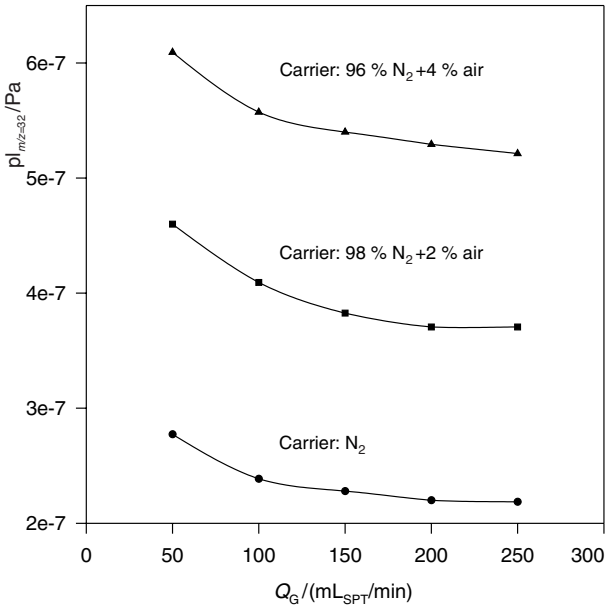

Fig. 5. Effect of carrier gas composition and flow rate on the MS response at a mass to charge ratio m/z=32 at a constant flow rate of 148 mL/min of water saturated with oxygen

the gas mixture in the ionisation chamber of the MS. In fact, the oxygen concentration of the carrier gas after passing through the membrane module is lower than that of air, *i.e.* the gas in the ionisation chamber at the experiment start-up. By the end of stage *i*), the MS is expected to be stabilised. In stages *ii*) and *iii*), the pattern of the electrode readings was closely followed by the pattern of the readings of the MS. The same occurred in stages *v*) and *vi*). In stage *iv*), there was a stable reading of the oxygen electrode, while the MS signal increased sharply from about 0.300 to 0.375 μPa. This is easily explained if we assume that the change in the carrier gas flow rate does not influence the flow of the molecules through the fibres from the liquid phase to the gas phase. This is perfectly acceptable, since it has been shown that the gas side resistance to the overall mass transfer is negligible (*2*,*3*). The mass transfer rate being constant, a decrease in the carrier gas flow rate will result in an increase of the concentration of the stripped component in the carrier gas, leading to a stronger response signal of the MS. In addition, a gain in sensitivity was observed. For example, a change in the dissolved oxygen concentration of about 80 % saturation resulted in a change in the MS response of 0.030 μPa in stage *iii*), and of 0.70 nPa in stage *vi*).

The responses of the MS when measuring a recirculating stream of water saturated with oxygen at different carrier gas and recirculated water flow rates were compared in order to study the influence of the gas and liquid flow rates in the module on the MS response. The lower the gas flow rate, the higher the signal obtained with the MS (Fig. 4). This confirms what was observed for stage *iv*) in Fig. 3, and again is explained by the longer residence time of the carrier gas within the fibres, thus allowing its higher enrichment in the transferred components. Lower gas flow rates were not tested, in order to avoid long response times. The liquid flow rate hardly influenced the MS response, although higher liquid flow rates in the module generally resulted in slightly stronger MS signals, due to an overall higher oxygen concentration in the liquid phase and higher mass transfer coefficients. At flow rates higher than those in the tested range, too many air bubbles were dragged from the fermenter and interfered with the measurements.

The influence of the composition of the carrier gas was also studied. As shown in Fig. 5, the MS response at m/z = 32 increased with the increase of the oxygen content of the carrier gas. This should have opposite effects on the response sensitivity: *i*) increase due to the measurements not being made near the MS background signal; *ii*) decrease due to the damping of the changes detected due to the oxygen content of the carrier gas.

Conclusions

A microporous hydrophobic hollow fibre membrane module was developed to work as a membrane inlet mass spectrometry interface for the analysis of dissolved gases and volatile components in liquid phases. The carrier gas flowing through the lumen of the fibres strips the liquid phase recirculating in a fermenter loop and is subsequently analysed by the MS.

Monitoring of dissolved oxygen concentration changes was successfully made using the developed interface. Operating conditions such as carrier gas and liquid flow rates in the membrane module and carrier gas composition were shown to influence the MS response. Lower carrier gas flow rates and higher carrier gas oxygen content produced higher signals, while the liquid flow rate hardly affected the measurements.

The potential advantage of the developed system lies on the possibility to simultaneously monitor gas and liquid phases by mass spectrometry. However, the sensi-

tivity of the measurements obtained by mass spectrometry for dissolved oxygen was much lower than that obtained with an oxygen electrode. Thus, current research is addressing mass transfer characterisation and assessment of the influence of the available membrane area in an attempt to optimise the response obtained with this interface.

Such a device may become very useful in the monitoring of volatiles, both in fermentation and biotransformation processes, avoiding the need for the development of specific electrodes.

Acknowledgements

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Kontinuirana kontrola otopljenih plinova preko ugrađene mikroporozne membrane masenom spektrometrijom

Sažetak

Masena se spektrometrija vrlo često koristi za kontrolu fermentacijskih procesa jer omogućava izravnu analizu protočnih i otopljenih plinova ili hlapljivih sastojaka u fermentacijskoj podlozi.

U ovom je radu, kao umetnuta međufaza, ugrađena mikroporozna hidrofobna membrana od šupljih vlakana. Membrana, smještena u vanjskom optoku, djeluje kao mjesto spajanja tijeka recirkulirajuće tekućine i tijeka plina nosača koji se analizira masenim spektrometrom. Prikazana je ostvarivost tog sustava, a razmotreni su i postupci poboljšanja kako bi se postigla zadovoljavajuća osjetljivost analize, a i brzina odziva na promjene.