Respiratory Physiology & Neurobiology 191 (2014) 67–74

Contents lists available at ScienceDirect

Respiratory Physiology & Neurobiology

journal homepage: www.elsevier.com/locate/resphysiol

A simple breath sampling method in intubated and mechanically ventilated critically ill patients

Lieuwe D.J. Bos[a,b,*], Yuanyue Wang[c], Hans Weda[c], Tamara M.E. Nijsen[c], Anton P.G.E. Janssen[c], Hugo H. Knobel[c], Teunis J. Vink[c], Marcus J. Schultz[a], Peter J. Sterk[b]

[a] Department of Intensive Care, Academic Medical Center – University of Amsterdam, The Netherlands
[b] Department of Respiratory Medicine, Academic Medical Center – University of Amsterdam, The Netherlands
[c] Philips Research, Eindhoven, The Netherlands

ARTICLE INFO

Article history:
Accepted 6 November 2013

Keywords:
Exhaled breath
Volatile organic compounds
Intensive care
Methodology

ABSTRACT

Volatile organic compounds (VOCs) in breath may serve as biomarkers of pulmonary infection or inflammation. We developed and validated a new breath sampling method for VOC analysis in ventilated patients.

Breath was collected from the ventilatory circuit using cheap disposables. VOCs were identified by gas-chromatography and mass-spectrometry (GC–MS) at various minute volumes during ventilation of an artificial lung (in vitro) and ventilated patients (in vivo).

Sixty-four VOCs emendated from the ventilator and tubing. Their concentrations had an inverse correlation with minute volume in *in vitro* experiments (median correlation coefficient: −0.61 [25–75th percentile: −0.66 to −0.43]). Forty-four of these "ventilator-associated VOCs" were also observed in vivo, without correlations with minute volume. In vivo experiments showed that only positive end-expiratory pressure influenced the concentration of breath VOCs. The sampling method was highly reproducible (median intra-class correlation 0.95 [25–75th percentile: 0.87–0.97]).

In conclusion, a novel, simple and repeatable sampling method was developed and validated for capturing exhaled VOCs in ventilated patients, which could allow for large-scale breath analysis in clinical studies.

1. Introduction

Analysis of expired air may be an attractive approach in disease recognition as breath contains metabolites from both pulmonary and systemic pathophysiologic processes (Moser et al., 2005). Intubated and mechanically ventilated patients are prone to pulmonary infection and injury (Chastre and Fagon, 2002; Slutsky, 1999; Ware and Matthay, 2000). Both attribute to changes in volatile organic compounds that can be detected in the exhaled breath (Aksenov et al., 2012; Bos et al., 2012, 2013a, 2013b; Fens et al., 2011; Hockstein et al., 2005). Recently, we performed an animal experiment in which we showed that exhaled breath analysis can be used to monitor the development of lung injury in rats that were infused with lipopolysaccharide (Bos et al., 2013a). Therefore, exhaled breath analysis could allow for real-time bedside diagnosis in critically ill ICU-patients.

Gas-chromatography mass-spectrometry (GC–MS) is gold standard for VOC separation, quantification and identification (Boots et al., 2012; Hübschmann, 2008). However, GC–MS is not available at the bedside. Advancements in sensor technology and rapid mass-spectrometry may allow for a bedside test and even continuous monitoring of ventilated patients in the near future (Bos et al., 2011, 2013a; Dolch et al., 2008). To facilitate clinical application of these analytical techniques, sample collection should be adapted for bedside use as well. Currently, sample collection from the ventilator circuit is difficult, requiring usage of non-disposable materials, such as sensors, mass flow controllers, tedlar bags and glass syringes (Filipiak et al., 2012; Miekisch et al., 2010). Alternatively, breath can be manually extracted from the ventilator circuit in a glass-syringe but this is prone to errors, labor intensive and cannot be combined with continuous analysis (Birken et al., 2006; Filipiak et al., 2012). It is also possible to obtain alveolar air through a suction tube (Shih et al., 2009). This method results in loss of airway pressure, which can induce atelectasis and subsequent

* Corresponding author at: Department of Intensive Care and Department of Respiratory Medicine, Academic Medical Center, Meibergdreef 9, G3-228, 1105 AZ Amsterdam, The Netherlands. Tel.: +31 205 666 345.
E-mail addresses: 1.d.bos@amc.uva.nl, lieuwe.bos@gmail.com (L.D.J. Bos), yuanyue.wang@philips.com (Y. Wang), hans.weda@philips.com (H. Weda), tamara.nijsen@philips.com (T.M.E. Nijsen), apge.janssen@philips.com (A.P.G.E. Janssen), hugo.knobel@philips.com (H.H. Knobel), anton.vink@philips.com (T.J. Vink), marcus.j.schultz@gmail.com (M.J. Schultz), p.j.sterk@amc.uva.nl (P.J. Sterk).

1569-9048/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.resp.2013.11.001

respiratory failure (Maggiore et al., 2003). Thus sample collection needs to be simplified with the use of disposable materials to allow for large clinical trials using novel analytical techniques.

In contrast to the controlled setting of an animal experiment, several confounding factors should be considered for sample collection in ventilated patients: contamination with VOCs from ventilator and tubing, the influence of ventilator settings (as these cannot be set to standard values due to the severity of illness) and repeatability. Therefore, the aim of this study was to develop and validate a simple method to sample VOCs in the breath of critically ill intubated and mechanically ventilated ICU-patients. We hypothesized that this method (a) can exclude contaminants from inspired air, ventilator or tubing, (b) account for the influences of the ventilator settings and (c) is repeatable.

2. Methods

2.1. Design and subjects

The study had three parts. First, we performed an *in vitro* study of contaminating VOCs in the inspired air. Second, we examined VOCs coming from the ventilatory circuit at various ventilator settings in the absence of a patient. And third, a study (*in vivo*) was carried out of VOCs in the circuit during mechanical ventilation of intubated patients at the ICU of the Academic Medical Center, Amsterdam, The Netherlands. This 40-beds ICU serves as tertiary reference for both medical and surgical patients. Exhaled air from consecutive newly intubated ICU-patients with the exception of postoperative patients after cardiac surgery was collected and analyzed once per day during the first three days of admittance or until death or discharge. A waiver of informed consent was obtained from the Institutional Review Board (IRB: 10.17.0729). This trial was registered at the Dutch trial register (NTR 2750, www.trialregister.nl).

2.2. Sampling method

For clinical care, the ventilation circuit was set up as shown in Fig. 1, and this set-up was not changed for the purpose of the study, besides the insertion of a side-stream connector. A co-axial tubing system was connected (Universal F2 breathing circuit, Medical product service GmbH, Braunfis, Germany) to a mechanical ventilator (Galileo ventilator, Hamilton, Bonaduz, Switzerland or Servo ventilator, Maquet, Rastatt, Germany) and a heat-and-moister exchanger (HME, Medisize, Hillegom, The Netherlands) was placed at the end. For exhaled breath collection, a T-piece connector (T-piece; 22M/22F with swivel, Medisize, Hillegom, the Netherlands) was placed between the HME and the swivel (Catheter mount, Medisize, Hillegom, The Netherlands). The swivel was connected to the endotracheal tube (Ruschelit safety clear plus, Teleflex medical, Athlone, Ireland), which is in direct contact with the upper part of the lower airways of the patient. To produce a side stream flow, the T-piece was mounted with 50 cm bubbling tube (Bubble tubing PHS3/30G 3 mm × 5 mm 30 m, Medisize, Vantaa, Finland), which was locked with a three-way stop-cock before insertion into the ventilatory circuit. Air was adsorbed on a stainless steel tube (6 mm O.D × 7 in., Supelco, Zwijndrecht, The Netherlands) filled with Tenax GR (250 mg/tube, Varian Chrompack, Middelburg, The Netherlands) using a fixed flow of 200 mL/min for 10 min. Two liters of mixed air corresponds to 1.3 L of expiratory air, after adjustment for an inspiratory:expiratory-ratio of 1:2. The flow was generated by a membrane pump (XaviTech V200 GAS 3.2-26V DC, Harnosand, Sweden) and controlled by means of a flow controller (MEMS

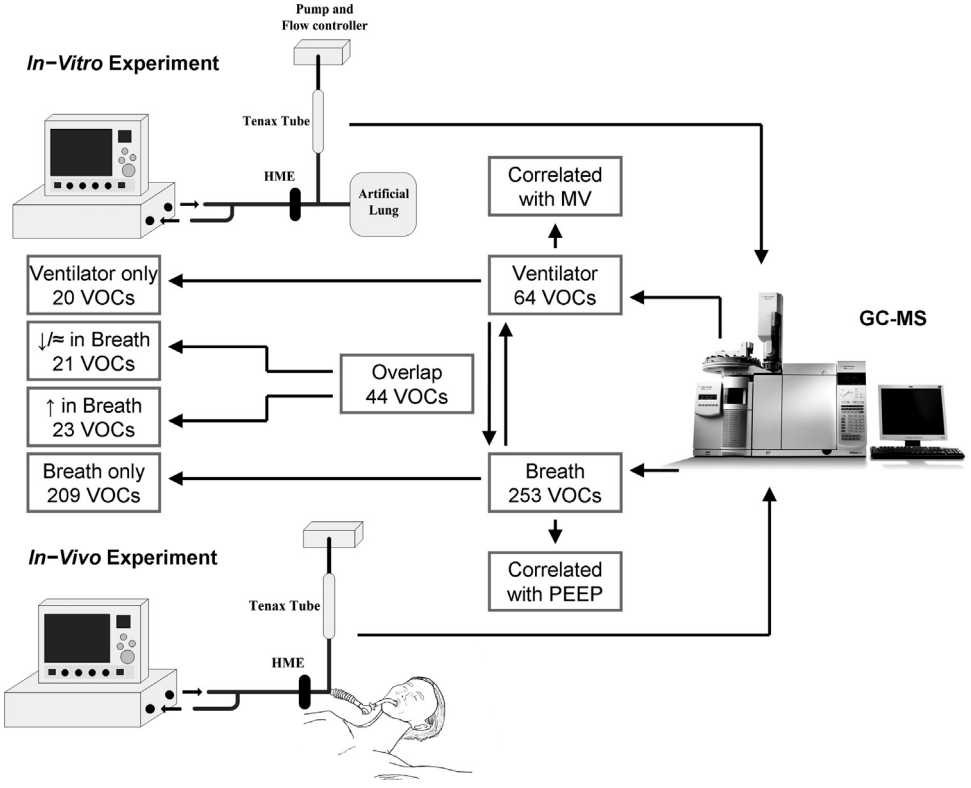

Fig. 1. Schematic representation of the sampling method. Air is collected through a T-piece distal of the endotracheal tube, proximal of the heat and moisture exchanger. To test the VOCs released by the system, a Tedlar bag was connected for air collection. 2 L of air is collected onto a Tenax tube with a flow of 200 mL/min for GC–MS analysis. To test the influence of positive end-expiratory pressure, minute ventilation and maximal airway pressure, an artificial lung was connected to the tubing. The side stream connector and tubing was also investigated separately. The typical flow (minute ventilation) in the ventilator, tubing and endotracheal tube is 4–17 L/min while the flow in the side stream connector to the tenax tube is always 200 mL/min.