

# Enhancing Electronic Nose Performance in Clinical Breath Analysis through Optimized Sampling Design

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**Abstract**— Electronic noses (e-noses) have shown promise for non-invasive lung cancer screening through the analysis of volatile organic compounds (VOCs) in exhaled breath, yet clinical translation remains limited by pre-analytical variability. This study investigates the influence of flexible polymer components, pumping strategies, and sampling bag reuse on VOC retention and contamination. It focuses on sampling-induced variability, rather than solely on sensor performance or data analysis. Controlled experiments using thermal desorption gas chromatography-mass spectrometry (TD-GCMS) evaluated VOC retention and residual contamination are presented. Results show that membrane pumps significantly increase variability and that bag washing cannot reliably eliminate contamination at low concentrations. These findings support the use of single-use bags, minimized storage times, and pump-free transfer to improve robustness of clinical e-nose breath sampling workflows.

**Keywords**—E-Nose, Sampling, Breath, Lung Cancer, Metal Oxide.

## I. INTRODUCTION

Early detection is the principal determinant of survival in lung cancer [1]; however, current screening modalities remain imperfect, resource-intensive, and not universally accessible. Exhaled breath analysis with electronic noses promises a non-invasive, rapid and potentially low-cost screening approach by using pattern recognition of volatile organic compound (VOC) mixtures emitted in breath [2]. Over the last two decades a variety of clinical e-nose studies have demonstrated diagnostic potential across respiratory and systemic diseases [3], yet real clinical use has been limited.

Sampling strategies in clinical e-nose workflows typically follow one of three paradigms: direct (on-line) sampling into the sensor system [4], off-line bag collection (e.g., Tedlar®, Nalophan®, FlexFoil®, FEP/PTFE) with delayed analysis [5], or active sorbent-tube enrichment followed by thermal desorption [6]. Each approach presents trade-offs. On-line sampling minimizes storage artefacts but is not always easy to integrate with some sensor technologies due to pressure and flow changes, which often leads to designs where the sensor chamber is flushed with the sample and then closed while the sensors reach a stable conductance [7]. Bag-based approaches facilitate decentralized collection and are technically easier to work with [8], yet polymer walls can emit background contaminants, adsorb VOCs and induce analyte permeation—effects that depend strongly on bag material, conditioning, temperature, and storage time [9-10]. Sorption methods provide enrichment for trace analytes but introduce variables related to sorbent chemistry and thermal desorption that modify the contents of the sample.

Several recurrent technical problems emerge from the literature and our own experiments: (1) loss or alteration of breath VOCs due to adsorption or emission from materials including pumps, seals, sampling bags and other wet parts

(i.e. in contact with sample/reference gas) [10,11], (2) condensation on cold spots resulting in polar VOC extraction in liquid phase [12], (3) contaminations from background air [13].

Here, we propose an optimized prototype architecture tailored for early lung cancer screening. This paper reports on the rationale for these choices, the prototype design, and part of the experimental data that motivated implementation.

## II. MATERIAL AND METHODS

### A. The prototype BRIDGE

BRIDGE is a transportable breath analysis prototype (Fig. 1). Since it works in cycles (alternating samples and reference air), it comprises a breath pathway and a reference-air pathway with VOC filtration using room-temperature activated charcoal followed by a heated humidifier. Canister air can also be used as reference air. Both pathways converge at a solenoid valve that selects the correct flow for the sensor chamber, which incorporates several metal-oxide (MOX) gas sensors and pressure, temperature, and humidity sensors.

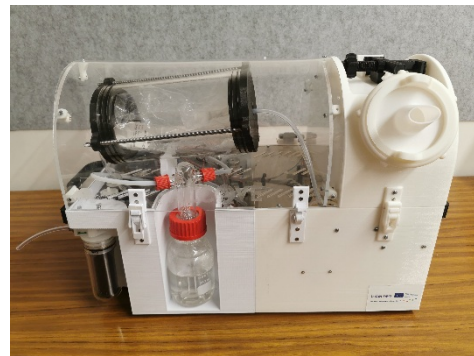


Fig. 1. Photography of the BRIDGE prototype (patent pending. Patent number EP 25217181).

The sampled patient breaths normally through a single-use conventional spirometer filter inserted into a heated sampling tube (40 °C). CO<sub>2</sub> concentration is continuously monitored with an adaptative capnography system that triggers the sampling of the alveolar breath in a FEP bag. An airtight chamber (i.e. “pressure chamber”), kept at negative pressure, contains the FEP bag and is opened to sample the alveolar breath. The sample tube is made so that no obstruction can occur, and no room air may be drawn into the sample bag. Once the bag is filled, the pressure chamber returns to atmospheric pressure, and a peristaltic pump sucks the sample to the sensor chamber. Between measurements, the system is flushed with filtered room air to prevent cross-contamination. The sampling bag is replaced between patients.

The sensor chamber consists of a compact cylindrical PTFE cell accommodating several MOx sensors arranged

radially to ensure uniform exposure and special design features to ensure minimal dead volume.

A startup time of an hour was set to let the sensors reach a stable signal and to set the whole system to a stable temperature. Signals are digitized on an integrated electronics board and transmitted to an Android-based interface. Data are displayed in real time, stored locally and retrieved via removable storage for subsequent preprocessing and pattern-recognition analysis.

The device chassis provided tool-free access to the spirometry filter, sampling tube, data storage, sampling bag, humidifier and charcoal filter for routine inspection, replacement or cleaning and disinfection.

The interface is automated for easy clinical use but allows for complete manual control if needed. The interface gives cues to follow a comfortable and regular breathing pattern.

All characteristics were chosen to obtain increased reproducibility of breath sampling, increase quality of sensor data and resulting “breath prints” (i.e. combined sensor data giving info on breath). Relative humidity, initially at 46% under reference air in the sensor chamber controlled at 40°C, increases slightly to 50% during breath exposure. This ensures that the sensor response is due to the composition of the breath rather than variations in humidity or temperature.

The system operated at low voltage (12 V DC) for transportable clinical use.

#### *B. Test on retention of breath VOCs by flexible polymers*

At an early stage of prototype development, concerns were raised regarding the potential influence of individual components of the BRIDGE on the retention and subsequent release of contaminating volatile organic compounds (VOCs). Membrane pumps generate flow through the deformation of elastomeric materials; in the present case, the RS® diaphragm pump incorporates a fluorinated rubber (FKM) membrane.

A gaseous mixture composed of pentanone (0.5 ppmv), heptanal (0.4 ppmv), and n-decane (0.2 ppmv) diluted in analytical-grade air was employed. The mixture creation technique and choice of compounds is detailed in previous publications [14]. The mixture was initially contained in a FEP sample bag, transferred to a second sample bag, and subsequently analysed by thermal desorption–gas chromatography–mass spectrometry (TD-GC-MS).

In a first configuration, sample transfer was achieved using an in-path membrane pump. In a second configuration, the membrane pump was removed from the sample path and positive pressure was generated outside of the first bag by a pressure-chamber.

Repeatability was evaluated by preparing five consecutive samples within a single day, with this procedure replicated on three separate days over two consecutive weeks. Each measurement consisted of drawing a fixed sample volume of 1 L from the second sample bag onto a Tenax® TA sorbent cartridge using a calibrated sampling pump (GilAir Plus; Gilian®). The time elapsed between bag filling and analysis was usually never longer than 2 hours.

#### *C. Test on contamination from bag reuse after cleaning*

The bag washing procedure was conducted as follows. Each sampling bag was first emptied of its residual contents using a pump, after which it was filled with clean analytical-grade air at 40% relative humidity (RH) and placed in an

oven maintained at 60 °C for 30 min. This emptying and refilling cycle was repeated three consecutive times, followed by a final evacuation of the bag contents.

Following washing, the bags were filled with analytical-grade air and conditioned at 60 °C for 30 min. They were then allowed to cool at ambient laboratory temperature for 5 min prior to sampling. A fixed volume of 1 L was subsequently adsorbed onto Tenax® TA cartridges for analysis.

A total of five bags, which had previously contained a mixture of pentanone (0.5 ppmv), heptanal (0.4 ppmv), and n-decane (0.2 ppmv) prior to the washing procedure, were analysed using this protocol within a single experimental day.

#### *D. TD-GCMS setup*

The cartridge is desorbed using a TD-100xr (Markes®) thermal desorber. The desorption occurs at 300°C with a trap focussing at -10°C. Mass flow controllers within the TD-100xr allow for a twenty-time split before the trap.

A Trace GC oven (Thermo Fisher Scientific®) with a VOC-specialized capillary column (Rxi®-624Sil MS, 0.25mm ID 1.4µm 60m, Restek®) separates the sample through gas chromatography. The GC oven program is as follows: hold 50°C for 5min, 10°C/min to 180°C, 6°C/min to 205°C, hold 205°C for 5min.

A DSQII mass spectrometer (Thermo Fisher Scientific®) uses electron ionization and a simple quadrupole to obtain both the spectrogram and chromatogram of the sample.

The GCMS setup is calibrated using three concentrations of the target VOCs diluted in methanol in three replications, by spiking Tenax®-TA cartridges using a Calibration Solution Loading-Rig (Markes®). All chemicals were acquired from Fisher Scientific® Belgium. More information can be found in [14].

### III. RESULTS AND DISCUSSION

#### *A. Developement of BRIDGE and intended testing*

The BRIDGE is intended to be evaluated in hospital settings as part of a multicentric clinical trial conducted within the framework of the ongoing ALCOVE project (Interreg FWV). The intended use of the electronic nose is not to be a diagnostic device but rather a screening one. The affordability and ease of use allow for widespread use in small healthcare centres and at proximity practitioners. The device would indicate if people at risk would require more tests and incite them to get checked at the hospital.

Furthermore, the overall performance of the device in a breath-based classification task must be assessed. For this purpose, a previously developed methodology has been used [14,15]. The resulting classification metrics will subsequently be compared with those obtained during the clinical trial. At the time of writing, these validation studies are ongoing, and the clinical trial is scheduled to commence in mid-2026.

#### *B. Test on retention of breath VOCs by flexible polymers*

For the configuration incorporating an in-line membrane pump, the overall relative standard deviation (RSD) was approximately 12% for pentanone, 22% for heptanal, and 14% for n-decane (Fig. 2). In contrast, when the pump was removed from the sampling pathway, the RSD values

decreased to 4.5% for pentanone and 10.7% for heptanal, indicating a marked improvement in analytical precision for these compounds. However, the RSD for n-decane increased to 17.8% under pump-free conditions. This lack of improvement suggests that the observed imprecision for n-decane is governed by additional factors independent of the presence of elastomeric pump components.

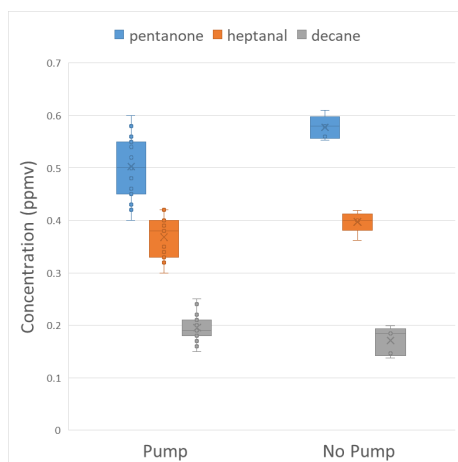


Fig. 2. Box-plot for concentrations of pentanone, heptanal and decane following transfer of sample between bags using either a pump (left) or a pressure chamber (right).

### C. Test on contamination from bag reuse after cleaning

The bag washing procedure exhibited substantial variability in its effectiveness (Fig. 3). Relative standard deviations (RSD) of the obtained peak areas were 114.6%, 92.2%, and 123.8% for pentanone, heptanal, and n-decane, respectively. In terms of absolute concentrations, the residues ranged from 1 to 11 ppb for pentanone, 5 to 22 ppb for heptanal, and 4 to 19 ppb for n-decane. Although these concentrations are relatively low, they may be significant when analysing mixtures containing biomarkers at ppb-level concentrations.

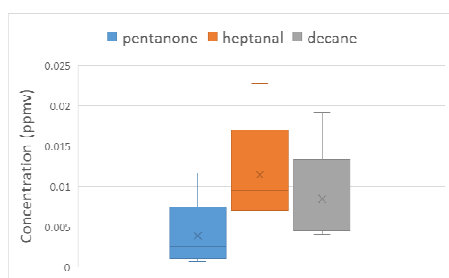


Fig. 3. Concentrations of pentanone, heptanal and decane obtained within gas sampling bags after a complete washing process.

## IV. CONCLUSION

This study addressed sources of pre-analytical variability that limit the repeatability and performance of electronic nose breath analysis, with a specific focus on early lung cancer screening. In this article, we presented a few experiments that oriented our choices in the design of BRIDGE. Experiments have shown that flexible polymer components introduce significant adsorption-desorption artefacts that increase variability over time. The use of a pressure-controlled sampling chamber effectively removed elastomeric elements from the sample path and improved repeatability for several analytes. Additional tests confirmed that FEP gas sampling bags washing procedures cannot

reliably eliminate residual contamination at low concentration levels.

The conclusions of the tests, among others, were used for deciding on the design choices of the BRIDGE.

Future work will focus on validating this architecture in a multi-centric clinical trial and on quantifying how these design improvements translate into diagnostic performance under real-world conditions.

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