High-throughput breath screening, using a portable selected ion flow tube mass spectrometer (SIFT-MS)

Introduction

Real time detection of volatile biomarkers in individual members of test populations could be useful for applications such as disease screening, security screening, monitoring of exposure to infectious agents or occupational hazards, and detection of substance abuse. Here we demonstrate the use of mobile SIFT-MS technology for high throughput breath testing, including real time simultaneous measurement of seven breath trace gases in 230 school children aged 5 to 13 years[1].

SIFT-MS as a breath analysis tool

SIFT-MS has several features that make it an effective breath testing technology, including accurate real time measurement and analysis that is easy for technicians to operate and subjects to use.

SIFT-MS allows real-time quantification of volatile compounds regardless of the sample's humidity. Additionally, absolute concentrations of trace gases in breath are determined by SIFT-MS without sample preparation, pre-concentration or analyte calibration.

A breath screening study

For the duration of this proof of concept investigation a Voice200® (Syft Technologies Ltd, Christchurch, New Zealand) instrument was deployed in a van, connected to standard 10 amp, 240 volt single phase power and used continuously for three four-hour sessions to conduct breath tests on 230 children at two sites.

Demographic data including height and weight were collected prior to breath testing. The children's ages ranged between 5 and 13 years. Table 1 summarizes the population demographics.

Table 1. Student population demographic data.

				Mean		
Age (years)	Number	Male	Female	Height (m)	Weight (kg)	BMI
5-6	29	14	15	1.16	24.8	18.2
7-8	65	34	31	1.27	32.0	19.6
9-10	70	34	36	1.38	39.3	20.4
11-12	54	30	25	1.51	52.0	22.6
13	11	7	4	1.54	55.0	22.5
Total	230	119	111			

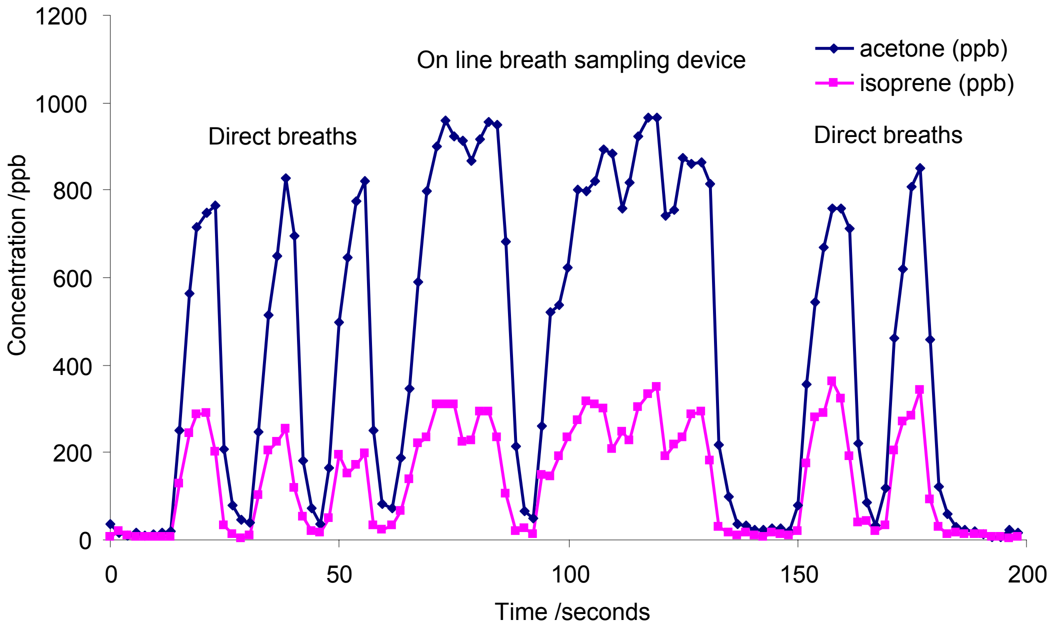
[1.] Ethics approval for this study was granted by the New Zealand Health and Disability Upper South A Regional Ethics Committee and parental informed, written consent for each participating child was obtained. Syft Technologies acknowledges the support of Aranui Community Trust Incorporated Society who provided community, cultural and school liaison. This study was financially supported by the New Zealand Ministry for Science and Innovation through a Research for Industry contract (SYFT0401). Syft also wishes to thank the staff, parents and children of the Aranui and St James Primary Schools for their support and willing participation.

A continuous breath stream of at least 10 seconds is required for accurate quantification of volatile organic compounds (VOCs) by SIFT-MS. However a single exhalation of this length is difficult to achieve in children. For this study a disposable plastic "T" tube mouthpiece was used[2]. It was attached to a disposable hydrophobic PTFE filter placed between the mouthpiece and the SIFT-MS sample inlet. Two one way valves were fitted to the T-piece so that ambient air inhaled through the nose could be "normally" exhaled from the mouth into the device through the sidearm.

Excess exhaled breath escaped from the mixing chamber through the valve opposite the SIFT-MS sample inlet with negligible flow restriction. As opposed to the concentration peaks and troughs of repeated tidal breath exhalations directly into the Voice200® sample inlet, the sampling device provides a continuous breath stream so that stable concentrations of breath analytes are measured (Figure 1). The mouthpiece and filter were changed for each participant.

To facilitate high throughput, the children were standing during testing (Figure 2). The flexible heated inlet of the Voice200® allowed the mouthpiece to be positioned at a comfortable height for each child.

Figure 1. Breath acetone and isoprene concentrations measured in real time from (a) five direct breaths, three at the commencement and two at the termination of testing and (b) using the disposable breath sampling device for two separate, sequential sampling times of approximately 30 and 40 seconds in the middle of the sampling session.



[2] The mouthpiece described does not eliminate the contribution of mouth and nasal cavity bacteria and enzymes to breath analytes such as ammonia. Also, because the plastic breath sampling device is used at ambient temperatures, it is prone to condensation and potential analyte adsorption. The contribution of these sources of interference must be thoroughly understood before a VOC can reliably be used in disease diagnosis or monitoring. Syft recommends the use of the Breath Analysis Inlet for all breath analysis investigations. Such an inlet was not available at the time of this study.

Figure 2. A child pictured breathing into the inlet of a Voice200® instrument.

Although not intended for diagnostic use in this case, the VOCs measured were selected as they are related to metabolic processes, and occur at a range of concentrations measurable by SIFT-MS. Acetone is produced by hepatocytes from decarboxylation of acetoacetate resulting from lipolysis or lipid peroxidation. Ammonia is associated with both endogenous protein and nitrogenous compound systemic metabolism as well as bacterial and enzyme activities in the mouth and nasal cavities. Isoprene is an early by-product of the mevalonate pathway sterol synthesis. These three compounds as well as ethanol, a product of gastrointestinal flora fermentation, exist at readily measured levels (50-2000 ppb) in the breath of all healthy normal individuals.

At lower concentrations (between 1-100 ppb) other compounds of interest included trimethylamine, increased levels of which have been associated with renal failure as well as abnormal metabolism, carbonyl sulfide (associated with cystic fibrosis and acute lung transplant rejection) and pentane, which is produced as a result of oxidative tissue damage.

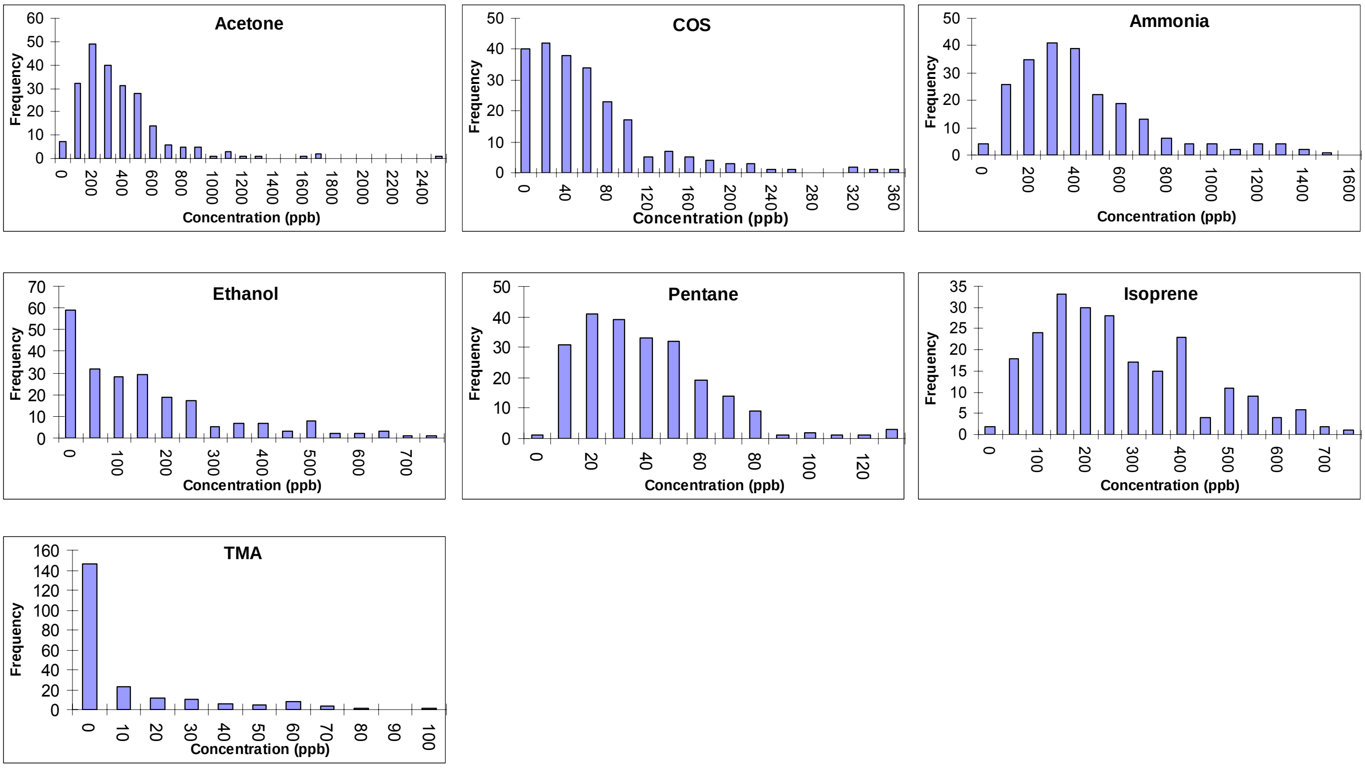
Table 2 lists breath trace gas concentrations and their range within the test population. The breath testing procedure was brief and easy for primary school children to perform. In addition to the advantage of portability, the Voice200® had sufficient sensitivity to measure simultaneously, breath trimethylamine at concentrations below 10 ppb, and acetone at higher concentrations.

Table 2. Selected breath analytes measured in 230 children (5 to 13 years old). For frequency distributions, see figure 3.

Breath analyte	Median (ppb)[A]	Interquartile range (ppb)
Ammonia	299	157-483
Acetone	254	141-431
Isoprene	219	118-354
Pentane	29	13-47
COS	34	5-68
Ethanol	68	nd-186
TMA	nd	nd-6

[A] nd = concentration below quantifiable limit.

Figure 3. Frequency distributions of breath analytes in 230 children (5 to 13 years old).



Summary

The number of children tested in this preliminary study was consistent with many population testing scenarios, but no attempt was made to maximise the throughput. Refinements to the breath sampling methods would further improve throughput.

The use of SIFT-MS allows a faster and wider range of analytes to be analysed compared to other breath analysis techniques. For GC-MS, sample collection is followed by preconcentration and lengthy analysis. For sensor technologies, sensitivities may be lower and quantitative data may be difficult to extract. SIFT-MS breath testing also offers numerous advantages when compared to traditional medical laboratory screening techniques for blood or urine, including a lower demand on technical experts and non-invasive sampling procedures.

Real-time analysis of breath for volatile marker compounds using a mobile SIFT-MS instrument allows rapid population screening that is not possible with traditional methodologies. This capability offers significant opportunities for rapid screening of at risk populations, particularly where speed of analysis and quick and clear results presentation are desirable. SIFT-MS breath screening can cover a range of applications, such as screening for infectious agents and disease, monitoring of exposure to occupational and environmental hazards, and detection of substance abuse.