WHITEPAPER

Breath Biopsy® Technology

Optimized tools for the reliable sampling and analysis of breath for research and clinical use

BREATH® BIOPSY

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Abstract

Analysis of biological samples such as blood, urine, and fecal matter is essential in both clinical research and clinical practice. Informative compounds within these sampling mediums are quantified to be used as biomarkers to inform diagnosis, monitor the progression of diseases, assess the safety of trialed novel drug therapies, measure the efficacy of treatments, and more. Breath is a waste product that is continuously produced from the body and is similarly enriched for biologically relevant compounds that could serve as non-invasive biomarkers; however, to date, there have only been a limited number of breath biomarkers translated for clinical use. This is primarily due to the wide range of different collection and analytical procedures utilized, without much standardization between methods for validating research findings across the field.

Breath biomarkers could provide substantial clinical benefit, and Owlstone Medical has spent many years developing optimized products and services to support the highest quality breath research. This includes tools to collect a high-quality breath sample with technical replicates, exclude contaminating background compounds and pre-concentrate breath samples, and quantitative analytical procedures to reliably distinguish “on-breath” versus “off-breath” compounds, provide robust and repeatable chemical identification, and more.

Key Points

- There is significant unmet needs in clinical research and practice that non-invasive breath biomarkers could address
- Breath collection and analysis has historically suffered from standardization issues, limiting the translation of breath biomarkers into the clinic
- Owlstone Medical’s Breath Biopsy technologies have been specifically designed with the challenges of breath in mind, to provide a robust and reliable platform to collect, analyze, and interpret breath data results for improved translation

Introduction

There are still significant unmet needs for many diseases: diagnosis is too often happening too late through a lack of good or appropriate early detection tools and a lack of suitable tests that can predict which treatments would be best, leading to lengthy trial and error. Clinical trials for drug development also currently suffer from many issues, including high cost, lengthy process, and a high failure rate: approximately 90% of candidate drugs that enter into at least phase I trials fail mostly due to a lack of efficacy (1). This could be addressed through better monitoring tools of endpoints that allow for more real-time monitoring for faster decision-making for shorter (and therefore less costly) trials, as well as non-invasive monitoring to improve ethical approval and participant recruitment. Over the past few decades, analysis of the volatile metabolites (otherwise known as volatile organic compounds, or VOCs) in the breath has attracted growing interest for their potential to act as non-invasive biomarkers of many physiological processes in the body for many clinical uses.

The idea that unique patterns of chemicals in breath could be used to diagnose disease is an ancient idea – Hippocrates himself wrote a treatise on breath aroma and disease whereby he attributed diseases to distinctive breath odors. One of these he referred to as “fetor hepaticus”, an unusual breath smell that he associated with liver failure. Although modern breath analysis has come a long way since smelling the breath and gaining medical information this way, this example establishes two crucial principles for breath science: changes in the normal levels of informative compounds in the breath can alert to disease states within the body, and that exhaled breath contains compounds originating from deeper within the body than just the respiratory system. Therefore, it is believed by many that breath could be another platform for medical analysis that could address many of the clinical unmet needs that still exist due to several methodological advantages of breath collection.

The advantages of breath analysis

Breath has many unique qualities that make it attractive for clinical use. One of the most immediately apparent is that breath can be collected conveniently and completely non-invasively. This can increase the uptake of diagnostic tests, especially for potential screening programs. A comfortable mode of sample collection also has benefits beyond just the improved comfort for patients and trial participants, including easier ethical approval during clinical trials and higher uptake for screening programs. Breath is an inexhaustible resource enriched with chemicals produced almost continually by the body. The volume of a single exhaled breath at rest is approximately 400 – 500 ml, with an average of 12 breaths per minute; this results in about 6 L of air being ventilated every minute (2). For an average person, their total blood volume circulates through their lungs approximately every minute (3,4), meaning that a breath sample taken over one minute contains compounds that have originated from the entire body (Figure 1).

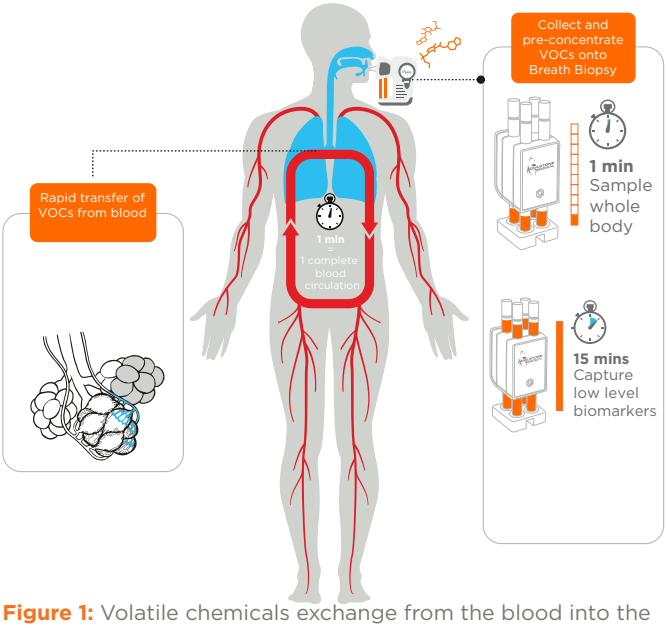

Figure 1: Volatile chemicals exchange from the blood into the lungs. It takes ~1 minute for the total blood volume to cycle around the body, so it is possible to analyze the entire circulating blood volume in a completely non-invasive manner via the breath.

A large volume of breath can be collected at any one time. The volatile compounds in breath samples can also be pre-concentrated before analysis, ensuring the capture of even very low-abundance chemicals. Theoretically, breath sampling can be sampled without spatial restrictions (as there is no need for large equipment), allowing for decentralized trial designs and at-home breath testing for disease monitoring and diagnosis. As the volatile compounds within exhaled breath are products of metabolic activity, measuring them in the breath can provide more immediate feedback on physiological processes compared to protein levels or genetic changes as seen in cancer. The time lag between bacteria in the gastrointestinal tract producing hydrogen, and detectable levels appearing in breath has been measured as five minutes or less (5), which demonstrates how closely breath composition reflects ongoing physiological processes. This contrasts with techniques like fecal analysis, which reflect a much larger window of time. Taking repeated breath samples over time can allow for longitudinal, almost real-time information for more in-depth monitoring. The potential power of this cannot be overstated, especially in combination with approaches such as our EVOC® probes.

It is important to note that volatile compounds can be analyzed from the headspace (air surrounding) of *in vitro*, *ex vivo*, and biofluid samples as a complementary analysis alongside breath. These types of setups include sampling the emitted volatile compounds from cell culture, tissue culture, and samples such as urine, fecal matter, and blood. Analysis of these samples allows for greater control over experimental conditions to collect more in-depth, mechanistic data about the production of volatile biomarkers in the disease area of interest, as well as response to drug treatments. Breath can also be collected from *in vivo* animal models through various methodologies, including the use of ventilation devices like the flexiVent®. This can investigate which processes in the body lead to the alteration of certain biomarkers in exhaled breath to inform clinical study, as well as streamline and optimize the validation of biomarkers, and ultimately, translation into the clinic.

The challenges of breath analysis

However, breath also has unique challenges that must be overcome to translate more breath-based biomarkers into the clinic. Some of these challenges are not unique to breath: a very small signal needs to be detected, and often specialist equipment is needed. Others are specific to breath and require innovation in breath research, this includes that breath is unlike other biological sampling mediums in that it is comprised of both compounds that have arisen from the body and exchanged into the breath at the alveolar surface in the lungs, and those that have been inhaled from the environment.

As volatile compounds are typically present in low abundance, reliable collection and accurate detection are essential to successfully identify candidate biomarkers. Despite the different methods available for breath research, it is a historical problem of the field that data are not always consistent and reproducible. Standardizing best practices in breath research is a key factor to establish before breath testing can find success in the clinic. This includes addressing the unique dual nature of breath with both exogenous and endogenous compounds by using filtered air for breath sample collection which can minimize environmental contaminants, and through utilizing “Breath or Blank” (BoB) study designs, whereby blank samples of the ambient air where sampling is taking place are collected to represent the roster of local contaminants. By comparing breath and blank samples, it allows for volatile compounds truly originating from the breath to be distinguished from those contaminating the sample from the background. Our award-winning Breath Biopsy® products and services have been developed with these strengths and challenges of breath in mind.

Breath Biopsy Technology

The ReCIVA® Breath Sampler

Figure 2: The ReCIVA Breath Sampler device.

To assist the collection of breath for research studies, Owlstone Medical has worked with leading experts in the field and developed a standardized device - the ReCIVA Breath Sampler. The ReCIVA pre-concentrates breath samples on adsorbent tubes, enabling a larger collection of air volume and offering the potential for greater sensitivity in detecting low-abundance compounds. The pre-concentrated breath samples remain stable for longer periods of time at room temperature, and the small size allows for easier transportation and storage.

Whatever your area of research interest, the ReCIVA was designed to be as versatile as possible while still providing quality breath sample collection. The ReCIVA simultaneously collects replicate samples contributing to the reliability and the robustness of the analytical results. The ReCIVA is compatible with our consumable Breath Biopsy Cartridges that includes four independent adsorbent tubes to fit inside the device for VOC collection. Tubes can be combined to enhance sensitivity or analyzed separately as replicate samples. The ReCIVA directly captures and pre-concentrates VOCs in breath onto the Breath Biopsy Cartridge. The direct extraction of VOCs from atmospheric gases means samples can be greatly enriched for VOCs on breath, even those at low abundance, simply by collecting samples for longer. This pre-concentration means that Breath Biopsy can offer higher sensitivity for trace VOCs, it also means Breath Biopsy samples are more robust during shipping or storage before sample analysis.

The volume of breath collected in studies usually varies widely, ranging from 20ml bottles (6) to 1-liter bags (7). This, coupled with the different breath fractions being collected (i.e. from the upper respiratory tract versus from deeper in the lungs), can significantly impact the results of studies. Measuring a fixed amount of collected breath volume consistently and capturing breath in its stable form with specified fractions all provide benefits for downstream processing and analysis of samples.

Figure 3: A demonstration of the ReCIVA Breath Sampler device in use with the coupled nose clip. In certain circumstances a mask can be used instead.

There are several benefits of using ReCIVA over other breath collection techniques. A common issue of other breath collection methods is lack of consistency in the volume of breath collected. Using the ReCIVA the volume of breath collected can be controlled, and therefore made consistent between patient samples. Alongside our CASPER Portable Air Supply, contaminants from the background air can be removed, reducing the risk of false discovery candidate breath biomarkers. Another common issue with breath collection (particularly when using Tedlar bags) is the condensation of breath on the surfaces, meaning that VOCs may be present in droplets and not captured by detection equipment. This is not an issue with the ReCIVA device as hydrophobic sorbent materials can be used in the sorbent tubes to limit water collection on the sample. The water content of a sample can be further reduced by dry purging of the tubes after sample collection, ensuring the maximum capture of biologically relevant compounds.

Breath collection using the ReCIVA device is simple. Typically, 1.25 litres of breath are collected onto each sorbent tube – with the analysis is completed with 2.5 liters: 2 tubes for analysis and 2 tubes for backup. This makes a total of 5 litres of breath collected, although this volume can be adjusted. To collect the full 5 litres it takes approximately 12-15 minutes of normal breathing into the device.

The ReCIVA breath sampler has been used successfully as part as many recent breath research

papers (8–11), including one published in Science Translational Medicine assessing those presenting with acute breathlessness accessing emergency care services (12). Several independent reviews have also been published by academics examining the feasibility of using ReCIVA. Harshman et al. concluded that their "results illustrate the utility of the ReCIVA Breath Collector for providing consistent exhaled breath onto adsorbent media, eliminating the pitfalls associated with collecting breath in bags, while gaining comparable results to samples collected by standard breath bags", and that " the ReCIVA sampler represents a significant step towards standardized exhaled breath sampling for off-line analysis and further research is warranted to improve overall exhaled breath sampling" (13). Holden et al. found that it was "possible and acceptable' to use ReCIVA to collect breath at the bedside of both adults and children as young as 5 years old, even while they were reporting acute breathlessness (14). Azim et al. conducted a systematic review of research examining exhaled VOC biomarkers for adult asthma and discussed our ReCIVA device, stating: "The optimal solution needs to balance practicality and precision, so as not to negate the clinical utility of breath sampling. The (ReCIVA) breath sampler developed by a broad consortium of breath researchers and engineers, represents one such solution" (15).

The CASPER® Portable Air Supply

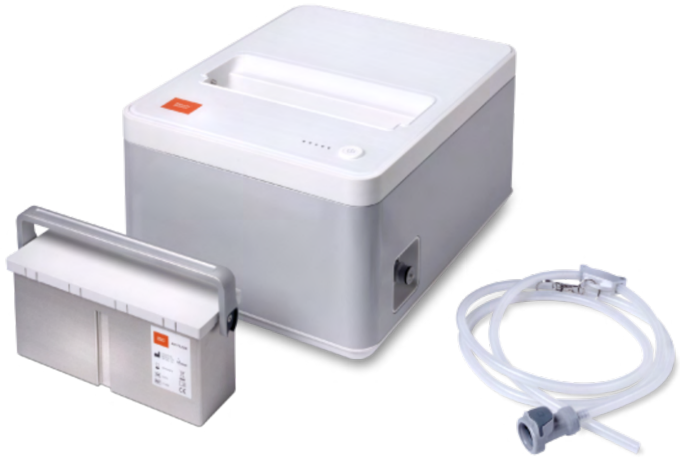

Figure 4: The CASPER Portable Air Supply that can be linked to the ReCIVA Breath Sampler as a source of clean, filtered air into the system.

While internal processes in the body produce a wide range of volatile compounds that can be detected in breath, we also breathe in volatile compounds that are present in the air. Pollution, furnishings, food, cosmetics, perfumes, and cleaning products are all common sources of VOCs in any air sample and have nothing to do with a patient's health. As an additional challenge, the selection of background VOCs is highly dependent on location. This means that collected breath samples often contain VOCs that are more related to location than to subject health and can be one reason for proposed breath biomarkers failing to pass validation. Many studies control for this by collecting all samples in the same

See our **ReCIVA Instructional Video** for a demonstration of using the device to collect a high-quality breath sample.

room, but this isn't a practical solution for large-scale trials or eventual clinical applications.

The abundance of different VOC species in breath can differ by as much as six orders of magnitude. While the most abundant species can be easy to analyze, they can make it harder to accurately detect and identify the rarer ones. Removing background VOCs that originate from the environment reduces the levels of some of the most abundant VOCs and ensures that more of the VOCs collected in each Breath Biopsy sample are relevant to a subject's health and metabolism. Therefore, to make biological and clinical inferences about the composition of breath samples, it is crucial that the number of volatile contaminants originating from the environment in breath samples are minimized. To address this, we have developed the CASPER Portable Air Supply, which can significantly reduce environmental VOCs in breath samples. This is particularly beneficial to control for variability in the ambient air in studies that are collecting breath from multiple different sites (16). The CASPER uses a mains-operated air pump which takes ambient air and passes it through an air filter unit before supplying it via a plastic tube to ReCIVA. The subject breathes in filtered air, reducing the level of background VOCs from the surroundings that are present in the collected sample.

Industrial air cleaners can remove volatile compounds from the air, but producing clean air is a complex and expensive process and the required machinery isn't portable. CASPER provides a more practical, economical and mobile solution while offering comparable reduction in background VOCs for breath collection. We used the Breath Biopsy platform (gas chromatography mass spectrometry - GC-MS) to analyze air produced by CASPER in comparison to industrial clean air and ambient air samples (Figure 5). This data demonstrates that the CASPER removes over 75% of background volatiles, a level of performance that makes it comparable to industrial clean air supplies.

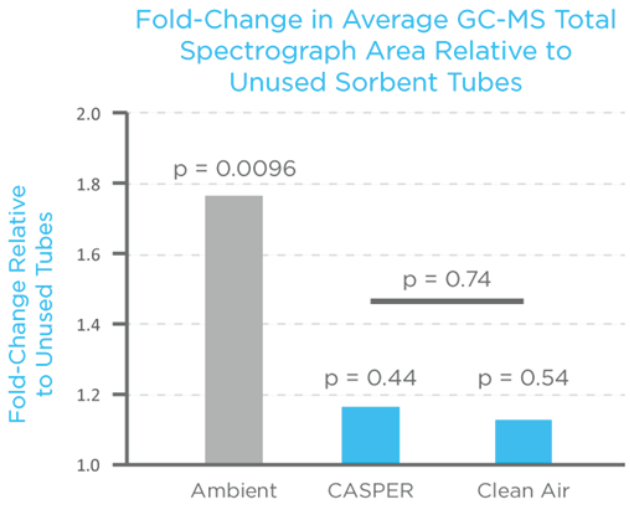

Figure 5: CASPER Portable Air Supply reduces ambient VOCs to an extent comparable to industrial clean air and statistically comparable to unused sorbent tubes. Data derived from total GC-MS spectrograph areas normalised against spectra generated from unused tubes. P-values show results of pairwise t-test comparisons. P-value for comparison to unused tubes are shown directly above each bar.

The CASPER has also been tested by introducing selected volatiles from essential oils into the air. Samples of the air in the room were collected using the ReCIVA Breath Sampler, and after it had been processed by CASPER. The VOCs from both sets of samples were analyzed on the Breath Biopsy platform (Figure 6).

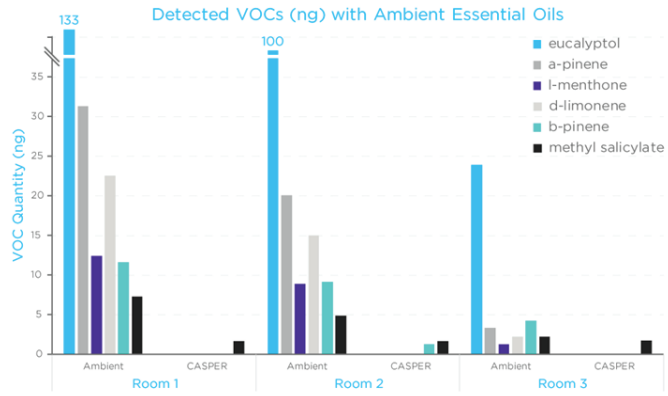

Figure 6: Ambient VOCs collected in rooms spiked with a non-standardized quantity of essential oils, with and without the use of CASPER.

The CASPER unites the airflow pump and filter into one casing, allowing direct connection to the ReCIVA Breath Sampler and making the whole system compact and portable.

The ReCIVA, CASPER, and Laptop with Collect software are available together in the Collection station...

...which includes a lightweight and easy to clean stainless steel trolley to maximize portability.

The Collect Software

The Breath Biopsy Collect software connects to and operates the ReCIVA breath sampler device, enabling you to collect specific breath fractions, monitor patient breathing in real time and gather patient metadata. Through continuously monitoring breathing patterns, the software works with the ReCIVA to effectively exclude air from anatomical dead space in the upper airways, unless required specifically otherwise. The exclusion of dead space air ensures that exhaled breath is primarily derived from lung areas rich in gas exchange, and enriched with volatile metabolites exchanged from the bloodstream. This can reduce the noise from internal VOC sources such as the mouth, upper airways and/or stomach, and ensuring the capture of breath that is enriched for VOCs exchanged into the lungs from the bloodstream that originate from informative physiological processes. The friendly user interface allows enhanced visuals, simple user instructions and easy navigation during the breath sampling process. The software will detect when a full breath sample has been taken, ensuring that a consistent volume of breath is taken across different participants or patients – which is a key variable for standardized and quality breath research.

By adjusting ReCIVA's collection settings with the Collect software interface, you can also collect volatile compounds from specific breath fractions, allowing you to differentially analyze compounds from the deep lungs, upper airways or mouth and throat, depending on your area of interest. This communicates with the ReCIVA's carbon dioxide and pressure sensors to monitor and learn the breath patterns of each subject in real time and only activate the pumps to draw air at the right times to collect the desired breath fraction (Figure 7).

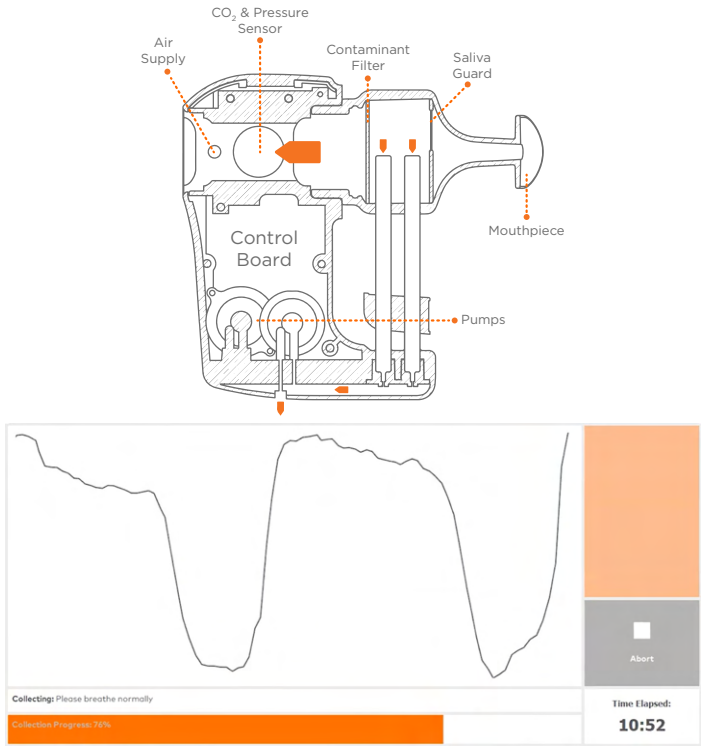

Figure 7: A schematic of the hardware of the ReCIVA breath sampler device, and the Collect software monitoring the breathing of a participant using the inbuilt sensors.

EVOC® Probes

Exogenous VOC (EVOC) probes are compounds designed to be metabolized, excreted, or absorbed by the body to result in a detectable EVOC product in exhaled breath (17). EVOCs are not normally found at significant levels in breath, therefore their specific concentrations in the breath can be more accurately interpreted as being related to the process of interest that is being investigated. This concept draws similarities to how labeling urea with 13 Carbon Urea (^{13}C-Urea) is currently employed as a metabolic probe in clinical settings to evaluate *H. pylori* infection within the stomach (18). The usual test for *H. pylori* is a breath test, whereby a substrate solution containing ^{13}C-Urea is ingested, and a sample of breath collected 30 minutes afterwards. Unlike the cells in the stomach, *H. pylori* is capable of metabolizing urea to produce ammonia and carbon dioxide, therefore presence of a *H. pylori* infection of the stomach is indicated by the levels of these compounds. Using a probe in combination with a breath test can unlock the potential for a diverse array of applications and can be used to target specific pathways of interest and amplify the signal from these pathways for better test performance (Figure 8).

We have recently used this approach with great success to trial limonene as an EVOC probe and dynamic breath test for liver disease (19). Limonene is a compound that falls under the FDA GRAS (Generally Recognized As Safe) list and metabolized by the cytochrome P450 enzyme CYP2C9 and CYP2C19, similar to a wide range of drugs. The rationale behind the use of limonene is that the hepatic abnormalities associated with liver disease can change the systemic bioavailability of compounds that are usually metabolized in the liver, and therefore, monitoring the rate at which a compound is metabolized and excreted from the body can be used to directly monitor the function of the liver, and consequently diagnose liver disease (Figure 9).

A further example of how this approach can be used is to shed light on drug metabolism - the intricate process of how drugs are transformed from prodrugs into their active forms, as well as metabolized for subsequent excretion. It is estimated 40% of prescribed medications fail to produce the anticipated effects, which could be due to insufficient levels of active compounds for therapeutic efficacy or excessive levels resulting in toxic accumulation. Gaining insights into an individual's drug metabolism status will help clinicians make decisions regarding drug selection and dosage.

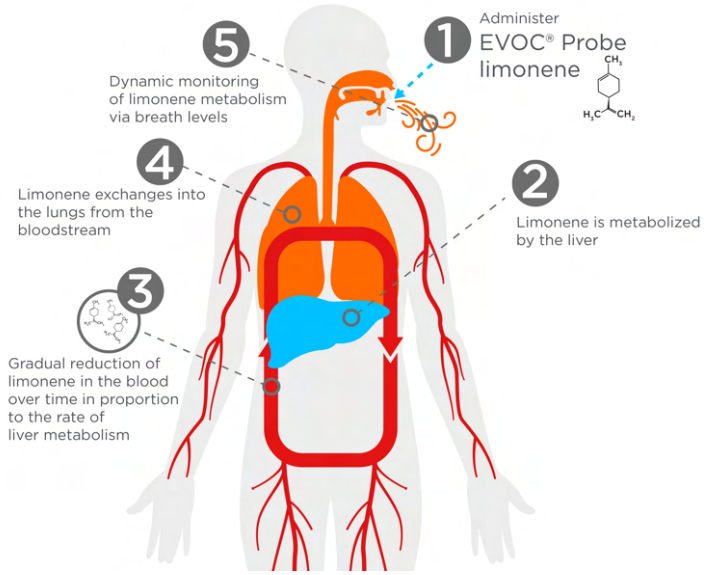

Figure 9: A schematic showing how limonene can be utilized as an EVOC probe to investigate liver function.

Different EVOC probes can be deigned and tailored to fit different study needs, and administered to your participants or cell culture to invoke a response from the biochemical pathway or tissue of interest. Next-generation more complex EVOC probes are also possible, as we have partnered with Bicycle Therapeutics to develop antigen-targeted diagnostic probes that use bicyclic peptides as their targeting mechanism. This collaboration is initially focussing on lung cancer screening as the first proof of principle for the broader opportunity by promoting selective accumulation of the probe at the tumor for increased signal and enhanced specificity.

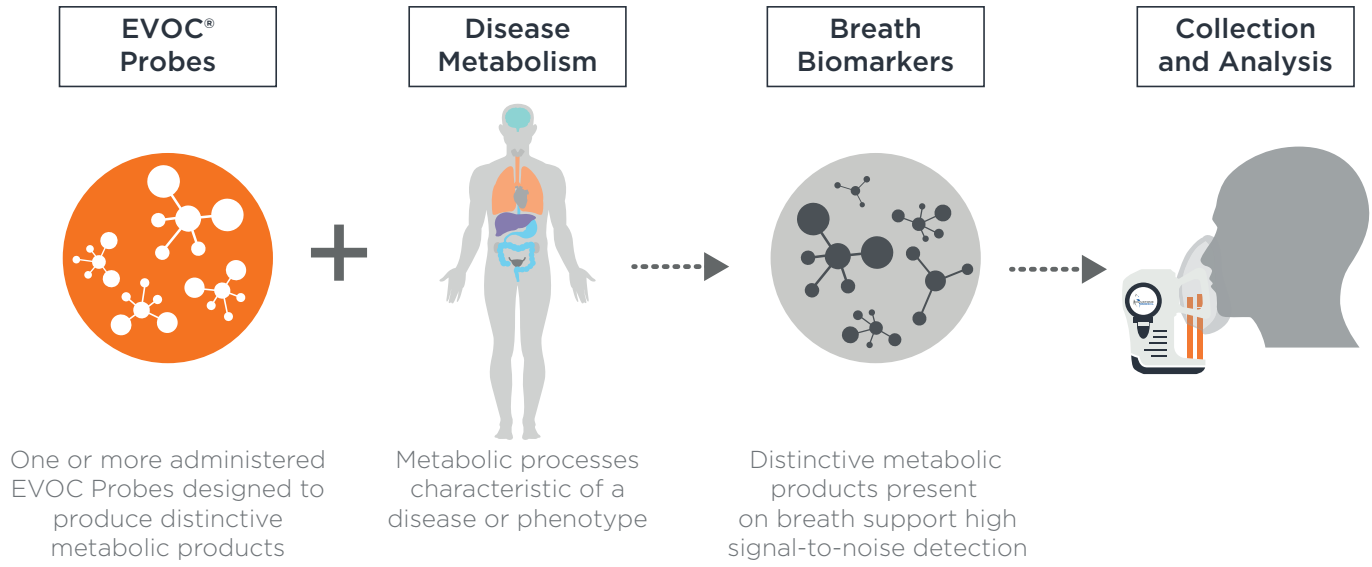

Figure 8: An example workflow of how EVOC probes can be incorporated into breath research studies to target specific metabolic pathways, or disease processes.

BREATH® BIOPSY OMNI

The complete end-to-end breath VOC analysis service

Figure 10: Breath Biopsy OMNI is a complete VOC analysis service from study design, breath collect, to analysis and interpretation.

To assist researchers in the field of breath biomarker discovery within clinical trials, the OMNI (Owlstone Medical Novel Insights) platform was developed. This innovative platform optimizes the identification and quantification of volatile compounds in the breath. The platform encompasses the collection of both breath and blank samples from the ReCIVA, with the blank samples forming a crucial control to significantly reduce the false discovery rates of candidate biomarkers, thereby enhancing the likelihood of uncovering true clinically significant breath biomarkers. Under the current OMNI methodology, a volatile metabolite is deemed an "on-breath" compound only if its concentration in the breath samples is above the concentration in the blank samples by at least three standard deviations.

The OMNI breath analytical process adheres to rigorous quality assurance stages, ensuring consistent and reproducible outcomes. As the analytical process only allows for a limited number of samples within one sequence run, it is crucial to minimize the technical variability from the different sequences run within a study. To ensure high quality results, each sample has an analytical sequence that incorporates over 50 standard compounds, each selected to represent the diversity of relevant compounds while maintaining consistent quantities. Because GC-MS platforms are susceptible to analytical drift, data normalization is required to process the differing outputs for the same input. To correct this variability, eight deuterated standards are included in every sample, facilitating a sample-by-sample evaluation of analytical performance. This comprehensive approach ensures a fit-for-purpose assessment of result quality in each sequence and facilitates the early detection of any issues arising during sample processing.

To ensure more consistent identification of molecular features from mass spectrometry analysis, we employ customized software settings that enhance feature extraction. The OMNI platform correctly detects 98% of signals from standard (volatile) compounds. With optimized feature extraction, the OMNI platform has the capability to detect over 70% more compounds in breath compared to an equivalent out-of-the-box feature extraction process. These improvements mean that more than double the number of prospective breath biomarkers are consistently detected by OMNI, revealing up to 500 VOCs detected in each breath sample.

The volatile metabolites identified through OMNI are categorized into different tiers of confidence, aligning with the standards set forth by the Metabolomics Standard Initiative (MSI). In addition to different tiers, retention index is also calculated to assist with the comparison of results between studies. This meticulous alignment provides high confidence in the identities of the detected volatile metabolites. Currently, we have validated over 200 compounds with standard references in our Breath Biopsy VOC Atlas (discussed in the next section), representing the volatile compounds in breath with the highest level of confidence. Our secondary tier of volatile metabolite identities is established through comparison with our internal HRAM (High-Resolution Accurate Mass) library. If this is not possible, third tier identifications of certain volatile metabolites are matched with the National Institute of Standards and Technology (NIST) library.

The VOC Atlas®

Current technical measurements reveal the presence of over 1000 distinct volatile compounds in exhaled breath. Given the significant variability of volatile compounds among the human population, studies employing untargeted approaches without adequate controls can readily yield a plethora of false positive hits. To address this critical requirement for improving biomarker discovery, Owlstone Medical has developed the Breath Biopsy VOC Atlas, the most extensive dataset of validated breath volatile metabolites.

The VOC Atlas is a continually evolving database, showcasing volatile metabolites commonly detected in the breath of a mixed population. The database comprises clinical studies that incorporate blank (air from the collection system) and breath samples from individuals, ensuring rigorous evaluation of frequently seen "on-breath" samples (as discussed in the preceding section on sample noise reduction). Volatile metabolites found commonly on-breath in these studies undergo validation against standard references to confirm their identities. Currently, the database confirms the presence of over 200 compounds, complete with the ranges of concentrations that they were found at within the heterogeneous cohort. These volatile compounds span multiple chemical classes, and we can provide confirmation to our customers and collaborators regarding the existence of compounds of interest in the database to assist with their research. The VOC Atlas is intended to be shared with our customers and collaborators as a resource in the future to help advance their breath research.

Many of the volatile compounds within our Atlas have been previously documented in the scientific literature. Noteworthy examples are acetone and isoprene, which are well known to be two of the most well-studied, and abundant volatile compounds found in human breath (20,21).

Furthermore, the Atlas highlights several short-chain fatty acids (SCFAs), which arise from the microbial fermentation of dietary fiber. These SCFAs are of particular interest due to their involvement in multiple processes that have significant impacts on human health (22,23). SCFAs associated with the microbiome have been implicated in various health contexts including cancer, neurogenerative disease, and inflammatory bowel disease (24–29). Trimethylamine (TMA), another volatile metabolite linked to the gut microbiome, is derived from the microbial metabolism of dietary choline, L-carnitine, and betaine. Increased levels of TMA in breath studies have been connected to kidney disease and colorectal cancer (30,31). Another volatile metabolite in the Atlas is 4-ethylphenol (4EP), a precursor to 4-ethylphenylsulfate (4EPS), a gut-microbially produced compounds which has been mechanistically linked to altered cognition through its disruption of the maturation of oligodendrocytes (the myelinating cells of the central nervous system) and myelination patterns in the brain (32).

The VOC Atlas provides precise information about the baseline levels of volatile metabolites and offers a scientific context for an expanding array of these molecules. Our team is continuously engaged in characterizing volatile metabolites within the Atlas, delving into the biochemical pathways responsible for their production and their interplay with metabolic processes in the body. A deeper understanding of the metabolic processes and disease relevance of these volatile compounds can facilitate the confident and targeted selection of candidate volatile metabolites when designing studies aimed at identifying breath biomarkers for specific diseases. As our research knowledge advances, we are committed to incorporating additional validated breath volatile metabolites and establishing their connections to specific organs and pathways, ultimately mapping them to specific diseases within the VOC Atlas.

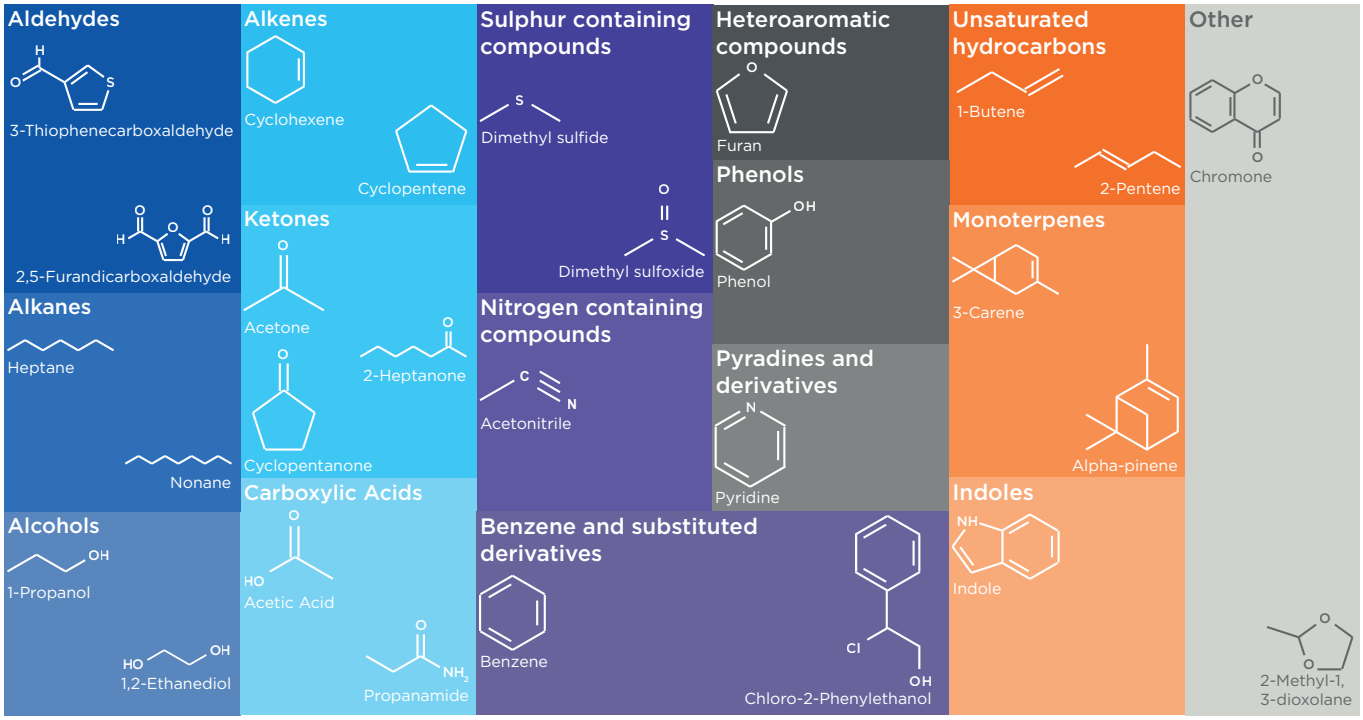

Figure 11: Examples of the volatile compounds that are contained within the VOC Atlas database.

Conclusion

There is a critical need for clinically informative breath biomarkers to support improved drug development, early detection of disease, better clinical monitoring, and precision medicine across a wide range of medical specialisms. Breath is a rich source of potential biomarkers as it contains volatile compounds produced all over the body from many different health and disease-associated pathways. Breath as a sampling medium offers additional benefits in terms of ease of use, patient comfort and potential for testing in home or primary care settings. Previously, the development of breath tests has been limited by a lack of robust and standardized methods and result reporting. This has resulted in many proposed breath biomarkers but few that have successfully advanced through validation to translation into the clinic. Due to the need for better tools, analytical platforms, and interpretation in breath research, a range of Breath Biopsy products and services have been especially developed and optimized in consultation with world leading researchers (Figure 12). The roster of Breath Biopsy products and services available encompass a complete end-to-end solution that can incorporate volatile metabolite analysis into any research study.

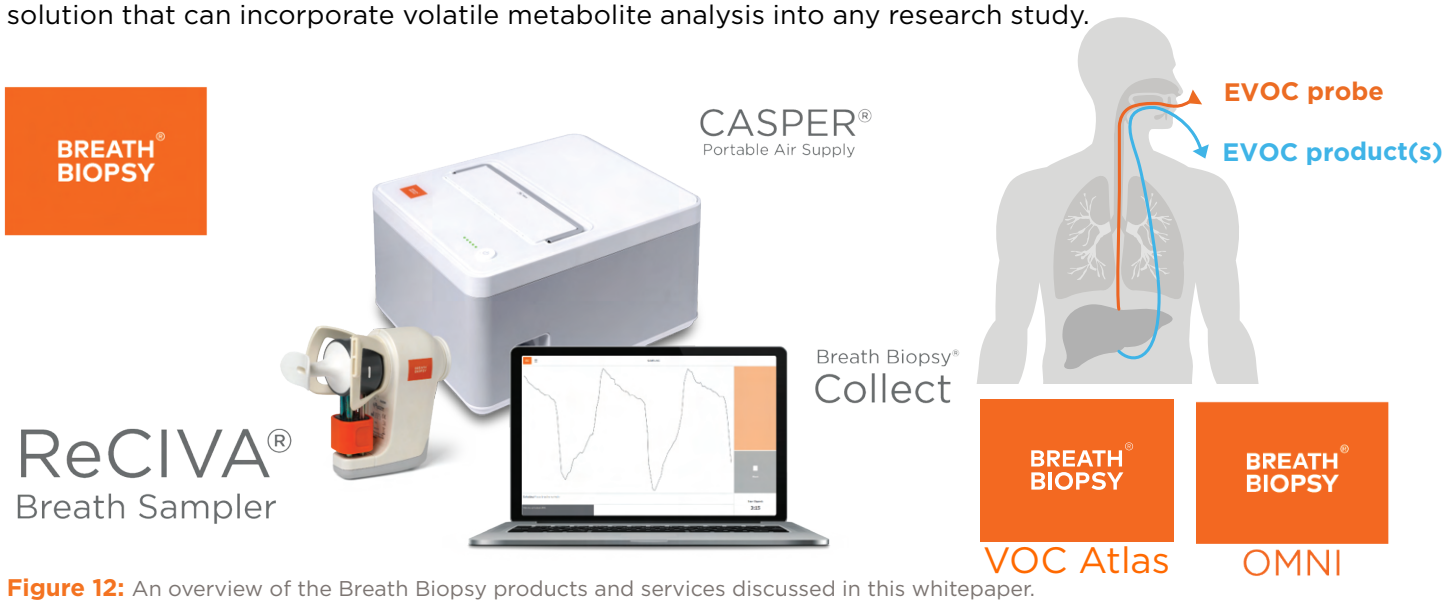

Figure 12: An overview of the Breath Biopsy products and services discussed in this whitepaper.

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