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The utility of breath volatile organic compound (VOC) sampling as a biomarker of sub-optimal nutritional status: a UK pilot study

S. Wilson-Barnes[1], L. Pongcharoenyong[1], L. Gymopoulos[2], K. Dimitropoulos[2], M. Jaksic[3], A. Mihajlović[3], B. Brkic[3], S. Lanham-New[1] and K. Hart[1]

[1]*Faculty of Health and Medical Sciences, University of Surrey, Guildford, UK,*
[2]*Centre for Research & Technology Hellas, Thessaloniki, Greece and*
[3]*BioSense Institute, University of Novi Sad, Novi Sad, Serbia.*

Poor-quality diets (PQD) are associated with non-communicable diseases(1). Furthermore, limited dietary variety may contribute to poor gut microbial diversity, which is associated with cancer risk(2). Technological advances provide opportunities to explore alternative health biomarkers. Specifically, volatile organic compounds (VOC) in exhaled breath may offer a non-invasive biomarker for nutritional status. This pilot study aimed, as part of a wider pan-European collaboration, to characterise the VOC profiles of adults with poor- quality diets to assess their utility as a biomarker of nutritional status.

Fifty participants were recruited in two sub-groups: i) adults with PQD (<3 portions of fruit and vegetables per day, *n* 33) and ii) adults with iron deficiency anaemia (ID; *n* 17). Participants attended the University after a 12 hour fast, with no smoking or chewing gum prior to sampling. Baseline anthropometric measures were taken, and a fasted breath sample was collected according to standard procedures into a Tedlar bag (Zefon Int'l, USA). Participants consumed a standard breakfast and completed online questionnaires evaluating general health and lifestyle. A final breath sample was collected 2 hours post-breakfast. Samples were shipped to researchers at the Biosense Institute (BIOS, Serbia), for extraction and analysis within 3 days of collection, using membrane-inlet mass spectrometry. Reagents used were methanol, ethanol, acetone, isoprene and n- pentane (Sigma-Aldrich, US) all in liquid phase. A favourable ethical opinion was received from the University ethics committee. Parametric analysis was conducted using SPSS (IBM), significance was set at $p < 0.05$, data are presented as mean ± SD.

Mean BMI was 32.7 ± 13.3 kg/m^2 for the whole sample, with no significant (NS) difference between the two groups; 30% were male. Fasted acetone levels were observed to be higher in the PQD than in the ID group (NS, 227.0 ± 246.5 vs 191.2 ± 206.4 ppb, $p = 0.298$). The PQD group presented with lower levels of fasted pentane (NS) (19.8 ± 19.6 vs 26.6 ± 30.5 ppb; $p = 0.498$). Furthermore, a self-reported urban (as opposed to rural) living environment (82%), was associated with higher fasted acetone (NS, 224.2 ± 246.2 vs 176.1 ± 164.4 ppb; $p = 0.290$) and ethanol (NS, 386.2 ± 316.9 vs 175.7 ± 104.4 ppb; $p = 0.091$) concentrations. There were no significant differences observed between the VOC profiles of plant-based (*n* 8) and omnivorous (*n* 37) participants.

In conclusion, this pilot study presents novel data on VOC profiles for two population groups at nutritional risk. Despite a lack of significant between-group differences the group as a whole presented with higher fasted pentane than recommended for healthy adults (0-10 ppb;(3)). Living environments appeared to impact breath profiles and warrant further investigation in various population groups to progress our understanding of the use of VOC profiles as nutritional biomarkers.

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