Breath Ethanol and Acetone as Indicators of Serum Glucose Levels: An Initial Report

Many volatile organic compounds are present in exhaled breath and may represent byproducts of endogenous biological processes. Ethanol is produced via alcoholic fermentation of glucose by gut bacteria and yeast, while acetone comes from oxidation of free fatty acids, influenced by glucose metabolism. We hypothesized that the integrated analysis of breath ethanol and acetone would allow an estimation of the blood glucose profile during a glucose load. We simultaneously collected exhaled breath gas, ambient air and serum glucose and insulin samples from 10 healthy volunteers at baseline and during an oral glucose tolerance test (OGTT).

The diagnosis and follow-up care of diabetes is based upon blood glucose measurements. Glucose loading is also used for the diagnosis of gestational diabetes. Millions of these measurements are performed daily, with significant discomfort or pain for the patient, and at considerable expense. In our research we hoped to prove that the integrated analysis of exhaled gases may serve as an alternative, non-invasive tool for glucose levels measurement.

Student Researcher: Winston Hu
Mentor: Pietro Galassetti, MD, PhD

Introduction

The human breath contains numerous volatile organic compounds (VOCs); many of them are present at concentration in the part per trillion (ppt) range.[1–4] While the origin and patho-physiological importance of most VOCs is poorly understood, elevation of breath levels of some of these gases is beginning to be associated with a variety of metabolic and pathological conditions.

Methods

We tested five males and five females who were non-smoking, were not on medication, and had no chronic diseases. Subjects were admitted to the UCI General Clinical Research Center (GCRC) in the morning following an overnight fast. An intravenous catheter was inserted in the basilic vein of the right forearm. Two baseline blood samples were drawn, at 10 minute intervals, to assess basal fasting glucose levels. As
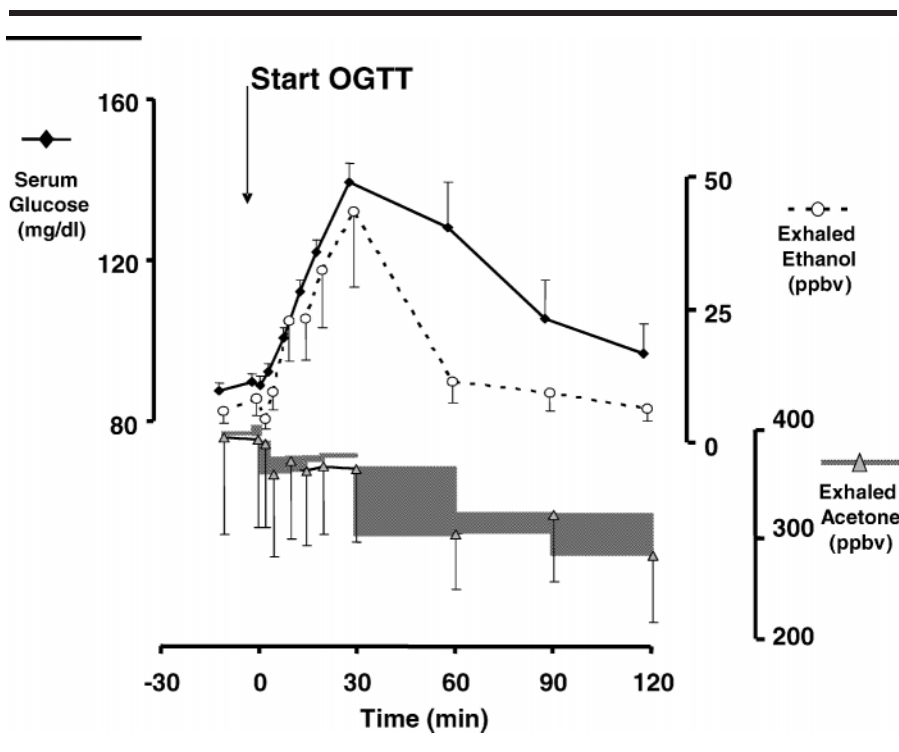

Fig 1. Average serum glucose and exhaled ethanol and acetone profiles in ten healthy volunteers during a standard oral glucose tolerance test after an overnight fast. Data are group means ± SE

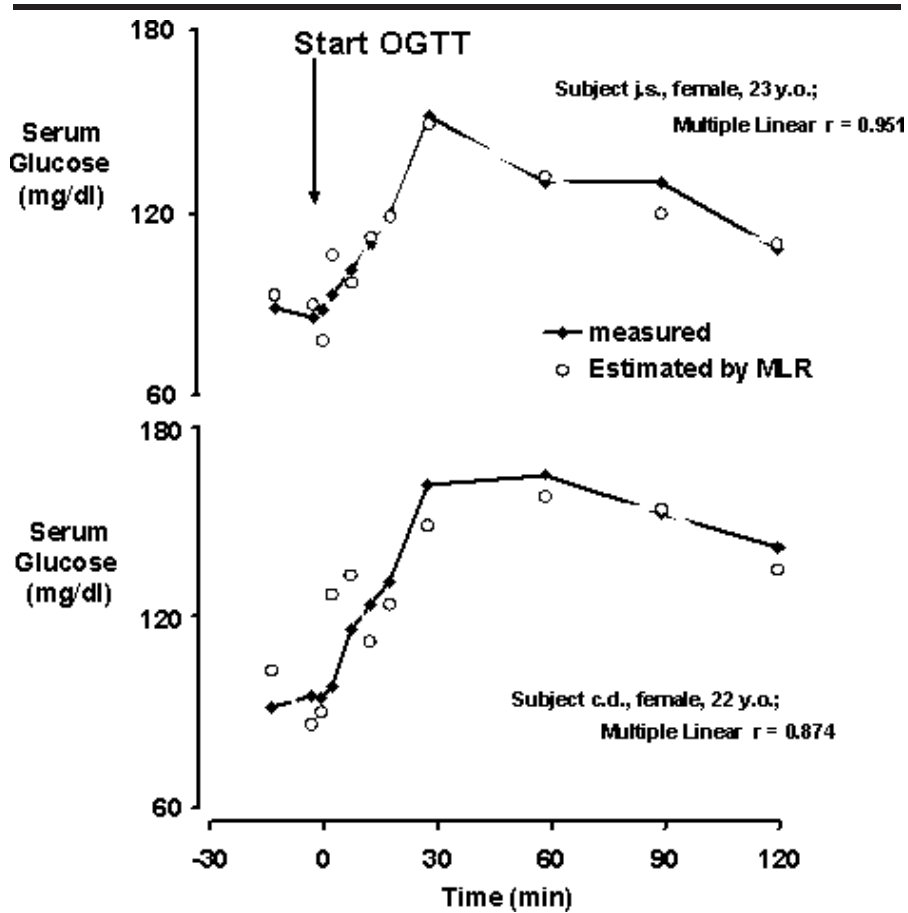From the University of California, Irvine Medical Center; Irvine, California.

Fig 2. Measured serum glucose profiles and glucose profiles estimated with multiple linear regression (ethanol; acetone; glucose) during a standard oral glucose tolerance test (OGTT) after an overnight fast in one subject with normal OGTT (top) and one subject with high 120-min glucose levels, indicating glucose intolerance

each blood sample was drawn, VOCs were collected via electro-polished 1.9-liter stainless steel canisters. A standard oral glucose tolerance test (OGTT) was then performed. Subjects ingested 75 g of glucose diluted in 296 ml of orange-flavored drink solution (Trutol 75, NERL Diagnostics, East Providence, RI). Following the ingestion of glucose, blood and VOC samples were taken at 2.5, 5, 10, 15, 20, 30, 45, 60, 90, and 120 minutes.

RESULTS

Serum Glucose, Insulin, OH-butyrate and Ethanol Levels

The mean baseline fasting serum glucose was 88 ± 2 mg/dL; peak serum glucose concentrations were recorded at 30 min (139 ± 5 mg/dL). This meant that by current diagnostic standards (normal OGTT results of 120 min blood glucose < 140 mg/dL), 9 out of 10 subjects displayed normal results. In one subject, a non-obese Hispanic female with family history of type 2 diabetes, a reading of 142 mg/dL at the 120 min time point resulted in a diagnosis of glucose intolerance, and the subject was referred for appropriate clinical follow-up evaluation.

Serum OH-butyrate was 38 ± 12 at baseline; levels gradually decreased through the study, with values of 27 ± 6 mM by 30 min, 19 ± 7 mM at 60 min, 9 ± 1 at 90 min and 6 ± 1 mM at 120 min (30, 60, 90 and 120 min values

$P < 0.05$–0.01 vs basal values). All serum ethanol measurements throughout the study were, as expected, below the lower detection point of the assay.

Breath Ethanol and Acetone

Baseline levels of ethanol in exhaled breath were 9.6 ± 3.1 parts per billion in volume (ppbv). Exhaled ethanol levels increased during the first part the OGTT peaking by the 30 min time point at 45.0 ± 14.2 ppbv. Exhaled ethanol levels rapidly decreased by 60 min back to near-baseline levels, and by 120 min were 7.9 ± 2.5 ppbv. Baseline levels of exhaled acetone were 392 ± 85 ppbv. After the start of the OGTT, acetone levels displayed a continuous decreasing trend, with values of 364 ± 71 ppbv at 30 min, 300 ± 64 ppbv at 60 min and 280 ± 64 ppbv at 120 min ($P < .05$ vs baseline).

Correlation Between Serum Glucose and Exhaled Gases

When a simple linear regression analysis was performed between either ethanol or acetone and glucose in each subject, both gases did correlate with glucose (ethanol vs glucose, average individual $r = 0.55$, acetone vs glucose, average individual $r = 0.40$). With either gas, however, in a few subjects the correlation was very low (lowest in ethanol-glucose 0.09, lowest in acetone glucose 0.06). After data analysis with multiple linear regression including glucose and both gases, however, an estimated glucose profile could be derived, that correlated with measured glucose values with an average individual r of 0.70, while no subject displayed an $r < 0.41$ and 6/10 subjects displayed an $r > 0.74$. Data from one representative subject are shown in Figure 2.

CONCLUSION

In this study, we made novel observations regarding the ability of breath ethanol and acetone analysis to estimate

serum glucose levels in human subjects. We had hypothesized that using a highly sensitive VOC breath analysis system would enable us to identify patterns of exhaled gases that correspond with serum glucose level. Indeed, we found that the integrated analysis of breath ethanol and acetone, both associated with glucose metabolism, correlated with changes in serum glucose levels.

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