

IDENTIFICATION OF BACTERIA IN HUMAN URINE THROUGH THE ANALYSIS OF EMITTED VOLATILE ORGANIC COMPOUNDS

Hypothesis / aims of study

Analysis of volatile compounds from body fluid can be used to diagnose disease. Volatile compounds emerge into the gaseous phase after the body has expelled the fluid and accumulate in the air above ('the headspace'). The profile of these volatiles is characteristic and reflects the activity of enzymes and their effects on substances in the fluid.

Urinary infection represent a significant health burden. Symptomatic patients with a positive urine dipstick are often prescribed antibiotics on an empirical basis as microbiology culture and sensitivity results often take 24-48 hours to become available. Around 15% of individuals are prescribed inappropriate antibiotics leading to prolonged symptoms and a greater chance of developing antibiotic resistant bacteria.

The aim of this study was analyse the volatile compounds produced in infected human urine using a gas chromatography system.

Study design, materials and methods

A batch of stock sterile urine was produced by filtering the urine of 8 healthy adult volunteers. 50ml samples of the stock urine were inoculated with uropathogenic bacteria and placed in an incubator at 37 degrees. Uninfected stock urine was used as a control. Samples were taken at hourly intervals for 6 hours. A bacterial count and optical density was taken at each time point. A 3ml sample of urine was placed in a gas headspace vial and heated in a water bath for 10minutes. Samples were analysed using a metal oxide gas sensor in combination with a gas chromatograph column. Sensitivity of the sensor was assessed using a 2cm³ sample of 50ppm ethanol/methanol mix on each day experiments were conducted. 2mls of headspace gas was injected into the GC column at each sampling point.

The study was repeated using 12 nitrite positive urine specimens obtained from the urology outpatient department. A sample of the infected urine was sent to the hospitals microbiology laboratory for analysis as per usual hospital protocol. 12 dipstick negative samples were used as controls.

The percentage response (height of chromatogram peaks in relation to baseline) and retention times (timing of peak from injection point) were analysed for each of the samples.

Results

A chromatogram was generated for each sample mapping the sensor's response to the presence of volatiles over a 42 minute time period.

Visual assessment of the chromatograms revealed characteristic differences in the volatile trace patterns for each bacteria. The percentage response and retention times for each bacterium were mapped and compared to controls. Percentage responses were greater with increasing bacterial concentrations however retention times at differing concentrations remained largely constant. Each bacterium produced unique retention time patterns. Most significant peaks were given off in the first ten minutes of sampling.

Interpretation of results

In both seeded, filtered human urine and infected patient samples, characteristic chromatographs were produced for different urinary tract infection causing organisms.

Concluding message

This small study suggests analysis of volatile compounds in the gas head space of urine may enable rapid identification of bacteria causing urinary tract infections in symptomatic individuals. This may allow more appropriate antibiotic choice, enabling effective treatment with reduced risk of developing antibiotic resistance.

Disclosures

Funding: North Bristol Trust small grant scheme **Clinical Trial:** No **Subjects:** HUMAN **Ethics Committee:** UK South West Central National Research Ethics Service **Helsinki:** Yes **Informed Consent:** Yes