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## Source, impact and removal of malodour from soiled clothing

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### ABSTRACT

Static headspace – multi-capillary column – gas chromatography – ion mobility spectrometry (SHS-MCC-GC-IMS) has been applied to the analysis of malodour compounds from soiled clothing (socks and T-shirts), pre- and post washing, at low temperature (20 °C). Six volatile compounds (VCs) (i.e. butyric acid, dimethyl disulfide, dimethyl trisulfide, 2-heptanone, 2-nonanone and 2-octanone) were identified. After sensory evaluation of soiled garments they were subjected to laundering with non-perfumed washing powder. The efficiency of the laundering process was evaluated by determining the reduction of each detected volatile compound (VC) post-wash (damp) for socks and T-shirts; VC concentration reductions of between 16 and 100% were noted, irrespective of sample type. Additionally the T-shirt study considered the change in VC concentration post-wash (dry) i.e. after the drying process at ambient temperature. Overall VC concentration reductions of between 25 and 98% were noted for T-shirt samples pre-wash to post-wash (dry). Finally, a potential biochemical metabolic pathway for the formation of malodour compounds associated with bacteria in axillary sweat is proposed.

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### 1. Introduction

Laundry has been an important domestic activity in the whole of human life. It is not known when and where human beings first started to wash their clothes. In the past laundry was commonly carried out in rivers and streams by hand and scrubbing with soap; this process is still used in developing countries today. Laundry washing is a means of removing dust and dirt from clothing and also of getting rid of malodour i.e. the presence of unpleasant smell on clothes. Nowadays advanced washing machines with several washing programmes (wash cycles) and effective detergents make washing easier and more efficient.

Personal malodour arises from the production of VCs generated by the action of microorganisms in breaking down the components of sweat, human skin cells and secretion substances from the glands. Characteristic malodour VCs that can arise from these processes include ammonia and hydrogen sulfide as well as short chain fatty acids; its occurrence can be a cause of personal embarrassment. As well as VCs generated from the human body, they can also arise from external sources, such as, the washing machine, as well as the drying environment. However, the characteristic malodour generated from laundries, which can be detected just after washing, can be due to poor hygiene in laundering resulting from

microbial fabric damage and biofilm build up inside the washing machine [1]. Traditionally in Europe, laundry has been washed at a high temperature (>60 °C). However, for environmental reasons, lower temperatures are now encouraged. At low temperature the generation of malodour is more common [1,2]. Detergent manufacturers have been designing detergents for low temperature washing (i.e. between 15–20 °C) and these are now available on the European market. The addition of perfumes in laundry detergents and fabric conditioners are used to neutralize unpleasant odours on clothing, which are not reactively removed in the washing process. It is apparent, by the range of products on supermarket shelves and their associated marketing that household consumers would be less satisfied with their garments if they were laundered with non-perfumed detergents. Modern detergents have been formulated as complex mixtures with various types of surfactants (ionic and non-ionic), bleach, enzymes as well as essential oils. Generally, surfactants in detergents are designed to remove dirt, in to the aqueous phase, while perfumes tend to remain on the garment.

Numerous factors have been identified for the formation of malodour in laundries, such as, the drying environment [3,4], bacterial colonisation (biofilms) [4,5] and metabolism of micro-organisms [6] as well as human odour [7]. However, limited studies have been carried out to investigate the bacterial colonisation (biofilms) in washing machines [4,5]. In household washing machines, microbial survival followed by biofilm formation occurs due to soiled garments and poor water treatments [5]. A number of microbe species have been isolated and identified from the biofilm within

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washing machines [1,4,5]. Among the garment and water carrying parts of washing machines “hot spot” biofilm formation have been discovered (i.e. product drawer, sump and rubber seal) [4]. Moreover a recent study [4] showed a possible link that contamination of fabrics occurs from the washing machines itself, based on the identification of the same bacteria in the washing machine rubber seal and on fabrics. According to this study potential VC markers for both high levels of bacteria and malodour in washing machines are dimethyl disulfide and dimethyl trisulfide.

In this paper, characteristic malodour marker VCs were determined using static headspace – multi-capillary column – gas chromatography – ion mobility spectrometry (SHS-MCC-GC-IMS). This analytical technique was selected over gas chromatography mass spectrometry due to its enhanced detection of sulfur-based compounds, speed of analysis and multi-dimensional detection capabilities [8]. The influence of the washing process on the reduction of VCs has been evaluated from pre-wash to post-wash on pre-selected soiled clothing items at low temperature. The washing process was evaluated using a developed scaled-down laundry approach using non-perfumed washing powder. The clothing items, namely socks and T-shirts, were worn by volunteers during either normal or extended physical activity, respectively. The study was extended to take in to account the drying process in the extended physical activity study.

## 2. Experimental

### 2.1. Chemicals/reagents

Acetone (CAS 67-64-1,  $\geq 99.9\%$ ), ammonia (CAS 1336-21-6, 28% NH<sub>3</sub> in H<sub>2</sub>O,  $\geq 99.99\%$ ), butyric acid (CAS 107-92-6,  $\geq 99\%$ ), dimethyl disulfide (CAS 624-92-0,  $\geq 98\%$ ), dimethyl trisulfide (CAS 3658-80-8,  $\geq 98\%$ ), 2-heptanone (CAS 110-43-0,  $\geq 98\%$ ), 2-nonanone (CAS 821-55-6,  $\geq 98\%$ ), and 2-octanone (CAS 111-13-7,  $\geq 98\%$ ) were all purchased from Sigma-Aldrich (Dorset, UK). Stock solutions (10,000 ppm) were prepared using acetone. Milli-Q water of conductivity 18.2 M $\Omega$ -cm was produced by a direct QTM Millipore system 165 (Molsheim, France) and was used in all dilution steps.

Sock samples (74% cotton, 19% polyester, 5% nylon and 2% lycra) and 100% polyester white sport T-shirts (Nike) were obtained from a local retail outlet (Newcastle, UK). Headspace (20 mL) crimp-cap vials and magnetic caps were purchased from Sage Analytical Ltd. (Lancashire, UK). Nylon Fire Bags (250 mm  $\times$  375 mm) were obtained from Crime Scene Investigation (Woburn Sands, UK) and were used for collection and storage of the soiled fabric samples. A leading granular laundry detergent (non-perfumed and perfumed) were obtained from the Procter & Gamble Technical Centre, Newcastle upon Tyne.

### 2.2. Instrumentation

A static headspace – multi-capillary column – gas chromatography – ion mobility spectrometer (SHS-MCC-GC-IMS) manufactured by G.A.S.-Gesellschaft für Analytische Sensorsysteme mbH (Dortmund, Germany), was used [8]. The SHS-MCC-GC-IMS was fitted with an automatic sampler unit (CTC-PAL; CTC Analytics AG, Zwingen, Switzerland) and a heated gas-tight syringe. A multi-capillary column (MCC) (Multichrom, Novosibirsk, Russia) was used for the chromatographic separation. The MCC comprised a stainless steel tube, 20 cm  $\times$  3 mm ID, containing approximately 1000 parallel capillary tubes, 40  $\mu$ m ID, coated with 0.2  $\mu$ m film thickness of stationary phase i.e. OV-5. Atmospheric pressure ionisation is generated by a Tritium (<sup>3</sup>H) solid state bonded source ( $\beta$ -radiation, 100–300 MBq with a half-life of 12.5 years). The IMS has a drift tube length of 50 mm. Separation in the IMS drift tube is achieved by

applying an electric field of 2 kV to the ionized volatiles in a pulsed mode using an electronic shutter opening time of 100  $\mu$ s. The drift gas was N<sub>2</sub> (99.998%) with a drift pressure of 101 kPa (ambient pressure). Samples were run under the following operating conditions: incubation conditions (time, 5 min; and, temperature, 95 °C); MCC-IMS conditions (syringe temperature, 85 °C; injection temperature, 80 °C; injection volume, 1.5 mL; column temperature, 35 °C; and, carrier gas flow rate, 10 mL/min); and, IMS conditions (temperature, 60 °C; and, drift gas flow rate, 500 mL/min) for butyric acid, dimethyl disulfide and dimethyl trisulfide whereas for 2-heptanone, 2-nonanone and 2-octanone the incubation conditions were as follows: (time, 5 min; and, temperature, 95 °C); MCC-IMS conditions (syringe temperature, 85 °C; injection temperature, 80 °C; injection volume, 1.5 mL; column temperature, 35 °C; and, carrier gas flow rate, 150 mL/min); and, IMS conditions (i.e. temperature, 45 °C; and, drift gas flow rate, 500 mL/min). All data was acquired in the positive ion mode and each spectrum is formed with the average of 42 scans. All data are processed using the LAV software (version 2.0.0, G.A.S). The software package enables both two- and three- dimensional data visualisation plots.

A tergotometer (Copley Scientific, Nottingham, UK) was used to simulate the washing machine. The tergotometer contained eight stainless steel vessels, each with a capacity of 1000 mL. The temperature within the stainless steel vessels of the tergotometer was controlled by a water circulatory heating system. The temperature was adjustable in the range ambient to 70 °C. Each stainless steel vessel was capable of being stirred within the range 50–200 rpm. Sub-samples of socks and T-shirt were added to the tergotometer in the approximate ratio of 2.5 g: 300 mL (fabric: water).

### 2.3. Procedure for sock analysis

Eight healthy volunteers (6 males and 2 females) were selected to participate in this study. Each participant received a pair of new socks and a wear protocol; each sock was enclosed in a uniquely coded sample bag i.e. right sock and left sock. Participants were not allowed to apply odorous products during the study i.e. deodorant or moisturiser. The wear protocol was as follows: each foot was rinsed thoroughly with tap water and then dried. Participants then wore the socks over a minimum period of 10 h during one day in a specified type of footwear i.e. shoes. After use the participants transferred each sock into the uniquely coded bag, which was sealed and stored overnight in a dark place. The sample containing bags were then returned to the investigator the following day. Each sample was olfactory graded according to a numerical scale ranging from 0 (no malodour) to 10 (malodorous).

The socks were then sub-sampled in three distinct areas i.e. toe, ball and heel by taking approximately 2.5 g of fabric. The sub-samples were then placed into 20 mL headspace vials and closed with a magnetic cap prior to SHS-MCC-GC-IMS analysis. Unworn socks were prepared as blank samples and analysed using the same methods; this allowed extraneous VCs to be identified and eliminated from future data treatment. Samples were washed according to the developed Tergotometer washing protocol using non-perfumed washing powder. After washing, the sock sub-samples were manually wrung out using fingers and each placed in a 20 mL vial for analysis as post-wash (damp) samples using SHS-MCC-GC-IMS.

### 2.4. Procedure for T-shirt analysis

Prior to wearing the T-shirts they were washed in a domestic washing machine (Servis, 1200 rpm) that itself was pre-cleaned. Pre-cleaning of the washing machine was done using Dr Beckmann Service-it Deep Clean washing machine cleaner by following the supplier instructions and washing at 60 °C for approximately 1 h.

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