Identification of a possible biomarker for colophony exposure

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Colophony is known to cause occupational asthma and dermatitis. Biological monitoring may be useful in assessing exposure. This paper describes a method for the analysis of dehydroabietic acid in urine and its potential use as a marker of colophony exposure. The method involves hydrolysis, solvent extraction, derivatization and analysis by gas chromatography–mass spectrometry. Twenty-eight workers from a soldering factory in South Africa were monitored. Results showed that levels of dehydroabietic acid in urine may be correlated with a subjective assessment of exposure.

Key words: Biological monitoring; colophony; occupational exposure; rosin; urine.

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Introduction

Colophony (rosin) is a natural product derived from pine resin with many industrial applications, including commercial fluxes and ‘rosin core’ solder in electronics soldering. The chemical constituents in colophony vary, but rosin is reported to consist of ~90% resin acids and 10% neutral matter. Isomeric forms of abietic acid make up ~90% of the resin fraction, the remainder being mostly dihydroabietic acid and dehydroabietic acid. The Methods for Determining Hazardous Substances (MDHS) document for colophony analysis in air [1] quotes abietic acid as the major constituent, with dehydroabietic acid as the secondary component.

Colophony is known to cause asthma and contact dermatitis, and control of exposure is important. Biological monitoring may be of help in assessing the adequacy of exposure control. There have been no previously published methods for biological monitoring of colophony, although biological effect markers have been determined [2] and the mechanism of sensitization has been investigated [3]. This paper presents a method of analysis for determining dehydroabietic acid in urine, and its use as a marker of colophony exposure, among a group of workers assembling electronic devices in a South African factory.

The factory manufactures electricity meters. An imported pre-printed circuit board is populated (components inserted) by hand. Solder resist is applied to keep the solder off certain places and the board is then wave soldered. The wave soldering machine is small and has a dedicated extraction system, but some fumes are evident at the entry and exit points. The boards are then examined, ‘touched up’ by hand soldering where necessary, and any excess solder is removed. The solder wire used is Multicore 60/40 (UK), which incorporates rosin flux, and Multicore X33 flux (resin free) was used in wave soldering.

The boards are coated with Multicore Xersin 2050 (which contains 2–3% modified rosins) in an enclosed machine. The boards (about A5 size) are hung on cross-members and rotated through the coating fluid, then taken out. The machine has an extraction system, but when the machine is opened at 1 or 2 min intervals, there is a noticeable increase in the ketone odour associated with the methylethylketone that makes up ~25–30% of the fluid.

The next step is to add other components, such as wiring and circuit breakers to connect the board, to the finished product. This requires hand soldering. Once complete, the assembled circuit boards are installed into the plastic outer casing, tested and packaged for despatch.
Methods

Analytical method

A stock solution of 23 mM dehydroabietic acid (>99% pure; Cansyn Chemical Corp., Toronto, Canada) was prepared in methanol:acetonitrile (50:50). From this, a working solution of 116 \( \mu \)M dehydroabietic acid was prepared in methanol. Standards were prepared by ‘spiking’ urine from unexposed persons with this working solution. An internal standard of heptadecanoic acid was used. Aliquots (2 ml) of urine were hydrolysed at \(-90^\circ\text{C}\) for 1 h with 250 ml of conc. HCl. Samples were then extracted into 8 ml of diethyl ether. The organic layer was removed and evaporated. The residue was derivatized using 50 \( \mu \)l of dimethylformamide dimethylacetal (Sigma-Aldrich, Gillingham, UK) at 75°C for 30 min. All reagents were of high-pressure liquid chromatography or derivatization grade.

The samples were next analysed by gas chromatography–mass spectrometry (EI+) using a BP-5 (or equivalent) column (30 m x 0.32 mm internal diameter, 1 \( \mu \)m film). Splitless (30 s) injections (1 \( \mu \)l) were made into the injector, which was held at 250°C. The oven was maintained at 140°C initially for 1 min, then heated at 40°C/min to 200°C, at which it was kept for 5 min. A further increase (5°C/min) took the final temperature to 280°C for 2 min. The transfer line was held at 280°C. Selected ion monitoring was used: \( \text{m/z} \) 314 for dehydroabietic acid; \( \text{m/z} \) 284 for heptadecanoic acid.

Field survey

Twenty-eight workers, from a South African factory that assembled and soldered electric meters, provided urine samples after a work shift. The shifts were 10 h long, with two half-hour breaks. The samples were frozen and packed in ice packs before being shipped to the UK, where they arrived still frozen. On receipt, samples were stored at \(-20^\circ\text{C}\) (under these conditions, samples are considered to be stable for at least 2 months).

The levels of airborne exposure to colophony were not known, as no environmental sampling had been undertaken. No local exhaust system for the removal of solder fume was installed. The general ventilation of the factory appeared inefficient; by mid-afternoon, carbon dioxide levels were about twice as high inside the factory as outside. Exposure levels were subjectively ranked 1–4 by the research team; the basis of the ranking system is illustrated in Table 1.

Results

Analytical method

The response was linear (least squares regression coeffi-
in the urine of exposed workers and is linear, with an intra-assay coefficient of variation of 8%.

The increasing mean for dehydroabietic acid levels with increasing exposure ranking and the borderline statistical significance ($P = 0.08$) of the correlation between exposure ranking and urinary dehydroabietic acid levels suggest that dehydroabietic acid in urine may be a useful marker of colophony exposure. Further studies are required that involve a full occupational hygiene survey encompassing both environmental and biological monitoring.

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References