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LETTER TO THE EDITOR

An Improved Method for Preparing Dimethyl Disulfide Adducts for GC/MS Analysis

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Abstract Dimethyl disulfide (DMDS) adducts are prepared from fatty acid methyl esters to locate the double-bond position in monoenoic fatty acids by gas chromatography/mass spectrometry. We improved our original procedure for preparing DMDS adducts with a simple device that can be made in any laboratory using a glass pipette and silica based packing material, Extelut NT.

Keywords Dimethyl disulfide adduct preparation · Double-bond position · Monoenoic fatty acid

Sir

In 1981, Francis [1] reported the usefulness of dimethyl disulfide (DMDS) adducts for locating the double-bond position in an authentic monoenoic fatty acid methyl ester by gas chromatography/mass spectrometry (GC/MS); electron impact ionization of DMDS adducts gives an abundant molecular ion, and the cleavage between the methylthio-substituted carbons produces a set of fragment ions that serve to locate the original double-bond position in a monoenoic fatty acid ester. In 1985, Leonhardt and DeVilbiss [2], Dunkelblum et al. [3] and Shibahara et al. [4] separately reported that DMDS adducts could be prepared from mixtures of double-bond positional isomers of monoenoic acid esters and from mixtures of saturated and unsaturated esters from natural sources (bacterial lipids [2], pheromone gland lipids [3] and plant lipids [4]). Each research group, however, used different reaction conditions for preparing the DMDS adducts (e.g., reaction times and temperatures: 24 h at 40 °C [2]; overnight at 40 °C [3]; and 30 min at 35 °C [4]).

Procedures to form DMDS adducts are widely used. The Lipid Library [5] lists more than 70 publications regarding DMDS adducts. Our research group has been preparing DMDS adducts for ~2 decades [4, 6–17]. We recently developed an improved procedure that reduces the hazard to analysts from the DMDS fumes and substantially reduces the intensity of the unpleasant odor. In addition, it shortens the preparation time. This procedure is especially beneficial when many samples of DMDS adducts must be prepared (e.g., to trace monoenoic acid bioformation [9, 10]).

The following is the new procedure: Use commercially available glass pipettes (ca. 15 cm × 5 mm id). As shown in Fig. 1, place ca. 10 mg of cotton (defatted with *n*-hexane before use) into the pipette to lightly plug the tip. Then, add ca. 0.6 g of the silica-based packing material, Extrelut NT (E. Merck, no.15093) (heated at 500 °C for 1 h before use), and tap the pipette to firmly pack the Extrelut NT. Next, pour 0.5 mL of a saturated $Na_2S_2O_3$ aqueous solution onto the Extrelut NT and let the pipette stand for ca. 5 min. The device is now complete and will be referred to as mini-column A. Analysts should only prepare the mini-column A immediately prior to use. One mini-column A can be used per sample.

While the mini-column A is being prepared, incubate a mixture of fatty acid methyl esters (1–1,000 μg as methyl monoenoates) with 0.1 mL of DMDS containing 1.3 mg of I_2 for 30 min at 35 °C in a reaction vial (e.g., the vials used with the auto injector of a GC instrument).

Then, pour the reaction mixture (containing DMDS, I_2 and the resultant DMDS adducts) along with 0.9 mL of

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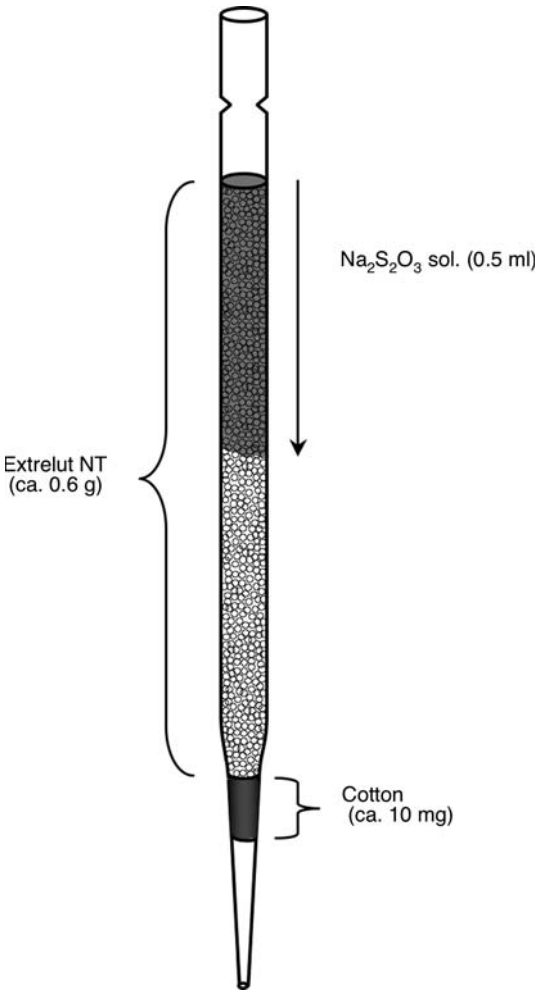

Fig. 1 Illustration of mini-column A, used to prepare dimethyl disulfide adducts. Instructions for making mini-column A are described in the text

diethyl ether/*n*-hexane (10:90, v/v) into the mini-column A. Rinse the reaction vial with 1 mL of diethyl ether/*n*-hexane (10:90, v/v) five times, and add this to the mini-column A. Collect the entire eluent (ca. 5 mL, allowing ca. 5 min to drain), and evaporate it to dryness in a stream of nitrogen gas. Dissolve the remaining DMDS adducts in *n*-hexane (an approximate volume), and then introduce them into a GC/MS system.

We compared the GC/MS data of DMDS adducts prepared using our previous procedure to the GC/MS data of DMDS adducts prepared with our improved procedure. There were no significant differences. We hope the modified procedure will prove useful.

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