Waters
Lab Highlights
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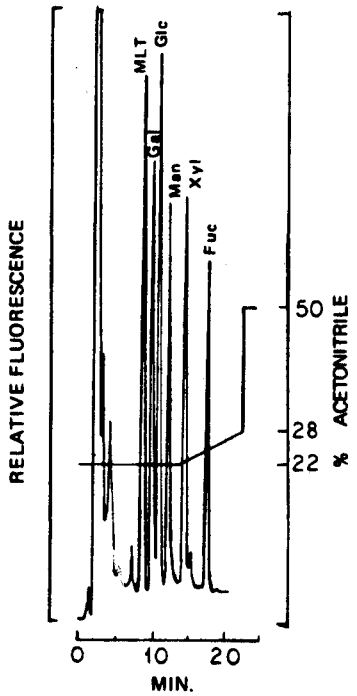
NEW, SENSITIVE METHOD FOR CARBOHYDRATES IN BIOLOGICAL SAMPLES

A procedure for the analysis of neutral sugars in biological specimens is described. The method entails acid hydrolysis of the sample to liberate monosaccharides, which are subsequently derivatized with dansyl hydrazine. The sugar-dansyl hydrazones are separated and quantitated by HPLC on a 5μ Radial-PAK™ C_{18} column with a gradient using acetonitrile and 10mM ammonium sulfate at pH 7 (see Figure 1). Fluorescent detection of the derivatized sugars permits 100-fold increased sensitivity compared to previously published time-consuming GLC methods.

The Redken research group showed that: This procedure is the "method of choice" for neutral sugar analysis of biological materials. They were also highlighted in a cosmetic trade magazine as reporting "a new method of determining the quantity and type of carbohydrates found in hair."

A glycoprotein of known composition (thyroglobulin) and hard keratin fibers served as models to critically evaluate the method on a highly resistant biological matrix containing low concentrations of neutral sugars. A chromatogram is shown below.

FIGURE 1



Chromatographic separation of standard sugar-dansyl hydrazones. Conditions for separation as described in text. Detector was Schoeffel FS970 Spetrofluoro Monitor (excitation wavelength, 240 nm; emission longpass filter, 550 nm; range, 0.2 AFS; sensitivity, 42%; time constant, 2.5 sec). Sample was 1 nmole each standard: mlt, maltose; gal, galactose; glc, glucose; man, mannose; xyl, xylose; fuc, fucose.

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Reference:
(1) W. F. Alpenfels, R. H. Mathews, D. E. Madden, A. E. Newsom, J. Liquid Chromatogr.