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Phospholipids Analysis by ^{31}P NMR

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PHOSPHOLIPIDS ANALYSIS BY ^{31}P NMR

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Cell membrane phospholipids can be identified and quantitated using ^{31}P NMR spectroscopy in conjunction with an analytical reagent composed of chloroform-benzene(d_6)/methanol-CsEDTA 2:1 ml/ml. 3 ml of this reagent dissolves between 0.01-100 mg crude tissue lipids obtained by the Folch procedure. When the source phospholipids are strongly contaminated with cations, it is necessary to modify the extraction method, backwashing with K-EDTA, 0.6 M, pH 6, instead of KCl. Also if source tissues must be stored for long periods of time, acetone desiccation is recommended. Using a 500 MHz ^{31}P NMR spectrophotometer (magnetic field=11.75 T), the extracted phospholipids yield narrow Lorentzian signals (1.8-3.2 Hz at half-height), with these widths at half-height corresponding to their $1/\pi T_2$ values. Chemical shifts (δ) at 24 °C, following the IUPAC shift convention and relative to 85% phosphoric acid, were determined as follows: CAEP, 21.09; LPG, 1.09; LPA, 0.83; LPE plas, 0.53; PG, 0.50; LPE, 0.43; PA, 0.25; CL, 0.18; LPI, 0.10; PE plas, 0.07; PE, 0.03; PS, -0.05; SPH, -0.09; DiMePE, -0.18; LPC plas, -0.20; LPC, -0.28; PI, -0.37; PAF, -0.70; PC plas, -0.77; PC, -0.84. This reagent permits assays of high precision and accuracy that use little spectrometer time and that are suitable for automated procedures.

HUMAN SYNOVIAL PHOSPHOLIPIDS

