

Simultaneous Gas Chromatographic Analysis of Amino Acids and Organic Acids

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N(O,S)-ethoxycarbonylation combined with tert.-butyldimethyl- silylation was optimized and validated for the simultaneous gas chromatographic (GC) analysis of amino acids and organic acids. Ethoxycarbonylation of amino, phenolic and sulfhydry groups with ethyl chlorofomate in aqueous solution was followed by tert.-butyldimethylsilylation of carboxyl and remaining polar groups for the direct GC analysis after solvent extraction. The present method was found to be potentially useful for the biochemical diagnosis of inherited metabolic disorders.

[PD4-20] [10/19/2001 (Fri) 09:00 - 12:00 / Hall D]

Simultaneous Quantitative Analysis of Sphingoid Base 1-Phosphates in Biological Samples by o-Phthalaldehyde Precolumn Derivatization after Dephosphorylation with Alkaline Phosphatase

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This paper describes a simultaneous analytical method for the measurement of sphingoid base 1-phosphates and sphingoid bases from a variety of biological samples. This method consists of two steps of sample pretreatment: the enzymatic dephosphorylation of sphingoid base 1-phosphates by alkaline phosphatase and the subsequent analysis of OPA derivatives of the liberated sphingoid bases by HPLC. By introducing C₁₇-sphingosine 1-phosphate and C₁₇-sphingosine as internal standards, not only phytosphingosine 1-phosphate, sphingosine 1-phosphate, and sphinganine 1-phosphate but also phytosphingosine, sphingosine, and sphinganine present in a sample could be quantified in 12 min on a C₁₈ reversed-phase column with a simple mobile phase of acetonitrile : water (90 : 10, v/v). With this HPLC method, we could reproducibly analyze the levels of sphingoid base 1-phosphates over a broad range of concentrations from 0.5 to 100.0 pmol from various biological samples including serum, cultured cells and rat tissue homogenates. The conversion of sphingoid base 1-phosphates into sphingoid bases increased the stability of the OPA adducts. Thus, this indirect measurement of sphingoid base 1-phosphates increased the sensitivity and reproducibility of the method. This HPLC method was also used to measure the changes in the levels of sphingoid base 1-phosphates in cultured cells after treatment with 1,25-(OH)₂D₃, a sphingosine kinase activator, or with fumonisin B₁, a sphinganine N-acyltransferase inhibitor.

[PD4-21] [10/19/2001 (Fri) 09:00 - 12:00 / Hall D]

Diagnostic Patterns for Capillary Electrophoretic Urinary Nucleoside Profiles from Patients with Liver Diseases

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An efficient capillary electrophoretic profiling method in micellar electrokinetic capillary chromatography (MEKC) mode was combined with simple pattern recognition methods for the correlation between urinary