

Amino Acid Analysis via the Hitachi Sensivate Elite Post-Column Reactor and LaChrom Elite® HPLC

Kendra Cox, Ph.D., Hitachi High Technologies America, Inc.

Amino acid analysis is an important technique involved in a wide range of bioanalytical applications including protein and peptide analysis, quality assessment of foodstuffs and clinical diagnosis of metabolic disorders. Historically, this technique has been performed on highly specialized chromatographic systems dedicated for comprehensive (up to 50-plus components) and quantitative amino acid analysis. Here we describe an alternative methodology for the analysis of amino acids in protein hydrolysates and foodstuffs, including γ -amino-n-butyric acid (GABA) and ornithine, using the highly flexible Hitachi LaChrom Elite® HPLC system with the Sensivate Elite Post-Column Reactor. Amino acids are analyzed via ion exchange separation, derivatized with ninhydrin, and detected by UV-Vis. The data here specifically describes the analysis of soy sauce¹.

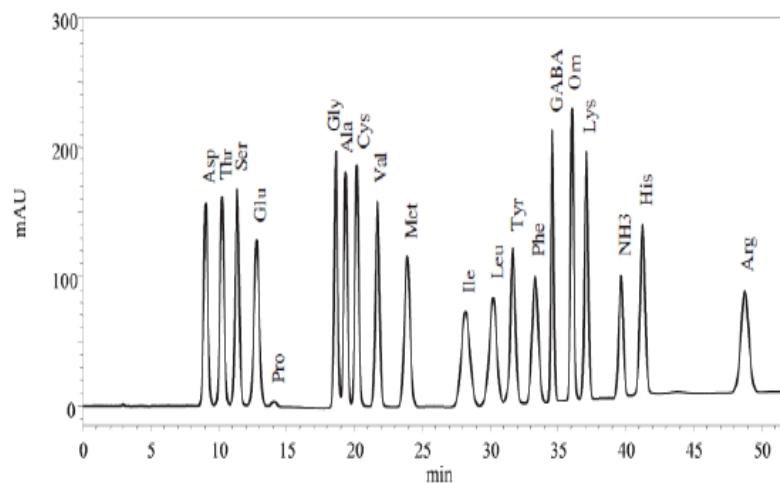
Experimental Conditions

Module	Conditions
Pump (L-2130)	Flow Rate: 0.4 mL/min
Eluent	Hitachi L-8900 buffer kit: PH-1, PH-3, PH-4 & PH-RG
Analytical Column	Hitachi ion exchange column for amino acid analysis – 4.0 mm ID x 150 mm (#2619PH)
Autosampler (L-2200)	Injection Volume: 10 μ L
Oven (L-2300)	Temperature: 57°C
Post-Column Reactor	Sensivate Elite, 0.5mL reactor block, 130°C, Hitachi L-8900 ninhydrin kit, 0.3 mL/min
Detector (L-2420)	UV-Vis, 570 nm
Standard	20-component amino acid mixture

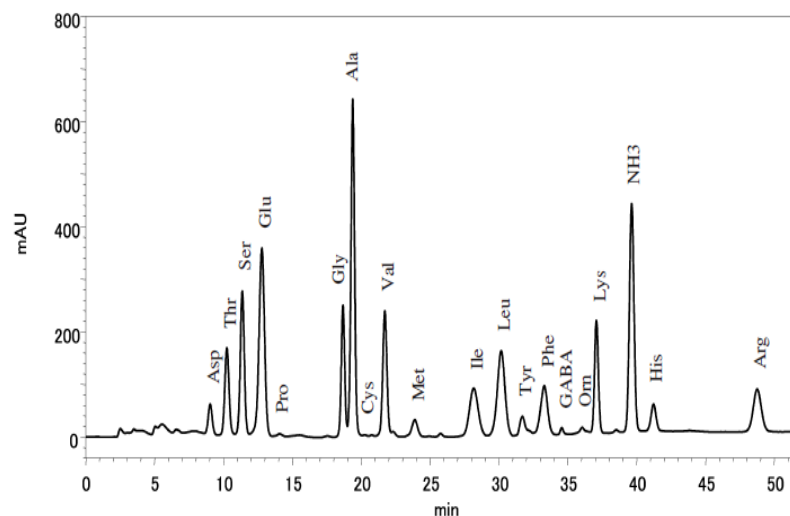
Results – Performance

Peak Area Reproducibility	RSD < 1.7% for each amino acid at 2 nmol
Retention Time Reproducibility	RSD < 0.1% for each amino acid at 2 nmol

Results – Chromatogram of 20-Component Mixture



Results – Chromatogram of Soy Sauce



Discussion

Hitachi's LaChrom Elite® liquid chromatography system with Sensivate Elite Post-Column Reactor is extremely effective at analysis of amino acids, including separation of GABA and ornithine, in a variety of matrices. The system exhibits excellent reproducibility and has the flexibility to be used for a variety of post-column reaction and conventional HPLC applications.

Reference:

1 – Technical Data LC96, Hitachi High Technologies Corporation.

Hitachi High Technologies America, Inc.

Life Sciences Division
 5100 Franklin Drive
 Pleasanton, CA 94588
 Toll Free: (800) 548-9001