CAPSULES
preliminary notes and applications from Bioanalytical Systems, Inc.

Thyromimetic Iodoamino Acids

Purpose
Separation and electrochemical detection of thyromimetic iodoamino acids.

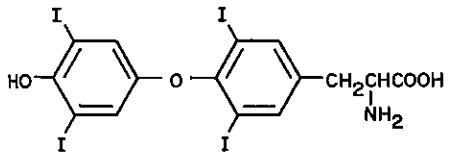

Figure 1. Thyroxine (T4)

The thyroid hormones T3 and T4 (F1) are widely studied for their key role in the biological processes involved in growth and differentiation. They influence metabolism and have an effect on many enzymes and organ systems. Fast, accurate and sensitive assays are needed for their study.

Existing Methods
Wet analysis, radioimmunoassay, derivatization-GC with electron capture, thin-layer and paper chromatography, electrophoresis, GC/MS. All suffer from either inadequate sensitivity or lengthy sample preparation. HPLC with UV or fluorescent detection is better but UV (220 nm) has poor detection limits, and fluorescence requires derivatization.

Reference
Rapid Separation and Determination of Thyromimetic Iodoamino Acids by Gradient Elution Reverse Phase Liquid Chromatography with Electrochemical Detection, M.R. Hadj-Mohammadi, J.L. Ward and J.G. Dorsey, J. Liq. Chromatogr., 7(1983) 511-526.

Conditions
Detector: BAS LC-4

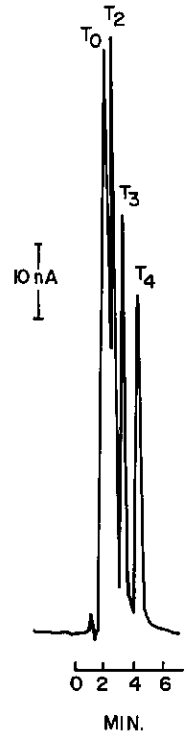

Figure 2. Results of isocratic elution of 4 thyromimetic amino acids.

Electrode: BAS TL-5 (Glassy Carbon)
Potential: +1.2 V vs Ag/AgCl
Column: 5 µm C 8 (250 x 4.6 mm) for isocratic separation; 5 µm C 18 (150 x 4.6 mm) for gradient elution
Mobile Phase: Isocratic: CH_3OH:H_2O:H_3PO_4 (70:30:0.2). Flow rate was 2 mL/min (F2). Gradient: A: 0.15% H_3PO_4 in H_2O; B: 0.20% H_3PO_4 in CH_3OH. Gradient was 0-90% B in 3 ramps. Flow rate was 2 mL/min (F3).
Detection Limits: 650 pg (T4) and 350 pg (T2, T3)
Linear Range: 0 - 1 µg (T4) and 0 - 500 ng (T2, T3)

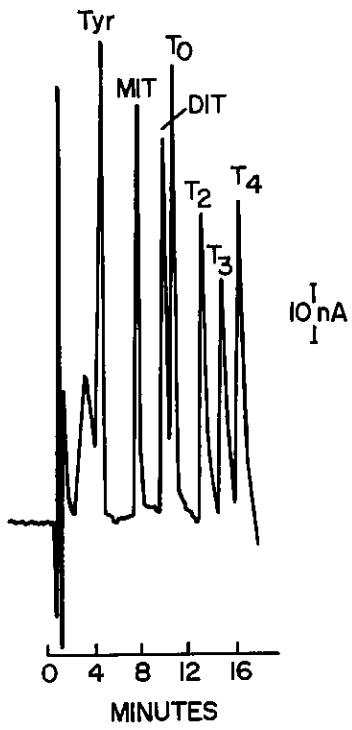

Figure 3. Results of gradient elution of 7 thyromimetic amino acids.

Sample Preparation

Samples were prepared from standards or pharmaceutical preparations. Injection volumes were 10 µL (gradient) and 5 µL(isocratic).

Notes

Gradient elution in LCEC is rare, and the authors provide a good discussion of their technique.

The isocratic elution of thyromimetic iodoamino acids presented above can be duplicated on the BAS 400 Liquid Chromatograph or the BAS 200 Problem Solver. The gradient elution procedure can be duplicated with the BAS 200.

The information in this publication is supplied as a service to our customers. Performance of the methodology has not necessarily been verified by BAS technical staff.