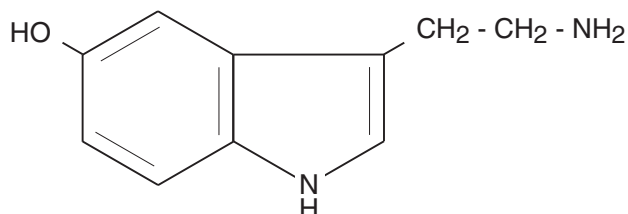


Determination of Serotonin in Rat Brain Microdialysate Using a UniJet SepStik Microbore Column and Radial-Flow Cell

1009

Purpose

Determination of serotonin (5-hydroxytryptamine, 5-HT, F1) in rat brain microdialysates. These low concentration samples were separated and detected utilizing a UniJet SepStik microbore column and a radial-flow Electrochemical cell.



Existing Methods

Thin-layer chromatography, gas chromatography, radioimmunoassay, and liquid chromatography with fluorescence or electrochemical detection.

Conditions

Systems: Liquid Chromatography equipped for microbore column chromatography.

Detector: BASi Electrochemical Detector.

Electrochemical Detector Electrode: 3 mm glassy carbon electrode (PN MF-2061)

Potential: + 650 mV vs. Ag/AgCl

Column: UniJet SepStik 3 μ m, C18, 100 x 1 mm (PN MF-8949)

Flow rate: 100 μ L/min

Mobile Phase 1: 1L (25 mM NaH₂PO₄, 50 mM sodium citrate, 27 μ M disodium-EDTA, 10 mM diethylamine HCl, 10 mM NaCl, 2 mM decane-sulfonic acid (sodium salt), pH to 3.2 with H₃PO₄); 175 mL acetonitrile.

Mobile Phase 2: 1L (0.1 M Monochloroacetic acid, 0.5 mM EDTA, 10 mM NaCl, 0.65 mM sodium octylsulfonic acid, pH to 3.1 with H₃PO₄); 100 mL acetonitrile.

Detection Limit: 0.25 pg 5-HT on column.

Notes

Typical chromatograms of 5-HT standard, microdialysate sample from rat brain and spiked microdialysate using mobile phase 1 are presented in F2. F3 shows the chromatograms using both mobile phase 1 and mobile phase 2 for determination of the same standard and microdialysate samples. A calibration curve of 5-HT is presented in F4. The concentration-time curves of 5-HT in microdialysate from rat brain are presented in F5.

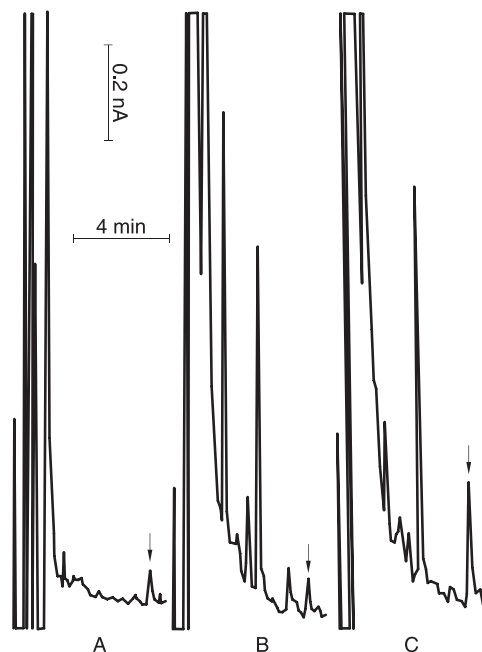


Figure 2. Chromatograms of 1 pg serotonin standard (A), microdialysate from rat striatum (B) and a mixture of serotonin standard and microdialysate from rat striatum (C) using mobile phase 1. 5 μ L were injected.

References

- Huang, T., Shoup, R.E. and Kissinger, P.T., *Current Separations*, 9 (1990) 139.
- Bohs, C.E., Linhares, M.C. and Kissinger, P.T., *Current Separations*, 12 (1994) 181.

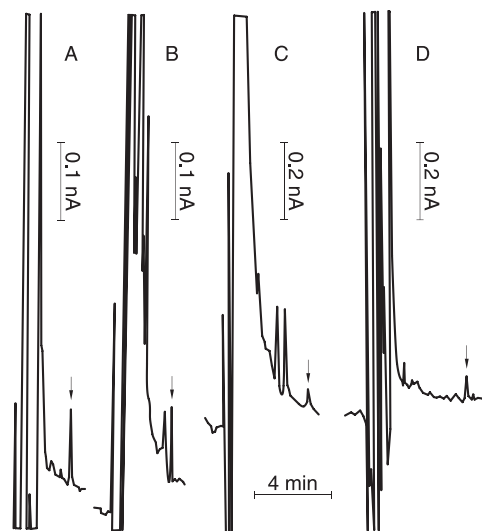


Figure 3. Chromatograms of 1 pg serotonin standards (A,D) and microdialysate samples from rat striatum (B, C) using mobile phase 1 (A and B) and mobile phase 2 (C and D). Microdialysis conditions same as in F1.

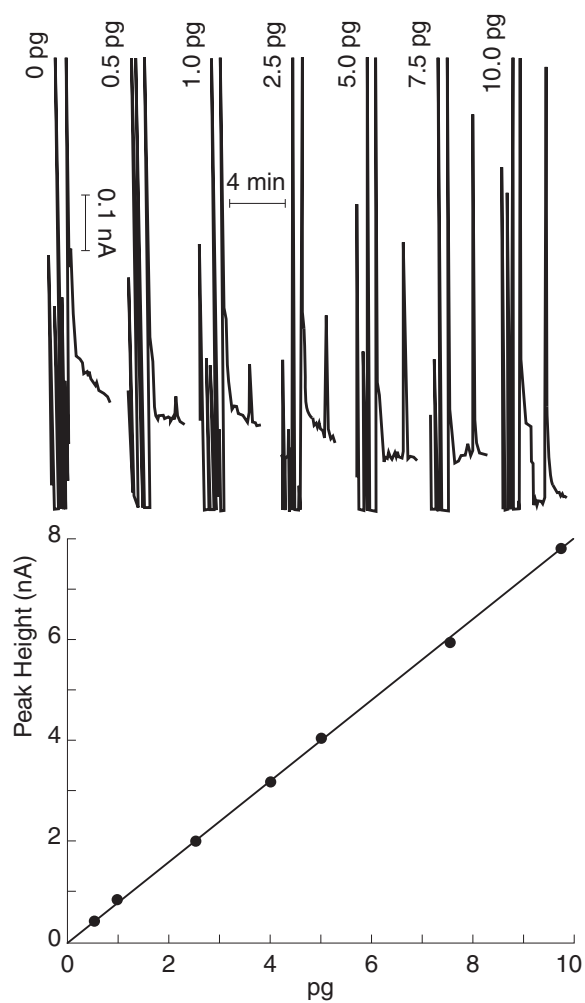


Figure 4. Calibration curve for serotonin using mobile phase 1.

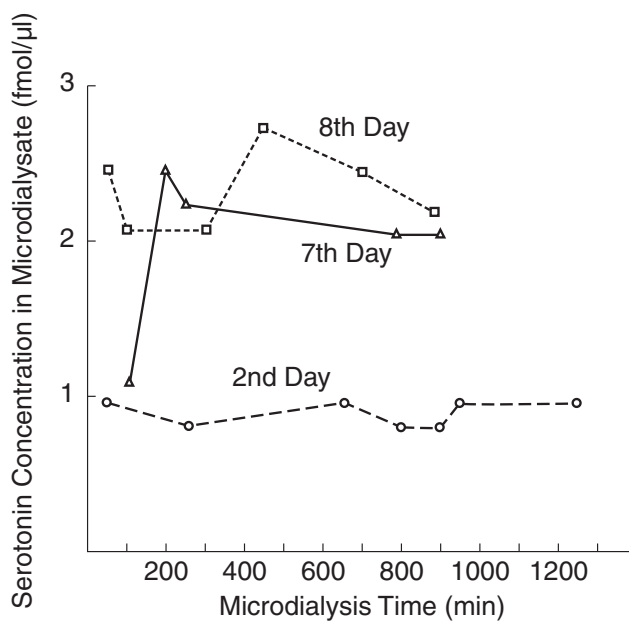


Figure 5. Concentration-time curve of serotonin in microdialysates from rat brain striatum. LCEC conditions same as in F1.