CAPSULES
notes and applications from Bioanalytical Systems, Inc.

# Determination of Purines in Microdialysates Using SepStik Columns

**Purpose:**
Determination of adenosine, guanosine and adenine (F1) in microdialysates.

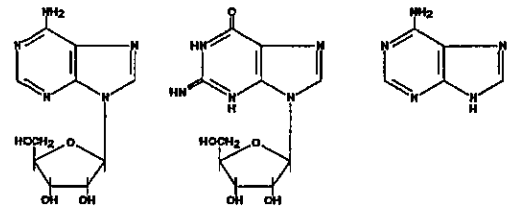


***Figure 1.*** Structures of adenosine (A), guanosine (B) and adenine (C).

In order to separate and sensitively detect purines in microdialysates, a SepStik microbore column was used. The 1 mm internal diameter increases the concentrations of the eluting purines up to 21-fold compared to standard LC columns.

**Existing Methods**
LCEC and LCUV with conventional columns.

**Conditions**
System: BAS 481 Liquid Chromatograph
Column: SepStik kit (BAS P/N MF-8949).
The packing was ODS 3 µM silica in a 100 x 1.0 mm bed.
Mobile Phase: 7 mM $NaH_2PO_4$ containing 3.5% $CH_3OH$. Adjust pH to 3.04 after adding $CH_3OH$.
Flow Rate: 80 µL/min actual measured flow rate. For conventional pumps, use BAS P/N MF-8947 Flow Splitter Accessory kit, and maintain about 2 mL/min flow rate at the pump. Adjust splitter ratio to give 75-85 µL/min through the microbore column by adjusting restrictor and/or pump flow rate.
Detector: BAS UV-108, with microcell.
Wavelength: 260 nm
Range: 0.002 AUFS
Rise Time: 1.0 sec.
Detection Limit: 50 pg injected yielded a S/N of 3. The injection volume was 5 µL.

**Sample preparation:**
Dialysate was directly injected onto the system.

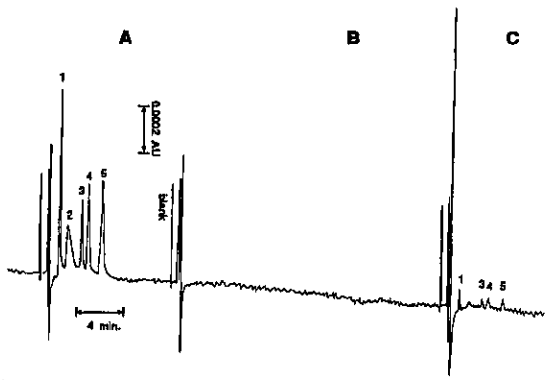


***Figure 2.*** Detection limit test
A: 500pg of each purine standard
B: Blank
C: 50pg of each purine standard

Peak Identification:
1. hypoxanthine
2. adenine
3. inosine
4. guanosine
5. adenosine

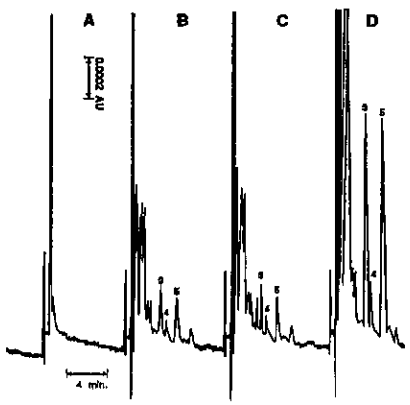


***Figure 3.*** Purines in various rat striatum dialysates.
A: Blank (Ringer's solution)
B, C & D: Various dialysate samples

Peak Identification as in F2.

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