Waters
Lab Highlights
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AUTOMATED SAMPLE CLEANUP AND ASSAY FOR PROCAINAMIDE FROM SERUM

The M590 Programmable Solvent Delivery System provides the capability to conveniently automate sample cleanup and chromatographic analysis from biological fluids (serum, plasma, urine, etc.). A complex procedure has been described by Roth et al. (1). The Model 590 pump equipped with a solvent select valve and high pressure switching valve along with a WISP™ and a M440 detector at 280 nm simplifies the Roth method.

The pump program, initiated by the WISP™ at ingestion, is listed in Table 1.

TABLE 1

M590 Program for Procainamide Method

Segment	Time (min)	Solvent	Flow (ml/min)	Valve Position
1	Initial	A	0.5	1
2	1.0	A	4.0	1
3	3.0	B	4.0	1
4	3.9	B	4.0	2
5	12.0	A	4.0	1

The plumbing connections for the high pressure valve are shown in Figures 1 and 2.

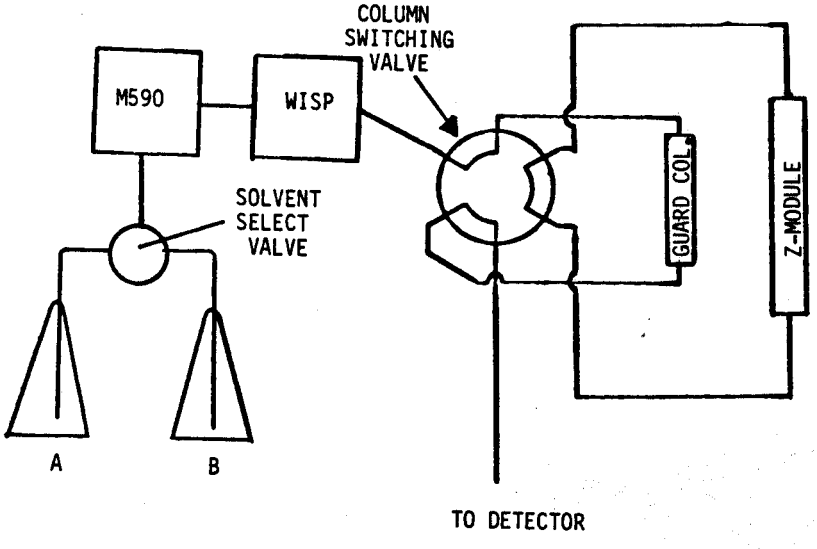

FIGURE 1: FLOW PATH FOR SAMPLE CLEANUP; SOLVENT A.

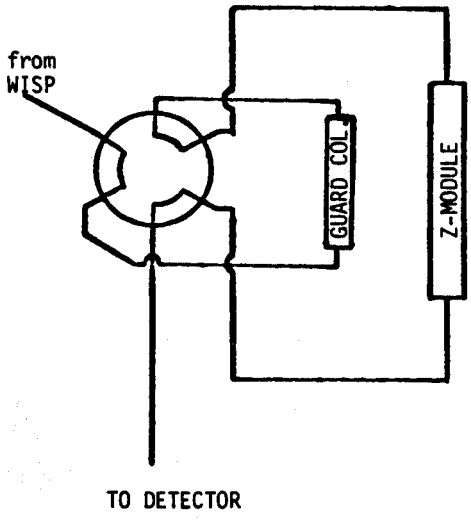

FIGURE 2: FLOW PATH FOR BACKFLUSH AND ANALYTICAL SEPARATION: SOLVENT B.

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At initial conditions, 100 µl of serum sample is injected into a mobile phase of 100% water (Solvent A). After 3.0 minutes, the analytical mobile phase (Solvent B) is selected, and before it elutes through the guard column, the high-pressure valve is switched to backflush the sample onto the analytical column and complete the assay. The valve then returns to the initial position, and Solvent A equilibrates with the guard column so the procedure may be repeated.

Chromatographic conditions are shown below.

Figure 3
Standards

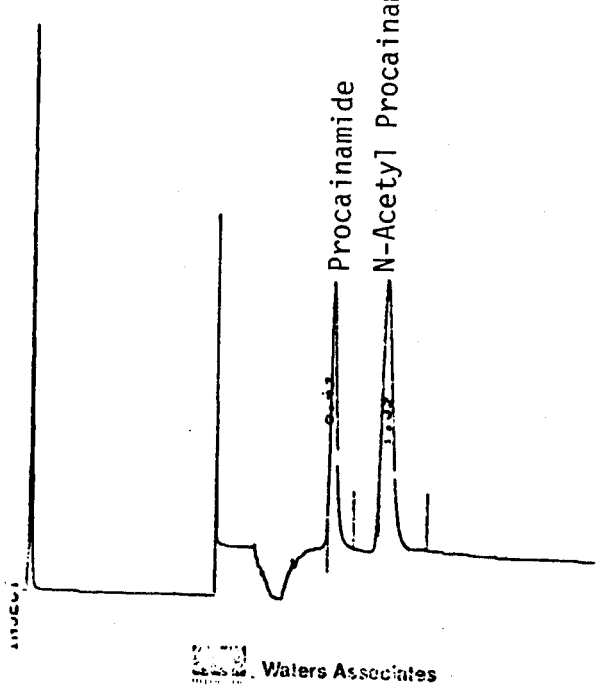
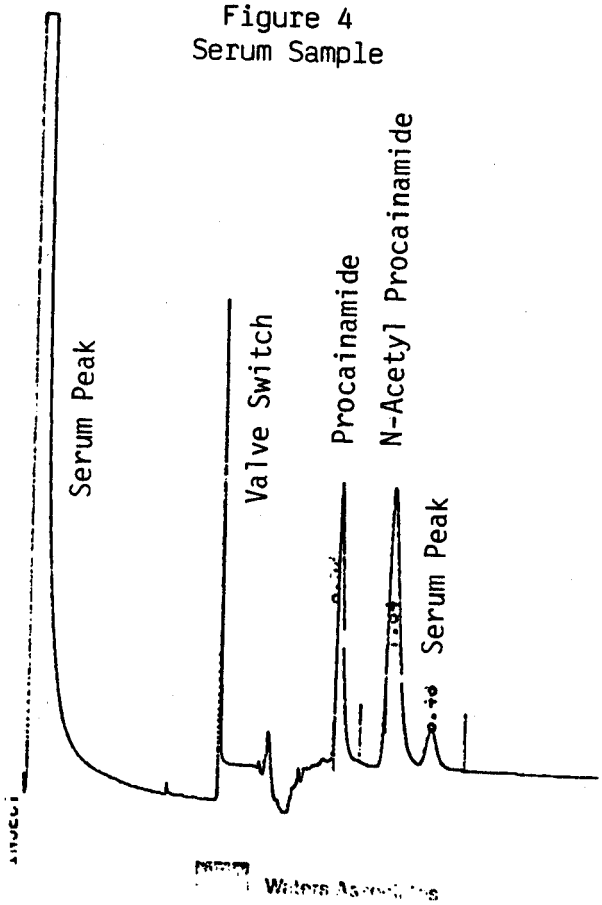


Figure 4
Serum Sample



Injection Volume:	100 µl Serum
Column:	Radial-PAK™ µBONDAPAK™ C_{18} in Z-Module™
Solvent A:	H_2O
Solvent B:	69% H_2O, 30% Methanol, 1% Acetic Acid, pH = 5.5
Detector:	M440, 280 nm, 0.1 AUFS

1. W. Roth, K. Beschke, R. Janch, A. Zimmer, F. W. Koss, J. Chromatog. 222, (1981) 13-22.