

SPESolid Phase
Extraction

Application Note

World Leaders in Sample Preparation

SPE145**ISOLATION OF AMINOGLYOSIDES FROM SERUM**

Spe-ed™ Cartridge	Cat. No. 2311- COOH Carboxylic Acid, 100mg/1mL. Process with <i>Spe-ed Mate</i> .
Cartridge Conditioning	0.5mL of 1 N hydrochloric acid. 2mL of distilled water.
Sample Addition	1mL of serum containing 10ug of internal standards (paromomycin).
Cartridge Wash	2mL of distilled water.
Analyte Elution	1mL of 0.2 M perchloric acid. Filter eluant through 0.2um nylon 66 syringe filter prior to analysis.
HPLC Conditions	C18, 15cm x 4. 6mm (5um)
Column	0.01 M pentane sulfonate / 5.6mM sulfate/ 7
Mobile phase	mM acetic acid in water : methanol (80:20)
Flow Rate	1.75mL/minute
Temperature	40°C
Detection	Fluorescence (excitation 365nm, emission 418nm, high voltage 600v)
Injection	20ul
Post Column	0.4 M H ₃ BO ₃ / 0.38 M KOH (6mL of 40% brij,
Research conditions	4mL of mercaptoethanol, 0.89 of o-phthalaldehyde added / 1 liter)
Reagent Flow Rate	0.4mL/minute.
Mixer	5cm x 4.6mm S\ S column packed with 75um glass beads
Reactor	3m x 0.5mm knitted Teflon capillary tubing

Note: Since sample matrix interferences and concentrations may vary from sample to sample, it may be necessary to adjust the wash and elution solvent/solution strength and/or volume to optimize isolation.

The logo for Applied Separations, featuring the company name in a bold, italicized sans-serif font with horizontal lines above the word 'Applied'.

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