

SPESolid Phase
Extraction

Application Note

World Leaders in Sample Preparation

SPE164**ISOLATION OF AMIKACIN FROM SERUM**

Spe-ed™ Cartridge	Cat. No. 12001-Octadecyl C18/18%, 100mg/1mL. Process with <i>Spe-ed Mate</i> .
Sample Preparation	Pipette 50uL serum, 25uL of 2 mol/L tris buffer and 100uL of working internal standard (kanamycin, 16mg/L into a 1.5mL polypropylene tube. Vortex and centrifuge for 1 minute @ 15,000g. Decant, supernate into polypropylene tube, add 30uL of TNBS solution (2.5gms. of trinitrobenzene sulfonic acid in 10mLs acetonitrile). Cap, vortex and place in a 70°C heating block for 30 minutes.
Cartridge Conditioning	2mL methanol 2mL water
Sample Addition	Add 700uL of 10 mmol/ L potassium hydrogen phosphate (1.82gms. of potassium hydrogen phosphate in 1L distilled water, adjust to pH 8.6 with phosphoric acid) to column vacuum off, then add 250uL of derivatized sample. Aspirate sample completely through the cartridge.
Cartridge Wash	3mL of 10 mmol/ L potassium hydrogen phosphate in water.
Analyte Elution	300uL of acetonitrile.

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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