

**SPE**Solid Phase  
Extraction

# Application Note

World Leaders in Sample Preparation

**SPE236****ISOLATION OF EXOGENOUS COMPOUNDS**

<b>Spe-ed™ Cartridge</b>	Cat. No.2724- Flow ABN, 3mL. Process with <i>Spe-ed Mate</i> .
<b>Sample Preparation</b>	Place 1.0mL whole blood into a 16x 125mm culture tube. Add 5mL of 0.1 M phosphate buffer (pH6). Add 0.1mL internal standards of hexobarbital. (50ug/mL) and benzphetamine (5ug/mL). Vortex 5 seconds.
<b>Cartridge Conditioning</b>	4mL of methanol. 4mL of water. 2mL of 0.1 M phosphate buffer (pH 6.0) <b>DO NOT ALLOW CARTRIDGE TO RUN DRY!</b>
<b>Sample Addition</b>	Apply sample using no more than 5in. mercury vacuum.
<b>Cartridge Wash</b>	2mL of 0.1 M phosphate buffer, pH 6 (< 5in.Hg vacuum). 1mL 1.0 N acetic acid (<5in.Hg vacuum). Centrifuge at 1,000 x g. for 10 minutes.
<b>Analyte Elution (acidic and neutral fractions)</b>	Place collection tubes under <i>Spe-ed</i> Flow cartridges. Elute with 5mL ethyl acetate (elute by gravity). Evaporate to dryness under N <sub>2</sub> at 65°C. Reconstitute with 100uL ethyl acetate. Analyze by GC/MS.
<b>Analyte Elution (basic fractions)</b>	Place collection tubes under <i>Spe-ed</i> Flow cartridges. Elute with 3mL 2% triethylamine in isopropanol. Evaporate to dryness under N <sub>2</sub> at room temperature. Reconstitute with 50uL reagent alcohol. Analyze by GC/MS.

**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".

930 Hamilton Street · Allentown, PA 18101  
610-770-0900 · 610-740-5520 (fax)  
www.appliedseparations.com