



## **BASIC DRUGS FOR HPLC ANALYSIS USING: 200 mg CLEAN SCREEN<sup>®</sup> EXTRACTION COLUMN**

Part #: ZSDAU020 without Tips or ZCDAU020 with CLEAN-THRU<sup>®</sup> Tips

### **1. PREPARE SAMPLE**

To 1 mL of 100 mM phosphate buffer (pH= 6.0) add internal standards\*. Mix/vortex. Add 1-5 mL of urine or 1 mL of blood, plasma/serum or 1g (1: 4) tissue homogenate.

Mix/vortex.

Add 2 mL of 100 mM phosphate buffer (pH 6.0)

Sample pH should be 6.0 ± 0.5.

Adjust pH accordingly with 100 mM monobasic or dibasic sodium phosphate.

Centrifuge as appropriate.

### **2. CONDITION CLEAN SCREEN<sup>®</sup> EXTRACTION COLUMN**

1 x 3 mL CH<sub>3</sub>OH.

1 x 3 mL D.I. H<sub>2</sub>O.

1 x 1 mL 100 mM phosphate buffer (pH= 6.0).

NOTE: Aspirate at < 3 inches Hg to prevent sorbent drying.

### **3. APPLY SAMPLE**

Load at 1 to 2 mL/minute.

### **4. WASH COLUMN**

1 x 3 mL D.I. H<sub>2</sub>O.

1 x 1 mL 100 mM acetic acid.

1 x 3 mL methanol.

Dry column (5 minutes at > 10 inches Hg).

### **5. ELUTE BASES**

1 x 2 mL CH<sub>3</sub>OH/NH<sub>4</sub>OH (98:2).

Collect eluate at 1 to 2 mL/minute.

NOTE: Prepare elution solvent daily.

### **6. EXTRACT**

To eluate add 2.0 mL D.I. H<sub>2</sub>O and 500 µL methylene chloride.

Mix/vortex.

Centrifuge at 2,000 RPM for 10 minutes.

Transfer organic lower layer to a clean test tube.

### **7. EVAPORATE**

Evaporate to dryness at < 40°C.

### **8. QUANTITATE**

Reconstitute in mobile phase and inject onto the HPLC.

**SOURCE:** UCT Internal Publication