VALIDATION SUMMARY	
Procedure Name	Solid Phase Extraction of Alkaline Drugs from Biological Fluids
Validation Summary	Validation experiments were performed using the current TOX203-10 solid phase extraction modified to analyze blood stains on clothing for a single investigation as follows: - Fabric samples (~ 2" x 2") were stained with 1 mL of blood samples spiked with fentanyl & norfentanyl at 5 ng/mL and cocaine, benzoylecgonine, ecgonine methylester, cocaethylene, methamphetamine, amphetamine, oxycodone, & noroxycodone spiked at 25 ng/mL (in duplicate). Each batch also contained fabric samples (1 of each) stained with 1 mL of negative control blood. - Each batch also contained the following 1 mL blood controls in test-tubes: negative blood, low control blood (5 ng/mL & 25 mg/mL as above), and a high control at 20 ng/mL (fentanyl/norfentanyl) and 100 ng/mL (all others). - Dried blood stained fabric samples were placed into test-tubes (16 x 100 mm) and soaked with methanol for ~ 4 hours. The methanol extracts were removed and placed into new test-tubes (16 x 100 mm) and dried to ~ 200 uL at 40°C (note: dried residue on the sides of the test-tubes will not be vortexed during the drying process but left behind). The ~ 200 uL methanol extracts were transferred into new test-tubes (16 x 100mm), reconstituted with 1 mL deionized water and extracted per TOX203-10. - Step I. Reconstitute was modified for all as follows: - To each test-tube: add 50 uL of methanol followed by 100 uL water, vortex, transfer to 0.22 um centrifugal filter tubes, and centrifuge for 5 minutes at 10,000 rpm. Filtrates will be transferred to ALS vials with inserts. TOX203 column, mobile phases and LC/MS parameters per TOX203-10 were used for this validation. Limit of Detection (LOD) experiments were evaluated (three fabrics in duplicate x 3 sets): all of the positive samples contained fentanyl & norfentanyl at 5 ng/mL and cocaine, benzoylecgonine, ecgonine methylester, cocaethylene, methamphetamine, amphetamine, oxycodone, & noroxycodone at 25 ng/mL.

APPROVALS			
Technical Approval	Redacted	Date	3/29/2024
Unit Chief Approval		Date	3/29/2024