K25 Detection of Drugs in Postmortem Blood and Skeletal Tissue (Bone)

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After attending this presentation, attendees will have a better understanding of the utilization and interpretive value of the detection of drugs in postmortem skeletal tissue (bone).

This presentation will impact the forensic community by informing forensic professionals how to utilize alternate matrices such as bone when traditional specimens such as blood, body fluids, and tissues are not available for toxicological analysis.

A recent multiple homicide case where only skeletonized and extremely putrefied fluids were available prompted the laboratory to establish a standardized approach for the analysis of bone in future cases. To do this, bones from non-decomposed specimens were evaluated. Cardiac and Femoral Blood (FB), along with routine postmortem specimens (iliac crest or vertebrae) were collected in 20 cases. Specimens were analyzed using a 13-panel Enzyme-Linked Immunosorbent Assay (ELISA), base and acidic/neutral, Enzyme Multiplied Immunotechnique (EMIT), acetaminophen, and salicylate analyses. Confirmations/quantitations were performed using gas chromatography/mass spectrometry.

Bones were stored at -20°C for three years prior to analysis. Cleaned bones were rinsed with deionized (DI) water, dried for 48 hours, manually crushed, and ground in a commercial grinder. Bone (3g) was added to 10mL methanol, vortexed 1 minute, incubated 48hrs at room temperature, and rotary extracted for 8hrs. Methanol supernatants were recovered by centrifugation, evaporated to near dryness, combined with 100µl of 0.1% methanolic HCl. Phosphate Buffer (PB: 2mL, 100mM, pH 6) and 4mL acetonitrile: methanol (2:1 v/v ratio) were added to evaporated extracts. Vortexed extracts were stored at -20°C for 12 hours, then centrifuged 5 minutes. Supernatants were reduced to 2mL by evaporation, and diluted to 5mL with PB. Bone extracts were analyzed in the same manner as blood. Drug-free bones from postmortem blood screening served as negative controls (n=3).

Opioids were detected in 8/9 cases, which screened positive by ELISA. In two heroin cases, only codeine was detected, 6.6ng/g and 15ng/mL, bone and FB, respectively in one case, no opiates were present in the other. Two cases contained hydrocodone, 36 and 8.3ng/g bone; 76 and 44ng/mL FB. Dihydrocodeine was detected in both FB samples, but not in bone. Oxycodone was detected in three cases at 161, 25, and 83.5ng/g, bone and 520, 65, and 920ng/mL FB. Oxymorphone was present in one bone specimen and in all three FB. Fentanyl, methadone, and propoxyphene were detected in one case each: (4.17ng/g, bone, 8.7ng/mL, FB), (217ng/g, bone, 530ng/mL, FB), (1.35ng/g bone, and 2.9mg/L FB), respectively. EDDP was present in FB, 220ng/mL, but not in bone. Norpropoxyphene was present in both matrices.

Methylenedioxypyrovalerone (MDPV) and N-benzylpiperazine (BZP) were present in individual cases in FB, urine, and bone. Synthetic cannabinoids JWH-18 and JWH-250 were detected in one case, bone, 0.33 and 1.4ng/mL and FB, 0.48, 4.6ng/mL, respectively, with AM-2201 and RCS-4 being found in bone only, synthetic cannabinoid analysis performed by NMS Labs. Cocaine/metabolites were present in four cases: benzoylecgonine, <20, 62, 258, and 512ng/g for bone, and 88, 96, 830, and >2000ng/mL for FB. EME was in two bone samples, anhydroecgonine methylester (AEME) was present in one bone sample and not detected in FB. Cocaine was present in one case at 58.4ng/g, bone, 278ng/ml, FB.

Nordiazepam was detected at 67.8ng/g, bone, qualitatively present in FB. Clonazepam was detected in two FB samples, but not in the corresponding bone. Cyclobenzaprine was present in one specimen, 183ng/g, bone, 14ng/mL, FB. Venlafaxine was present in one case, 768ng/g, bone, 0.69mg/L, blood. Norvenlafaxine and lidocaine were present in both matrices. Dextromethorphan was present in bone, 968ng/g and >2000ng/L for FB, in an overdose case. Diphenhydramine was present in two FB, 1.45 and 0.075mg/L, but was only seen in the first at 300ng/g, bone. Cotinine correlated in 6/7 cases with FB and urine. Nicotine and caffeine were not found in bone, both detected in FB. Doxylamine, promethazine, and ibuprofen were also found in both bone and FB.

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In these cases, most drug findings in blood were also detected in bone. In no cases were drug concentrations higher in bone than in blood. These findings demonstrate that bone may be a useful matrix to document drug usage in postmortem cases when traditional specimens are unavailable.

Bone, Skeletal Tissue, Alternate Specimens

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