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### **K73 Determination of Presence and Quantification of Ketamine, Norketamine, and Dehydronorketamine in Dosed and Buried Rat Remains at Different Stages of Decomposition**

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After attending this presentation, attendees will understand the potential to detect and quantify ketamine in buried and decomposing remains using a three-step homogenization, Solid Phase Extraction (SPE), and a Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS) method. Attendees will also become aware of the effects of postmortem interval and body parts on detection.

This presentation will impact the forensic science community by demonstrating the effects of burial on the distribution of drugs and enabling the toxicological analysis of organs in buried and decomposing bodies.

To date, several studies have been carried out to identify drugs in decomposed/decomposing remains, yet few studies on buried remains have been published. Studies on buried remains have tested only skeletal tissue in controlled testing environments; no other tissue matrices sampled from buried remains have been tested.

The goal of this study was to determine if ketamine could be detected in soft tissues collected from buried rats, and if so, in what quantity compared to the dosage given. Rats were dosed at three levels: 20mg/kg (n=13); 30mg/kg (n=13); and, 40mg/kg (n=13). Control/untreated animals were also tested (n=4). Injections were made daily for ten days. Rats were then euthanized, two sets (n=21) at an hour after the last injection and one set (n=15) ten days after the last injection. One rat at each dosage (0mg/kg, 20mg/kg, 30mg/kg, and 40mg/kg) was analyzed without burial for comparison. The remaining rats were buried and then exhumed at different stages of decomposition (77, 188, 293, 793, and 3,104 Accumulated Degree Days (ADDs) after burial) to determine the effect of burial length and stage of decomposition on drug distribution and detection. The rats were dissected and samples were taken from the brain, heart, and liver. When those were no longer discernible, the general viscera in the areas of the brain, heart, and liver were sampled. At 77 and 188 ADDs, organs were still intact. By 293 ADDs, some organs had started to liquefy and by 793 ADDs, most organs were liquefied. Brain samples were most affected by decomposition and were not available from some animals in the later stages of the research. Samples were weighed, diluted with saline (1:1 for brain and liver, 1:2 for heart), and homogenized using a Biotage® Bead Ruptor 24. The samples were then centrifuged and the drugs were extracted from the supernatant using SPE. The amount of drug present, ketamine, norketamine, and dehydronorketamine, was quantified in soft tissues using LC/MS/MS with ketamine-D4 as the internal standard.

As expected, ketamine, norketamine, and dehydronorketamine were not detected in the tissue samples from the rats that were euthanized ten days after the last dose. Ketamine and its metabolites were detected in samples from the dosed rats that were not buried and exhibited a dose-response relationship. The drugs were also detected in most of the buried rat tissue samples, with higher concentrations in the higher dosed rats. Where discernable tissue was available, liver concentrations were found to be higher than the heart and brain concentrations.

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#### **Ketamine, Buried Remains, LC/MS/MS**