



K33 Methadone Detection in Postmortem Oral Swab Samples

Valerie M. Hummert, BS*, Marshall University, Forensic Science Center, 1424 3rd Avenue, #5, Huntington, WV 25701; David J. Clay, BA, Myron A. Gebhardt, MS, Kristen M. Bailey, MS, and James C. Kraner, PhD, Office of the Chief Medical Examiner, 619 Virginia Street, West, Charleston, WV 25302

After attending this presentation, the attendees will have an understanding of the use of oral swab samples obtained after death for the detection of methadone.

The presentation will impact the forensic community and/or humanity by demonstrating how oral swabs may be valuable in establishing methadone use in cases of fatal drug overdose.

Recent studies have shown that oral fluid samples are useful for detecting drug use. The purpose of this study was to evaluate whether a specific drug, methadone, could be detected in oral swab samples obtained after death. At present, methadone is among the most commonly detected drugs in fatal drug overdoses in West Virginia.

Oral swabs were obtained in cases in which the cause of death was suspected to be drug-related. Autopsy technicians collected the samples using standard laboratory cotton-tipped swabs by rubbing the swab along the buccal mucosa. Samples were eluted by vortexing with 1.0 mL of methanol and were centrifuged to remove debris. The supernatant was collected, dried under nitrogen, and reconstituted in 0.10 mL of methanol. GC/MS analyses were performed using an Agilent 6890 gas chromatograph interfaced with a 5973 mass-selective detector.

Saliva collected from non-methadone using donors was used for validation with deuterated methadone as an internal standard. Preliminary experiments demonstrated that an unidentified substance present in the swabs co-eluted with methadone. Several attempts were made to modify GC parameters in order to circumvent this interference. These were unsuccessful in resolving the two compounds; therefore, selected-ion monitoring was employed for analysis of methadone in the swabs. Target and qualifier ions acquired for methadone and deuterated internal standard were: methadone 72, 294, 223; methadone-*d*9 78, 226, 178. The analysis had a linear range of 36.5 ng/swab to 365 ng/swab ($r^2=0.991$) and a limit of detection of 29.2 ng/swab. Precision of the assay was demonstrated with intraday ($n=3$) and interday ($n=3$) coefficients of variation of 5.72% and 13.4%, respectively, using a control containing 292 ng/swab. Average methadone recovery was 27.3% when spiked saliva (0.1 mL) was added to the swab.

Cases that were confirmed to be methadone-positive and quantitated in blood were chosen for the study. The average weight of material collected on the swab was $83 \text{ mg} \pm 41 \text{ mg}$.

All calibrators and controls were required to be $\pm 20\%$ of their intended values. Twenty-six case samples were analyzed with a maximum of five case samples included in each assay. Assays also included four calibrators (36.5, 73.0, 183, 365 ng/swab), two positive controls (54.8, 292 ng/swab), and one negative control (saliva with no methadone added).

Methadone was detected in 17 of the 26 samples, three of which were below the LOQ ($< 36.5 \text{ ng/swab}$). The amount of methadone in the samples varied from 38.9 to 333 ng/swab. The methadone metabolite, EDDP, was not studied, but it was noted that methadone was not detected in any of the swab samples from cases for which EDDP in blood was found to be below our limit of detection, 0.01 mg/L.

Methadone, Oral Fluid, Postmortem