K19 Preparation of Oral Fluid for Quantitative Determination of Opiates and Amphetamines by LC-MSMS

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After attending this presentation, attendees will better understand critical issues related to the analysis and quantitation of drugs of abuse in oral fluid.

This presentation will impact the forensic sciences community by expanding the current knowledge on issues related to the analysis of drugs in oral fluid and the distribution of drugs in oral fluid.

Keeping in mind that: (a) substitution therapy policy has recently been implemented in Taiwan; and, (b) oral fluid as an alternate specimen for monitoring drug use has attracted considerable interest, this project was carried out to develop a sample preparation method for effective analysis of opiates and amphetamines in oral fluid by the liquid chromatography-tandem mass spectrometry (LC-MSMS) technology.

Various heating and deproteinization parameters were evaluated for their effectiveness in: (a) removing forth, contaminations, and protein;
(b) preserving original drug composition in the specimen; and, (c) carrying out direct electrospray LC-MSMS analysis. Oral fluid specimens were first processed by the sample preparation protocol, then analyzed by a LC-MSMS system (Agilent 6410 Triple Quadrupole Mass Spectrometer with an electrospray interface and an Agilent 1200 RRLC System) using an Agilent Zorbax SB-Aq (2.1 mm ´ 150 mm, 3.5 µm particle) analytical column operated at 40 °C. The mobile phases adapted for gradient elution are: (A) methanol and (B) 0.1% (v/v) formic acid in water.

The established protocol achieved 1 ng/mL as the method's limit of detection for amphetamine, methamphetamine, 6-acetylmorphine, 6- acetylcodeine, morphine, and codeine. The method's limit of quantitation was 1 ng/mL for the first four compounds listed above and
2.5 ng/mL for morphine and codeine. The method was also successfully applied to the analysis of 34 oral fluid specimens collected from patients participating in the substitution therapy program following the institution's IRB guidelines. Data generating by the "sample preparation/direct LC-MSMS" protocol were superior to those obtained by portable testing devices and gas chromatography-mass spectrometry approaches. For example, one portable testing device could only identify 3 amphetamines and 1 opiates positives, while this method hereby developed quantitated the presence of methamphetamine, amphetamine, morphine, 6-acetylmorphine, and codeine in 20, 17, 7, 3, and 1 specimens. With the limited size of specimen available, the GC- MS approach could not detect the presence of the drugs of interest in many of the specimens that were found (by the newly developed methodology) to contain these dugs at low ng/mL concentration levels.

Oral Fluid, Drugs of Abuse, Liquid Chromatography-Tandem Mass Spectrometry

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