K46 Detection of Carboxylated Metabolites of XLR-11, UR-144, and Their Pyrolysis Products in Oral Fluid

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The goal of this presentation is to inform attendees about the complexity of analyzing thermally unstable synthetic cannabinoids like XLR-11 and UR-144. The possibility of monitoring synthetic cannabinoid metabolites in oral fluid and its potential applications will be discussed.

This presentation will impact the forensic science community by demonstrating that synthetic cannabinoid metabolites can be detected in oral fluid specimens and their detection can possibly prove useful in avoiding passive exposure defense in drug court cases.

Introduction and Objectives: Synthetic Cannabinoids (SC) are the most frequently found group of emerging drugs in routine testing. Like marijuana, they are ingested by smoking, produce similar subjective effects, and, hence, are referred to as "Synthetic Marijuana." XLR-11 and UR-144 are currently the most popular but thermally unstable components in "Synthetic Marijuana" preparations. In this study, detection of carboxylated metabolites of both UR-144 and its pyrolytic product (UR-144 3,3,4-trimethyl pentenoyl isomer) are reported for the first time. Seventy-four oral fluid specimens previously confirmed positive (32) or negative (42) by Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS) for parent drugs XLR-11 and/or UR-144 were re-analyzed for the presence of UR-144 N-pentanoic acid and UR-144 degradant pentanoic acid, common metabolites of XLR-11 and UR-144.

Methods: Oral fluids were collected using the Quantisal™ device, resulting in a 1:4 dilution. Standard reference materials for carboxy metabolites of UR-144 and its pyrolytic product were obtained from Cayman Chemical®. Acid metabolites for the UR-144 and its pyrolysis product were observed when human liver microsomes were incubated with commercially available XLR-11 and XLR-11 degradant respectively for a separate study, demonstrating these to be common metabolites for XLR-11 and UR-144. Carboxylated metabolites were extracted from 0.5mL of acidified oral fluids (60µL phosphate buffer added, pH 1.8) by liquid-liquid extraction with hexane:ethyl acetate (7:1). The top layer was dried down and re-constituted with 50µl methanol for a 20µL injection onto the LC/MS/MS system in negative Multiple Reaction Monitoring (MRM) mode using Electrospray Ionization (ESI). Separation was performed on a 5µ biphenyl column with 0.7mL/minute flow rate. Mobile phases were 0.1% formic acid with 2mM ammonium formate and 0.1% formic acid with 2mM ammonium formate in acetonitrile. The gradient started at 20% organic, was held for 0.5 minutes and increased to 50%, 60%, and 90% after 0.6, 2.9, and 3 minutes, respectively, before returning to initial conditions at 4 minutes. The method was applied to 74 authentic oral fluid specimens with an administrative cut-off of 10pg/mL.

Results: Full-scan positive mode ionization MS data from microsome incubations of XLR-11 degradant showed a metabolite with the same mass and similar fragmentation to that of UR-144 N-pentanoic acid. In negative ion MRM mode, the same precursor and product ions as the UR-144 N-pentanoic acid standard were present but eluted earlier than the standard. Therefore, this metabolite was first presumptively identified as carboxylated UR-144 degradant and later confirmed by comparison with the synthesized standard. In oral fluids tested, UR-144 N-pentanoic acid metabolite was detected at or above 10pg/mL in 15 (50%) of the 32 specimens positive for parent and 2 (5%) of the 42 negative specimens. The concentrations of the UR-144 acid metabolite ranged from 15-500pg/mL. The carboxylated metabolite for UR-144 degradant was also detected in all specimens positive for UR-144 N-pentanoic acid.

Conclusions: This is the first study reporting detection of carboxy metabolites of the most prevalent synthetic marijuana ingredients XLR-11 and UR-144. Their quantitative evaluation in oral fluid opens up possibilities for application as markers for SC abuse. Further research on their application and perhaps more sensitive techniques are needed to fully exploit their potential in oral fluid testing. Monitoring oral fluid for SC metabolites could prove useful in minimizing passive exposure defense in drug testing cases.

Synthetic Cannabinoids, XLR-11/UR-144, Pyrolysis

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