K6 Simultaneous Detection of Psychedelic Amphetamines in Urine By LC/MS/MS

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After attending this presentation, attendees will be familiar with a technique for the simultaneous detection of eleven designer amphetamines in urine by liquid chromatography/tandem mass spectrometry (LC/MS/MS).

This presentation will impact the forensic science community by providing a new method for the simultaneous detection and quantitation of emerging drugs of abuse in urine.

Psychedelic amphetamines are a relatively new class of designer drug in the United States. These drugs were initially popular in Europe and Asia, but the 2C-, 2CT- and DO- series of amphetamines are now routinely seized throughout the United States. A number of these substances are not scheduled in the Federal Controlled Substances Act, offering users a legal alternative to the more traditional designer amphetamines like 3,4-methylenedioxymethamphetamine (MDMA). Many of these newer designer amphetamines produce profound hallucinogenic effects due to their structural similarity towards both mescaline and amphetamine. The pharmacology and toxicology of these drugs are considerably less studied than their conventional counterparts. Their prevalence among toxicological casework in the U.S. is not known, but many toxicology laboratories do not screen for these substances.

The drugs included in this study were 4-bromo-2,5- dimethoxyphenethylamine (2C-B), 4-methylthioamphetamine (4-MTA), 2,5-dimethoxy-4-ethylamphetamine (DOET), 2,5-dimethoxy-4-iodoamphetamine (DOI), 2,5-dimethoxy-4-methylamphetamine (DOM), 2,5-dimethyoxy-4-ethylthiophenethylamine (2C-T-2), 2,5-dimethyoxy- 4-(i)-propylthiophenethylamine (2C-T-4), 2,5-dimethoxy-4-(n)- propylthiophenethylamine (2C-T-7), 2,5-dimethoxyphenethylamine (2C-H), 2,5-dimethoxy-4-iodophenethylamine (2C-I), and 2,5- dimethoxy-4-bromoamphetamine (DOB). A positive electrospray ionization (ESI) LC/MS/MS procedure was developed to allow simultaneous detection and quantitation of these substances following solid phase extraction (SPE).

Negative urine was fortified with the drugs of interest and extracted using SPE. In the absence of commercially available deuterated analogs, mescaline-d9 was chosen as the internal standard. An alkaline extraction using a copolymeric mixed-mode SPE column was used to isolate the drugs. Optimal drug recoveries were achieved using 2% ammonium hydroxide in 95:5 v/v methylene chloride:isopropanol. Separation was achieved using a C18 LC column and gradient elution (5% methanol in 50mM ammonium acetate and 100% acetonitrile in 50mM ammonium acetate). The total run time was approximately 5 minutes. Data was acquired using the following ions (precursor ions are underlined): m/z 262, 245, 230 for 2C-B; m/z 182, 165, 117 for 4-MTA; m/z 224, 207, 179 for DOET; m/z 322, 305, 105 for DOI; m/z 210, 193, 178 for DOM; m/z 242, 225, 134 for 2C-T-2; m/z 256, 239, 197 for 2C-T-4; m/z 256, 239,197 for 2C-T-7; m/z 182, 165, 150 for 2C-H; m/z 308, 291, 276 for 2C-I; m/z 276, 259, 231 for DOB. Limits of detection for all target analytes were 1-2 ng/mL and limits of quantitation were 1-6 ng/mL. Precision and accuracy was evaluated at 20 and 75 ng/mL. For all drugs, accuracy at 20 ng/mL was 95-115% and CVs were 4.3-10.0% (n=4). At 75 ng/mL accuracy was measured in the range 83-120% and CVs were 3.7-12.0 (n=4). Ion suppression and interferences from common amphetamines and endogenous phenethylamines were included in the study.

The technique allows for the simultaneous low-level detection of eleven psychedelic amphetamines in urine samples. The method will be

further developed to determine the prevalence of these drugs in toxicological casework and to further develop a confirmation and quantitation procedure for these and other designer drugs in blood samples.

Designer Amphetamines, LC/MS/MS, Solid Phase Extraction

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