K50 Current Research Initiatives in Toxicology at the National Institute on Drug Abuse

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After attending this presentation, attendees will be able to describe new research from Chemistry and Drug Metabolism (CDM), detailing National Institute on Drug Abuse (NIDA) findings on urinary cannabinoid excretion, oral fluid cannabinoid stability, Liquid Chromatography with Tandem Mass Spectrometry (LC/MS/MS) confirmation of urinary synthetic cannabinoids, and performance impairment and blood THC concentrations in driving cases.

This presentation will impact the forensic science community by revealing CDM investigations of illicit drug agonists, antagonists, and drug dependence treatment pharmacotherapies. Controlled drug administration studies in drug users were conducted under Institutional Review Board-approved protocols and Investigational New Drug applications from the Food and Drug Administration.

CDM investigates the pharmacodynamics and pharmacokinetics of illicit drug agonists and drug dependence pharmacotherapies. Phase I controlled drug administration studies are conducted in drug users under Institutional Review Board-approved protocols and Investigational New Drug applications from the U.S. FDA. Recently, this study focused on mechanisms of action of cannabinoid agonists, development of evidence-based drug policy, and legislation for oral fluid testing and emerging designer drugs. In this presentation, new research findings are shared on urinary cannabinoid excretion, oral fluid cannabinoid stability, LC/MS/MS confirmation of urinary synthetic cannabinoids, and performance impairment and blood THC concentrations in driving cases.

Twelve chronic frequent and nine occasional cannabis users smoked one 6.8% Δ^9-tetrahydrocannabinol (THC) cigarette. Cognitive, subjective, psychomotor and physiological responses, and urinary cannabinoid pharmacokinetics were characterized. THC, cannabidiol (CBD), cannabinol (CBN), 11-hydroxy-THC (11-OH-THC), 11-nor-9-carboxy-THC (THCCOOH), THC-glucuronide, and THCCOOH-glucuronide concentrations were simultaneously quantified by LC/MS/MS and normalized to urine creatinine. THCCOOH (frequent N=12; occasional N=6), THC-glucuronide (frequent N=12; occasional N=9), and THCCOOH-glucuronide (frequent N=12; occasional N=9) were identified; THC, 11-OH-THC, CBD, and CBN were not detected. Highest concentrations (ng/mg creatinine) in frequent and occasional cannabis smokers, respectively, were: THC-glucuronide 3.5-60.7 and 3.0-35.1; THCCOOH 2.7-17.2 and 0-7.2; and THCCOOH-glucuronide 146-548 and 20.8-298. Concentration-time curves for the excretion of urinary cannabinoids and the presence of potential markers for recent cannabis smoking will be presented.

Analyte stability is critical for interpreting drug concentrations, although there are few data on cannabinoid stability in oral fluid, an important new drug testing matrix. Cannabinoid stability in authentic oral fluid collected with the StatSure® and Oral-Eze® collection devices after controlled smoking of one 6.8% THC cigarette was evaluated. Stability pools were prepared for each participant (n=16) by combining oral fluid collected in the first 13.5 hr. Pools were aliquoted into polypropylene cryotubes and stored at room temperature (RT), 4°C, or -20°C. Baseline specimens were quantified within 24 hr, and the remaining aliquots analyzed after one week at RT and 4°C, and four weeks at 4°C and -20°C. Specimens were considered stable if concentrations were within ± 20% of baseline. Specimens collected with the Oral-Eze® and StatSure® devices were stable for THC and THCCOOH at 4°C for one week and one month, while after longer frozen storage a small number of specimen concentrations increased or decreased more than ± 20%. Mean THC and THCCOOH oral fluid concentrations were 93.0 – 96.7% of target after refrigeration for one or four weeks, and 89 – 117% (THC) and 81 – 110% (THCCOOH) for one month frozen. Elution and stabilizing buffers in oral fluid collection devices help maintain cannabinoid concentrations in oral fluid, as well as improving cannabinoid recovery from the collection pad.

Synthetic cannabinoids are an important new designer drug class. Assays are needed to identify these drugs in human urine. An LC/MS/MS method was developed for the qualitative confirmation of ten synthetic cannabinoids (JWH-018, JWH-073, JWH-081, JWH-122, JWH-200, JWH-210, JWH-250, AM 2201, and RCS-4) and their hydroxyalkyl, hydroxyindole, and carboxy metabolites in human urine. Specimen preparation includes hydrolysis and protein precipitation, followed by monitoring of a single MRM transition in a survey scan that triggers an enhanced product ion (EPI) scan at three different collision energies. This information-dependent acquisition experiment is conducted on an ABSciex 5500 QTrap. Qualitative results from several hundred authentic urine specimens will show prevalence of parent and metabolites and metabolite patterns.

Finally, this latest investigation on cannabis effects on driving is presented. In collaboration with other toxicologists, driving under the influence of drugs (DUID) cannabis cases were compiled and analyzed and police reports on apprehended drivers under the influence of cannabis and blood THC concentrations were evaluated. Representative individual case reports with varying THC concentrations are presented, as well as aggregate/summary statistics. The case reports focus on cannabis-only cases, to avoid complications imposed by polypharmacy.

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Urine Cannabinoids, Oral Fluid Stability, Designer Drugs

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