K30 Toxicological Analysis of Synthetic Cannabinomimetic Spice Drugs

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After attending this presentation, attendees will be informed of an LC/MS/MS method for determining the concentration of "spice" drugs in forensic blood and urine specimens.

This presentation will benefit the forensic science community by providing a method for qualitatively and quantitatively detecting seven emerging indole cannabinoid drugs of abuse using liquid chromatography-tandem mass spectrometry (LC/MS/MS). The lack of methods available to analyze these drugs makes detection difficult in suspected cases of "spice." Therefore, as the prevalence of use increases, the need for validated detection methods becomes important.

Synthetic cannabinomimetic drugs have been studied primarily for their activity as CB_1 and CB_2 cannabinoid receptor agonists.
Additionally, their strong binding affinity for CB_1 receptors has made

these synthetic drugs potent marijuana alternatives which have become increasingly popular in recent years. Many of these drugs are sold as herbal incense under the name "spice" or more commonly "K2" in the United States. JWH-018 is the most commonly found drug in these herbal blends. The drugs analyzed in this study include JWH-015, JWH- 018, JWH-019, JWH-073, JWH-200, JWH-250, and WIN55212-2.

"Spice" toxicity can present itself in conflicting psychological states such as nausea, excitability, sedation, and panic. Physiological changes can include sweating, tachycardia, dyspnea, and xerostomia. Numerous hospitalizations as well as a suicide have been following reported acute doses of "spice." Furthermore, long term effects include panic attacks, blurred vision, muscle spasms, and a case of diagnosed dependence syndrome.

Liquid-liquid extractions were used to extract the drugs from blood and urine. Drug standards were spiked into negative blood and urine specimens. Various extraction conditions were compared in order to optimize extraction efficiencies of the drugs in both blood and urine. In the end, pH10 sodium borate buffer and ethyl acetate provided the best extraction efficiency. LC/MS/MS was used to analyze the extracted drugs and develop a method to qualitatively and quantitatively identify the drugs. Prior to LC/MS/MS analysis, drug optimization on the instrument was performed in order to select the appropriate qualifier and quantifier ions for each drug as well as the fragmentor and collision voltages. To ensure optimal chromatography, diazepam-D5 was chosen as the internal standard after comparison with hydrocodone-D3 and fentanyl-D5. Methanol with 0.1% formic acid was chosen as the mobile phase as it allowed for adequate separation of the compounds of interest.

The method was validated using the laboratory's validation guidelines. A five-point calibration curve was developed from 1-250 ng/mL. Linear ranges were from 1-100 ng/mL for all drugs except JWH- 200 and WIN55212-2 which maintained linearity from 1-250 ng/mL with R^2 greater than 0.995 for all drugs. To validate the method, two extractions were performed on separate days. Accuracy and precision were calculated at 10 ng/mL and 100 ng/mL using three replicates for each concentration. LOQ for all drugs was 1ng/mL.

This presentation provides a rapid, sensitive method for determining the presence and concentration of several indole-based cannabinomimetic drugs in blood. The combination of chromatographic separation and ion monitoring with LC/MS/MS allows for multiple drugs to be accurately detected. This method can prove useful with the increasing rate of synthetic cannabinomimetic drug use in the population.

Spice, Cannabinoid, LC/MS/MS

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