

# The Importance of Collaborations Between Private and Public Health Laboratories in Response to Designer Drugs of Abuse

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## ABSTRACT

The increasing emergence of synthetic drugs is a difficult challenge for public health officials, clinicians, and law enforcement. The collaboration between researchers at the Arkansas Department of Health - Public Health Laboratory (ADH-PHL) and Cayman Chemical Company has characterized biomarkers for the development of validated human specimen testing. This collaboration has produced peer-reviewed publications, educational presentations, and development of metabolic reference standards for clinical testing. Importantly, the Center for Drug Detection and Response has been established this critical partnership to support clinical testing and impact legislative policy.

## BACKGROUND - PRIOR TO 2010

### Arkansas Department of Health

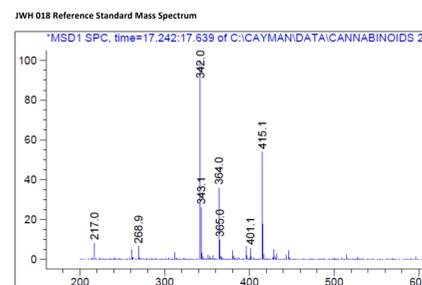
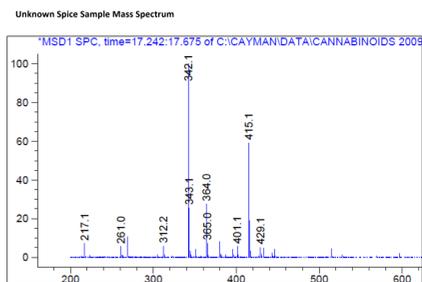
- 2004 - 80,000 sq ft new laboratory build
- ADH-PHL Laboratories perform more than 800,000 tests on 500,000 specimens
- Toxicology Laboratories began first studies to identify synthetic cannabinoid metabolites in 2010
- Manages the Center for Drug Detection and Response at the University of Arkansas for Medical Sciences ([www.uams.edu](http://www.uams.edu))

### Cayman Chemical Company

- 1980-present - Development of research tools to help researchers understand basic biological mechanisms
- Cayman has been actively involved in the cannabinoid research society since the 1980's, providing tools to probe cannabinoid receptors
- Began synthesis of synthetic cannabinoids for use as forensic standards in 2009

ICRS 2010 "Automated Ion Trap Screening Method for Identification of Synthetic Cannabinoids (SCs) in Herbal Incense Products"

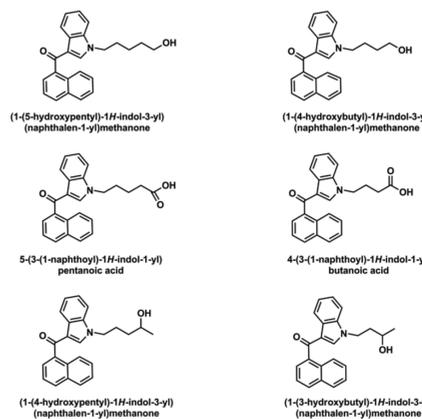
- JWH 018 and 5 other SCs were identified in 5 incense products



## CANNABINOID METABOLITE IDENTIFICATION

Initial studies at ADH-PHL were aimed at identification of the urinary metabolites of JWH 018 and JWH 073

- 6 suspect metabolites of JWH 018 and 6 metabolites of JWH 073 were synthesized by Cayman
- For each compound, 3 major metabolites were confirmed and quantified in human urine samples by ADH-PHL
- Additional studies by ADH-PHL demonstrated uridine diphosphate-glucuronosyltransferase (UGT) activity, suggesting a likely mechanism of clearance



### Reference List

Moran, C.L., et al., Quantitative Measurement of JWH-018 and JWH-073 Metabolites Excreted in Human Urine. *Anal Chem*, 2011, **83**(11): p. 4228-36.  
 Chimalakonda, K.C., et al., Solid-phase extraction and quantitative measurement of omega and omega-1 metabolites of JWH-018 and JWH-073 in human urine. *Anal Chem*, 2011, **83**(16): p. 6381-8.

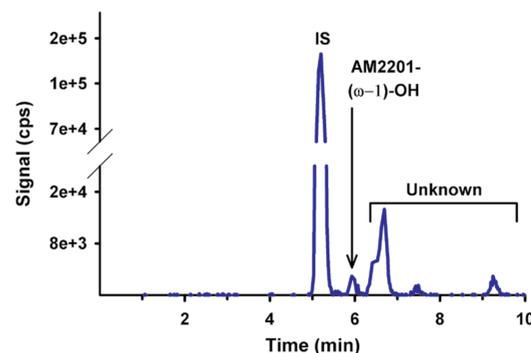
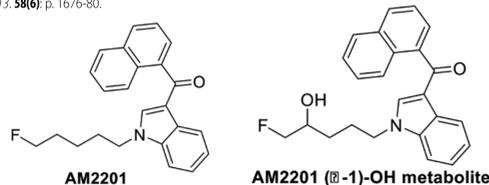
## SECOND GENERATION SYNTHETICS

DEA Scheduling of JWH 018, JWH 073 and several other synthetic cannabinoids led to the introduction of a second generation of synthetic cannabinoids including AM2201, the most popular of the second generation synthetic cannabinoids

ADH-PHL demonstrated that AM2201 could undergo similar metabolism to JWH 018 in recombinant cytochrome P450s and human liver microsomes

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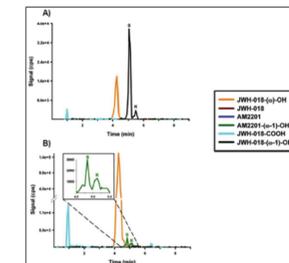
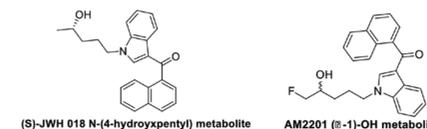
Patton, A.L., et al., K2 Toxicity: Fatal case of psychiatric complications following AM2201 exposure. *Journal of Forensic Sciences*, 2013, **58**(6): p. 1676-80.



## STEREOSELECTIVE METABOLISM OF JWH 018 & AM2201

Additional work between ADH-PHL and Cayman resulted in development of a chiral method to evaluate the enantiospecificity of human metabolism of JWH 018 and AM2201, which may be relevant for understanding clinical toxicity of synthetic cannabinoids.

- A.) Human urine sample positive for JWH 018 ( $\omega$ -1) metabolite. The S-enantiomer is predominantly formed (as glucuronide).
- B.) Human urine sample positive for AM2201 ( $\omega$ -1) metabolite. A mixture of enantiomers is formed.

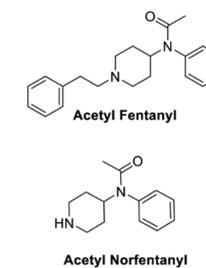


### Reference List

Patton, A.L., et al., Targeted Metabolomic Approach for Assessing Human Synthetic Cannabinoid Exposure and Pharmacology. *Anal Chem*, 2013, **85**(19): p. 9390-9.

## Acetyl Fentanyl

- Acetyl fentanyl (aka desmethyl fentanyl) is a designer synthetic opioid analog of fentanyl that is 5 times more potent than heroin.
- Acetyl fentanyl has been identified in seized pills in Montreal (May 2013 CBC News report) and has been implicated in 10 deaths in Rhode Island since March 2013 (Morbidity and Mortality Weekly 62(13), 2013).
- Based on the metabolism of fentanyl to norfentanyl the suspected metabolite of acetyl fentanyl was anticipated to be acetyl norfentanyl, however this had not been demonstrated.
- Acetyl fentanyl will give a false positive result in the fentanyl ELISA test therefore GC-MS or LC-MS/MS methods are necessary for confirmation and quantification of this analog and its metabolite.



## Acetyl Fentanyl LC-MS/MS METHOD

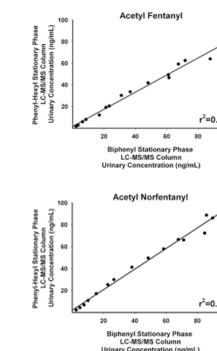
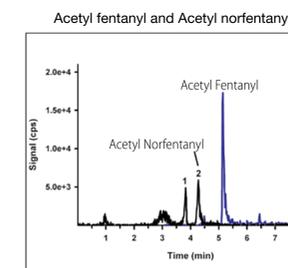
### Method

4.6x50mm, 2.6um Phenomenex Ph-Hex column at 30°C • MP A 10mM ammonium formate in water • MP B 0.1% formic acid in methanol • 600µL/min flow rate 0-4min 2-98%B, 2min 98%B • API 4000 Qtrap/Positive ESI • Acetyl fentanyl SRM 323->188, 105amu • Acetyl norfentanyl SRM 219->84, 136amu

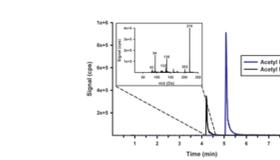
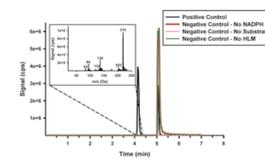
Concentrations are expressed as response ratios of each analyte over their corresponding -d<sub>5</sub> deuterated Internal Standard

### Reference List

Patton, A.L., et al., Quantitative Measurement of Acetyl Fentanyl and Acetyl Norfentanyl in Human Urine by LC-MS/MS. *Anal Chem*, 2014, **86**(3): p. 1760-6.



## Acetyl Fentanyl METABOLIC STUDIES



### Reference List

Patton, A.L., et al., Quantitative Measurement of Acetyl Fentanyl and Acetyl Norfentanyl in Human Urine by LC-MS/MS. *Anal Chem*, 2014, **86**(3): p. 1760-6.

## CONCLUSIONS AND FUTURE WORK

### Conclusions

- A collaboration between ADH-PHL and Cayman Chemical was established to address some of the analytical challenges surrounding emerging drugs of abuse.
- This work has successfully led to the identification of the major metabolites of several emerging synthetic cannabinoids.

### Future Work

Prepackaged toxicology kits for mass spectrometric analysis of drugs of abuse and their metabolites ("ToxBox") ADH-PHL is rapidly validating methods for toxicological profiling and quantitative analysis of drugs of abuse.

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