

## Detection, confirmation, and quantitation of several drugs of abuse in urine using LC/MS/MS

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**AIM:** Analysis of urine for detection and confirmation of illicit drug use is increasing. Typically, samples are analyzed for detection of commonly abused illicit drugs: THC, cocaine, opiates, amphetamines, and PCP. The initial screening of samples is often performed using immunoassay, with confirmation and quantitation often performed by GC/MS. The cutoff levels range from 10 ng/mL for 6-MAM to 2000 ng/mL for morphine and codeine. The use of LC/MS/MS to detect, identify, and quantitate drugs of abuse in urine is investigated.

**METHODS:** The analytes of interest and their respective cutoff levels are as follows: THC-COOH at 15 ng/mL; benzoylecgonine at 150 ng/mL; morphine at 2000 ng/mL; codeine at 2000 ng/mL; 6-MAM at 10 ng/mL; phencyclidine at 25 ng/mL; amphetamine at 500 ng/mL; and methamphetamine at 500 ng/mL. A Shimadzu HPLC stack was interfaced to an Applied Biosystems/MDS Sciex 3200 Q TRAP hybrid triple quadrupole/linear ion trap mass spectrometer. Mobile phases A and B were water and ACN, respectively, with 0.1% formic acid added to each. A flow rate of 0.500 mL/min was used and 10  $\mu$ L of sample was injected for analysis. Separation was achieved on either a Betabasic-18 or Aquasil C18 column, 2.1 mm x 50 mm. Samples consisted of urine spiked with the analytes of interest at various levels. 1 mL of spiked urine was diluted with an equal volume of 10% aqueous methanol and injected for analysis without additional sample preparation.

**RESULTS:** All analytes were successfully detected and quantified well below the required cutoff levels and the LOQ for most analytes was 1 ng/mL or lower. All eight analytes could be analyzed in five minutes. The simple sample preparation saved time and minimized sample handling, which also helped minimize any error that could be introduced into the analysis. Interference from the matrix was not significant and did not affect the retention time of interest for any compound. The linear ion trap functionality of the instrument was used to generate high quality MS/MS spectra of the purported analytes. These spectra could be searched against a library for unambiguous identification and confirmation.

**CONCLUSION:** A simple and quick LC/MS/MS method was developed to analyze urine for illicit drug use. Both qualitative and quantitative experiments could be performed and sample preparation was reduced to simple dilution of the sample. Quantitation limits were on the order of 1 ng/mL or better, which was well below the required limits for all analytes. Confirmation could be achieved by acquisition of MS/MS spectra of each detected analyte and using a library search to confirm its identification.

**KEYWORDS:** *LC/MS/MS, Drugs of abuse, Confirmation, Quantitation, Urine*

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