Automated Extraction of Carboxy THC from Urine on an ASPEC XL4™ Solid-Phase Extraction System without the Use of SPE Cartridges

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AIMS: Procedures for the extraction of 11-nor-Δ⁹-tetrahydrocannabinol-9-carboxylic acid (carboxy THC) from urine by liquid-liquid extraction and solid-phase extraction (SPE) methods have been well documented and employed worldwide. Automated SPE method for extracting carboxy THC from urine is now popular, due to its better reproducibility, increased throughput, and reduction in labor costs. However the cost of SPE cartridges is significant, especially for laboratories like ours dealing with large volume of specimens. The aim of this study was to develop an alternative automated method for extracting carboxy THC from urine without the use of SPE cartridges.

METHODS: An automated extraction protocol for extracting carboxy THC from urine was developed on the ASPEC XL4™ Solid-Phase Extraction System, employing liquid-liquid extraction principle. The process involves an initial clean-up solvent extraction step of hydrolyzed urine at basic condition (pH > 10) to remove neutral and basic interfering substances and a final solvent extraction step at acidic condition (pH < 2) for carboxy THC. Extraction is achieved by programming the System utilizing its liquid handling functions such as dispensing, mixing, and loading. The collected fractions are evaporated and derivatized with PFPA/PFPOH. The derivatives, after drying, are reconstituted in hexane for GC-MS analysis.

RESULTS: The extraction efficiency of the method (84%) was comparable to that reached with manual liquid-liquid extraction (88%) and with automated SPE (84%) on ASPEC XL system (Langen et al. 2000. J Anal Toxicol 24: 433-437). Excellent data concordance ($R^2 > 0.995$) was found for two patient specimen sets (n = 52) using this method and the manual liquid-liquid extraction method. The limit of detection, limit of quantitation, and the upper limit of linearity of the developed method were found at 1, 2, and 1500 ng/mL respectively. There was no detectable carry over after 10,000 ng/mL analyte. For a batch of 76 samples, the process uses 450 mL hexane/ethyl acetate (5:1) as extracting solvent and 1 L 30% methanol in water as rinsing solvent and takes 5 hrs to complete.

CONCLUSIONS: The method is comparable to both manual liquid-liquid extraction and automated SPE methods. It removes the costly SPE cartridge item from the automation process. It also removes the very demanding manual handling task of capping SPE cartridges for the ASPEC XL4™ system. The developed method is proved to be a simple, speedy and economical alternative to the currently popular automated SPE method in drug analysis of urinary carboxy THC.

Keywords: Carboxy-THC, Urine, Automation