A Rapid iMethod™ Test for the Analysis of THC-COOH in Urine

iMethod™ Test for THC-COOH using the Spark Pico System Version 1.0 for Cliquid® Software

The following description outlines the instrument requirements and expected results obtainable from the Spark Holland iMethod™ Test for the quantification of THC-COOH when using a Spark Holland Symbiosis PICO integrated online SPE and HPLC system with a mistral column oven, an AB SCIEX 4000 series (API 4000™ or 4000 QTRAP®) LC/MS/MS instrument.

Cannabis is one of the most widely-used illicit drugs in the world. It is the most frequently detected drug in cases of driving under the influence of drugs in several countries. Cannabis use is detected by identifying the presence of metabolites of the major psychoactive constituent of marijuana, Δ9-tetrahydrocannabinol (THC), in biological fluids, for example, urine. The major metabolite found in urine is 11-nor-Δ9-tetrahydrocannabinol-9-carboxylic acid (THC-COOH), which exists in both the free and glucuronide form. The continued increase of cannabis abuse has created a greater demand for sensitive, rapid, and reliable methods for confirming the presence of this drug in biological samples.

Sample preparation is based upon dilution of the urine sample followed by hydrolysis of any glucuronide present and automated on-line clean up using a Spark HySphere C8EC-SE sorbent cartridge upon injection in the Symbiosis PICO system. More in depth sample preparation, and instrument parameter information is included as part of the standard operating procedure provided with the method, as is the required analytical columns.

The on-line clean up solvents and mobile phase consists of the use of water, acetonitrile and formic acid with sample clean up on a Spark HySphere™ C8EC SE cartridge and separation on a Phenomenex Gemini C6-Phenyl 50 X 3 mm 3 µ HPLC column. An example chromatogram of the separation achieved is shown below in figure 1.

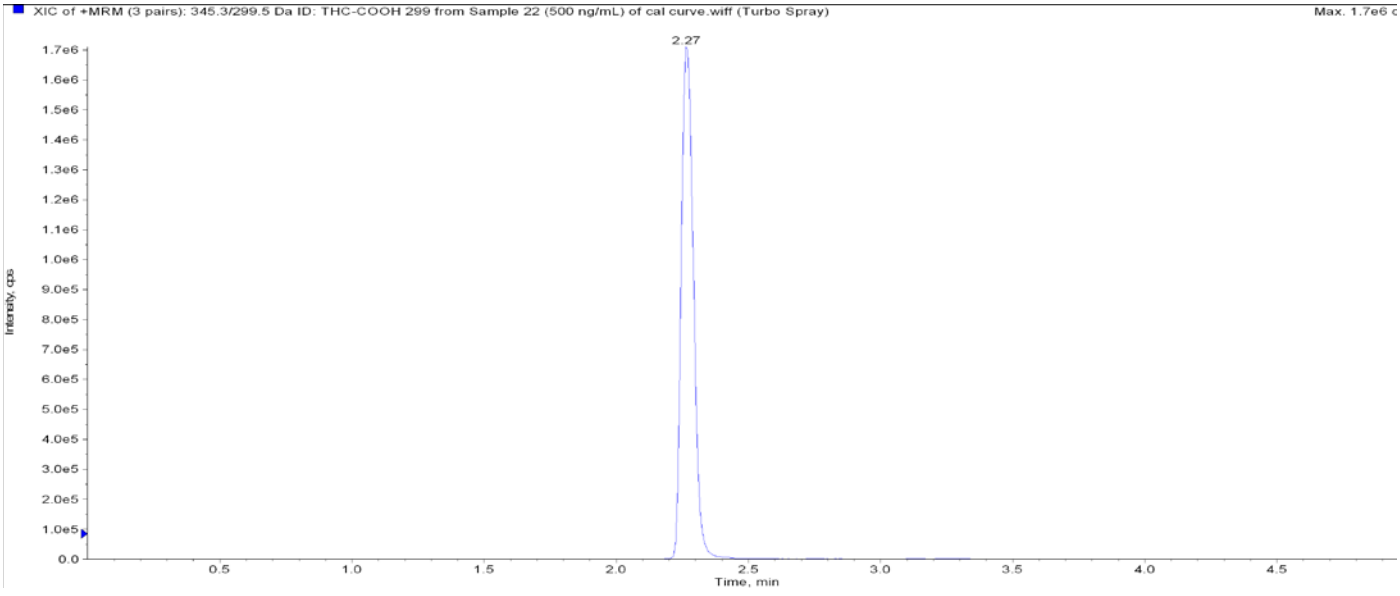

Figure 1: 500 ng/mL THC-COOH spiked in urine

Results

The following calibration curve is representative of the performance obtained on the instrument using the method described here

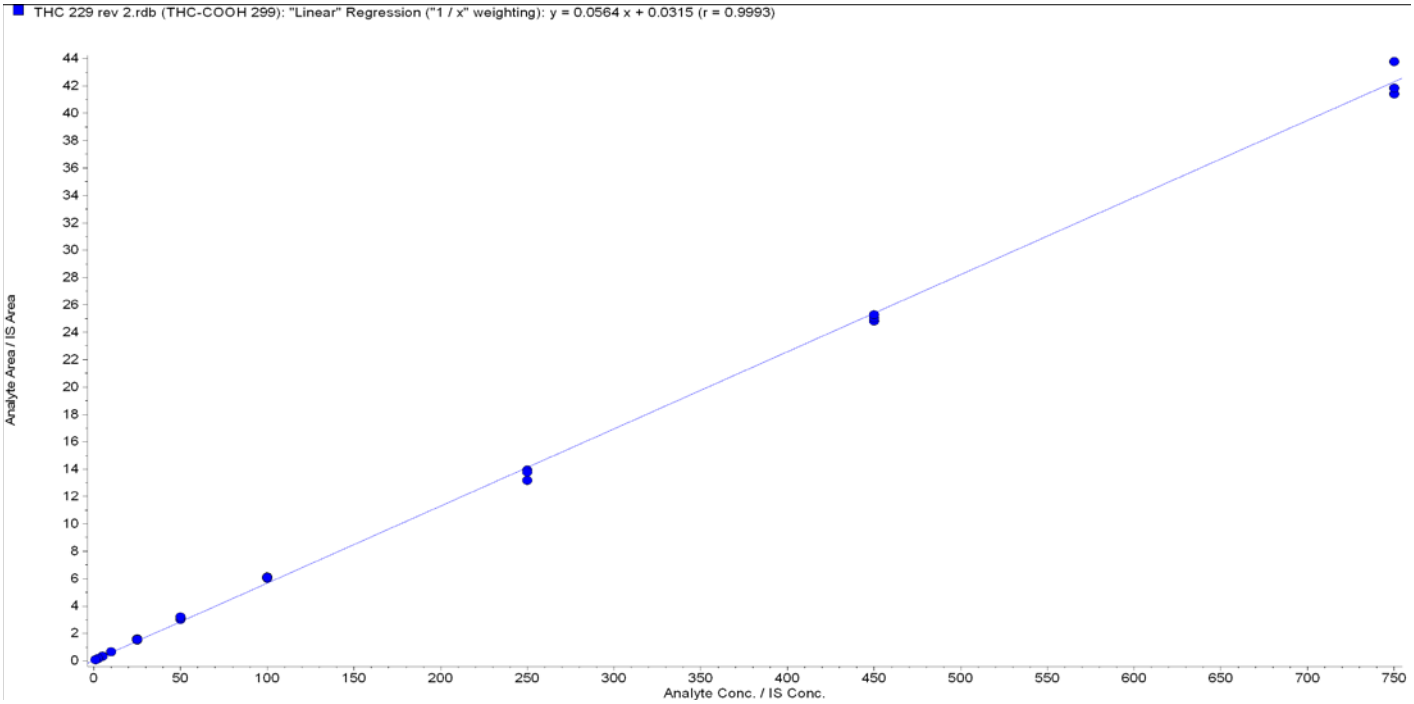

Figure 2: Three combined calibration curves ranging from 1 ng/mL to 750 ng/mL in urine. R=0.9993 with a 1/x weighting created by following the procedure in the Sample Preparation section of the standard operating document provided.

Please note that the results presented above were obtained using a single instrument and single set of standards and samples. Prior to production use, the method should be fully validated with real samples, and the results here may not be typical for all instruments. Variations in LC column properties, chemicals, environment, instrument performance and sample preparation procedures will impact performance, thus these results should be considered as informative rather than representative.

System Requirements

In order to run this method as outlined above, the following equipment and reagents are required:

- An AB SCIEX 4000 Series LC/MS/MS System
- Spark Holland Symbiosis PICO Online SPE HPLC system with a Mistral column oven
- THC-COOH standards (www.cerilliant.com)
- LC/MS grade methanol, and acetonitrile
- A Phenomenex Gemini C6-Phenyl 50 X 3 mm 3 µ HPLC column
- Spark HySphere C8EC-SE sorbent cartridges
- Pipettes and standard laboratory glassware

Please note that the Phenomenex HPLC column is required but not included with this iMethod™ Test.

Important Note

The purchase and use of certain of the chemicals listed above may require the end user to possess any necessary licenses, permits or approvals, if such are required in accordance with local laws and regulations. It is the responsibility of the end user to purchase these chemicals from a licensed supplier, if required in accordance with local laws and regulations. The suppliers and part numbers listed below are for illustrative purposes only and may or may not meet the aforementioned local requirements. AB SCIEX is not responsible for user's compliance with any statute or regulation, or for any permit or approval required for user to implement any iMethod™ procedure.

The iMethod™ Test described above has been designed by AB SCIEX to provide the sample prep and instrument parameters required to accelerate the adoption of this method for routine testing. This method is provided for information purposes only. The performance of this method is not guaranteed due to many different potential variations, including instrument performance, tuning, and maintenance, chemical variability and procedures used, technical experience, sample matrices, and environmental conditions. It us up to the end user to make adjustments to this method to account for slight differences in equipment and/or materials from lab to lab as well as to determine and validate the performance of this method for a given instrument and sample type. Please note that a working knowledge of Analyst® Software may be required to do so.

Publication number: 1822610-01

Headquarters
353 Hatch Drive | Foster City CA 94404 USA
Phone 650-638-5800
www.absciex.com

International Locations
For our office locations please call the division headquarters or refer to our website at
www.absciex.com/offices