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Determination of *Cannabis* compounds in blood samples by headspace solid-phase microextraction and gas chromatography-mass spectrometry

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AIMS: To develop a method for the analysis of Cannabis compounds (THC and THC-COOH) in blood samples using headspace solid-phase microextraction (HS-SPME) followed by on-fiber derivatization and gas chromatography-mass spectrometry (GC-MS).

METHOD: The blood sample is placed into a headspace vial and after the addition of sodium chloride and the deuterated internal standards, the sample is submitted to HS-SPME, using a polydimethylsyloxane (PDMS) fiber. Then, for derivatization, the fiber is directly placed into the headspace of a second vial containing trifluoroacetic anhydride and 1,1,1,3,3,3-hexafluoro-isopropanol. The perfluorated derivatives of the THC and THC-COOH, adsorbed on the fiber, are analyzed by GC-MS on an Agilent 5973 mass spectrometer in electron ionization mode and selected ion monitoring mode using an HP-1MS capillary column.

RESULTS: In order to gain optimal conditions, several parameters have been considered during the method optimization process. They include, among others: a) the influence of the addition of different salts, such as sodium carbonate, sodium chloride, ammonium sulfate and sodium sulfate; b) extraction time and temperature; c) different derivatization reagents, such us trifluoroacetic anhydride (TFA), pentafluoropropionic anhydride (PFPA) and heptafluorobutyric anhydride (HFBA); d) derivatization time and temperature. Sodium chloride was found to be the best salt and TFA the best derivatizing agent. Both provided the highest recoveries and more stable derivatives. The optimized method was validated for the two compounds under study. Quantification limits are 0.25 ng/mL and 0.50 ng/mL for THC-COOH and THC, respectively. Validation parameters were determined at three concentrations -- low, medium and high. Recovery data ranged from 26 to 32.2% for THC-COOH and 21.5 to 30.4% for THC. The intra-assay precision ranged from 2.3 to 6.2% (THC-COOH) and 4.1 to 6.9 % (THC). The inter-assay precision ranged from 3.4 to 6.9 % (THC-COOH) and 5.3 to 9.9 % (THC). With respect to the PDMS fiber, it lasts for around 100 samplings. The efficacy of this method was demonstrated by comparing the results in 10 samples with those obtained by SPE and all of them were similar.

CONCLUSION: The method has demonstrated to be simple; only two steps are involved, rapid, sensitive and specific. It is being routinely applied in our laboratory.

KEYWORDS: *Cannabis, Blood, HS-SPME*

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