Abstract

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Title of Thesis: UHPLC-HRMS analysis of canabinoids in biological matrix

Cannabis is still considered to be the most widespread drug worldwide. The main molecule responsible for its effects is psychotropic cannabinoid Δ^9-tetrahydrocannabinol, which is converted into active metabolite 11-OH-Δ^9-THC and inactive metabolite 11-nor-9-carboxy-Δ^9-THC in the human body. The determination of these three molecules in human serum is one of the basic analytical assays that are performed in toxicological laboratories, as it reflects the state of current drug exposure. Another cannabinoid - cannabidiol (CBD) - is also gaining worldwide awareness, and its therapeutic effects have been the subject of many clinical studies. CBD has no psychoactive effect and its potential for the treatment of a wide range of diseases was first fully acknowledged with the launch of the first drug (Epidiolex®) on the US market in 2018 with CBD being the main active ingredient.

The aim of this thesis is development and validation of the sensitive method for determination of Δ^9-THC,11-OH-Δ^9-THC, Δ^9-THC-COOH, CBD and 7-COOH-CBD in human serum using UHPLC-HRMS. The optimized extraction process consists of protein precipitation with isopropanol followed by liquid-liquid extraction in an acidic environment. The extractant consists of a mixture of *n*-hexane and ethyl acetate (96:4, *v/v*) with the addition of acetonitrile, the acidification of the extraction mixture, which is necessary for the extraction of cannabinoid acids, is achieved by addition of 1M HCl. The method was validated according to the validation protocol published by the European Medicines Agency in 2011, all validation parameters fulfilled the set validation criteria. We succeeded in developing a reliable and sensitive bioanalytical method that is suitable for introduction into clinical practice.

Key words: Cannabinoids, LC-HRMS, Bioanalysis, Validation