O41. Interest of qualitative and quantitative profiling of endocannabinoids for evaluation of their implication in drug addiction process

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Introduction: In the middle of the 90's, the discovery of endogenous ligands for cannabinoid receptors opened a new era in this research field. Amides and esters of arachidonic acid have been identified as these endogenous ligands. Arachidonoylethanolamide (anandamide or AEA) and 2-Arachidonoylglycerol (2-AG) seem to be the most important of these lipid messengers. In addition, virodhamine (VA), noladin ether (2-AGE), and N-arachidonoyl dopamine (NADA) have been shown to bind to CB receptors with varying affinities. During recent years, it has become more evident that the EC system is part of fundamental regulatory mechanisms in many physiological processes such as stress and anxiety responses, depression, anorexia and bulimia, schizophrenia disorders, neuroprotection, Parkinson disease, anti-proliferative effects on cancer cells, drug addiction, and atherosclerosis.

Aims: This work presents the problematic of EC analysis and the input of Information Dependant Acquisition based on hybrid triple quadrupole linear ion trap (QqQLIT) system for the profiling of these lipid mediators.

Methods: The method was developed on a LC Ultimate 3000 series (Dionex, Sunnyvale, CA, USA) coupled to a QTrap 4000 system (Applied biosystems, Concord, ON, Canada). The ECs were separated on an XTerra C18 MS column (50 × 3.0 mm i.d., 3.5 µm) with a 5 min gradient elution. For confirmatory analysis, an information-dependant acquisition experiment was performed with selected reaction monitoring (SRM) as survey scan and enhanced produced ion (EPI) as dependant scan.

Results: The assay was found to be linear in the concentration range of 0.1-5 ng/mL for AEA, 0.3-5 ng/mL for VA, 2-AGE, and NADA and 1-20 ng/mL for 2-AG using 0.5 mL of plasma. Repeatability and intermediate precision were found less than 15% over the tested concentration ranges. Under non-pathophysiological conditions, only AEA and 2-AG were actually detected in plasma with concentration ranges going from 104 to 537 pg/mL and from 2160 to 3990 pg/mL respectively. We have particularly focused our scopes on the evaluation of EC level changes in biological matrices through drug addiction and atherosclerosis processes. We will present preliminary data obtained during pilot study after administration of cannabis on human patients.

Conclusion: ECs have been shown to play a key role in regulation of many pathophysiological processes. Medical research in these different fields continues to growth in order to understand and to highlight the predominant role of EC in the CNS and peripheral tissues signalisation. The profiling of these lipids needs to develop rapid, highly sensitive and selective analytical methods.