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Development of a method based on HPLC-DAD for the detection and quantification of glutamate and gamma-aminobutyric acid

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Introduction: With the increasing incidence of neurodegenerative diseases, the need arose to study several molecules that may be useful to the study of new therapies and to treat the symptoms that come with these diseases, as is the case of depression. GABA and glutamate have a very important role in the homeostasis of the organism and molecules of interest and potential from the point of view of diagnosis are revealed. HPLC is a method that allows the detection and quantification of various analytes in different biological matrices, allowing rapid results with high precision, sensitivity and specificity.

Materials and methods: All chromatographic assays were performed using a Hitachi LaChrom Elite® HPLC system, with separation on a Lichrospher LiChroCART ®

250-4 100 (5 μm) RP-18 column and with DAD detection. The parameters of flow rate, mobile phase composition, temperature and wavelenght settings were varied during the process of the development of the method. The validation of the method was performed according to ICH guidelines, which was tested in standard solutions of GABA and glutamate and in samples of serum, urine and yeast extract.

Results and conclusions: At the end of several assays, the chosen method allowed for a 10 minutes analysis with specific detection of GABA and glutamate and a quantification in the order of μg/mL. Even though the quantification was possible in the standard solutions, the same did not occur in the biological matrices tested. Although it still needs to be optimized for biological matrices, the method developed allows an easy, fast and economically sustainable analysis of GABA and glutamate. It is, to date, the only method with DAD detection that allows the simultaneous detection of GABA and glutamate without recourse to derivatization of the sample.

Keywords: Neurodegenerative diseases, depression, GABA, glutamate, HPLC-DAD