Study of a potential biomarker for Alzheimer's disease by LC-MS-MS

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Introduction

Alzheimer's disease is the most common form of dementia and affects 35 millions of people in the world today. The diagnostic is usually possible after signs of the first symptom. However recent studies show that neuron damage can begin 10 years before this. Thus the research of new biomarkers to detect and identify this disease earlier on is of great interest. In this article, the protein A is studied as a potential as biomarker of this disease in human plasma. Two forms of this protein are analyzed and quantified by micro-LC coupled with tandem mass spectrometry. A parameter X, the ratio of the two protein's forms, is also determined for samples from healthy patients (Control Sample) and patients with Alzheimer's disease (AD Sample). The results show a difference between the two sets of samples.

Material and methods

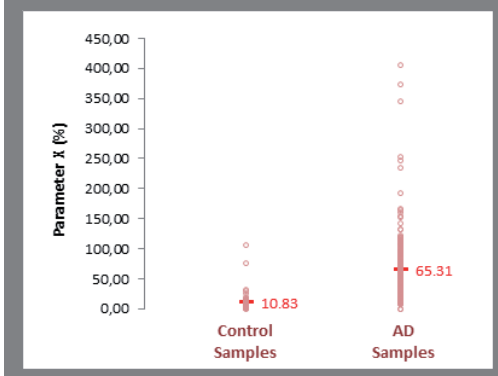
The protein A was digested by trypsin and purified by Solid Phase Extraction (SPE) with an Oasis HLB cartridge 3 cc 60 mg (Waters). Digestion allowed to break down the protein into small molecules, peptides, which are adapted to mass spectrometry analysis. A specific peptide was chosen. Two forms of this peptide were separated and analyzed by micro liquid chromatography (Column C18,3 µm, 1mm*10 cm) coupled with tandem mass spectrometry. A triple quadrupole mass analyzer in Selected Reaction Monitoring (SRM) mode was used. SRM mode is a powerful method for quantitative measurement of target peptides and has been a tool for quantification for number of decades. Moreover, this mode is highly specific and sensitive.
For each from of peptide, three SRM transitions (m/z values selected in Q1 and Q3) were followed but only one was used for quantification.
The SRM transitions are described in Table 1.

PROTEIN A	m/z in Q1	m/z in Q3
form 1	**516,263**	734.4
		621.3
		416.2
form 2	**524,361**	847.4
		750.3
		637.3

Table 1: SRM transitions followed during the analysis. The values in red were used for the quantification.

For each form of the peptide, a peak was obtained on the chromatograms. Each peak was integrated and the parameter X, ratio of the two forms, was deduced. This was repeated for all of the samples. An average X parameter was calculated for the two types of sample.

Figure 1 : Control samples and AD Samples parameters X of human plasma's protein A



Results and discussion

As showed in Figure1, the parameter X of the protein A is less important for Control Samples than for AD samples. An average difference of 54.3 % was observed. These results seem encouraging, however before concluding it is necessary to also study the influence of other parameters such as sample preservation, sampling procedure, and the cleanliness of the equipment.

Conclusion

Protein A (and its parameter X) shows a true potential as a biomarker of Alzheimer's disease. Further studies should be undertaken to confirm these results in order to validate protein A as a new biomarker.

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