Changes in the human hippocampal proteome during Alzheimer's disease.

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Research question and background
Because of the current lack of effective treatment and early presymptomatic diagnostic markers of Alzheimer's disease (AD), detailed insight into disease mechanisms involved at the various stages of AD is highly important. The pathological hallmarks, amyloid beta plaques and intra cellular aggregation of hyperphosphorylated tau in neurofibrillary tangles, are used to objectively stage the disease. In this study we have analysed the proteome of the hippocampus, and in particular the CA1 region and subiculum, as this is one of the most vulnerable and early affected regions in AD, with the aim to identify changes in the human proteome and underlying disease mechanisms.

Methods and tissues used
A total of 40 patients representing all stages of AD (Braak 0 to VI) were selected. Hippocampal regions CA1 and subiculum were isolated from human post-mortem brain tissue using laser capture microdissection and protein lysates were prepared. After separation of the proteins by SDS-PAGE and in-gel trypsin digestion, the extracted peptides were analysed by LC-MS/MS using Orbitrap mass spectrometry. MaxQuant software was used for protein identification and quantification. Cluster analysis was used to identify proteins that are co regulated during the disease course.

Results and conclusion
Analysis of the mass spectrometry data resulted in the quantification of a total of 3216 proteins of which a subset of 372 proteins were found significantly regulated. Known proteins such as GFAP and tau were increased with increased Braak stage, as expected. We also identified a range of proteins that have not been associated with the disease before. Interestingly one group of exhibited an "early up late down" pattern. We could validate our results by immunoblotting and immunohistochemistry.
Newly identified proteins and pathways will provide insight into the complex biology of AD and may lead to new biomarkers and/or therapeutic drug targets.