Neuropeptide profiling in rat hippocampus using capillary 2D-LC-MS/MS

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Capillary two dimensional liquid chromatography with electrospray ionization tandem mass spectrometry (2D LC-ESI-MS/MS) was applied to investigate the influence of hypoxic induced stress upon neuropeptide excretion levels in rat hippocampus. Strong cation exchange chromatography (SCX) was applied in first dimension and reversed-phase chromatography (RP) in the second dimension. Rats of three age groups (8, 32 and 50 weeks) were exposed to different CO_2 influx in the inhaled air to examine any stress-related and age factors in the neuropeptide excretion. MS/MS spectra of peaks detected in the same SCX fraction were submitted to Mascot for identification of the compounds. Multivariate analysis of variance method, 50-50 MANOVA, found significant effects of both influx rate, age and interactions between influx and age as viewed across all peptides. A univariate analysis of variance of individual peptides revealed 5 peptides significant effected according to p-values adjusted for false discovery rate using rotation test ($p<0.05$). In addition we also report results of raw p-values ($p<0.05$) suggesting main effects of influx on 17 peptides, main effects of age on 12 peptides and interaction effects on 14 peptides.