ISCHEMIC INJURY ELICITS THE UNCONVENTIONAL SECRETION OF PROTEIN HALLMARKS OF ALZHEIMER'S DISEASE ONSET AS SEEDS FOR INTERNEURONAL PROPAGATION.

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Objectives: Understanding the ischemic injury contribution to Alzheimer's disease (AD) onset, pinpointing to post-translational modifi cations, turnover alterations, and secretion of proteins and networks identified as AD hallmarks.
Materials: Neuronal cultures prepared from hippocampi of E18-E19 embryos from pregnant Sprague Dawley rats. 5%CO2: 95%N2 gaseous mixtures (Sapio); hypoxia chamber (Billups-Rothenberg).
Methods: DIV8 neurons were subjected to oxygen and glucose deprivation (OGD). After medium replacement with a glucose-free balanced salt solution (ogR), cells were incubated for 3 hours in the chamber saturated with the gaseous mixture. The restoration solution (glucose and B27) was administrated to neurons (ogR) for one hour (R1h) or overnight (R16h).
Results: We observed Tau and amyloid precursor protein (APP) post-translational and protein level modifi cations after OGD/ogR. Tau cleavage and secretion occurs at R1h through microvesicles (MVs) population, including LC3 and/or LAMP1 positive vesicles, marker of autophagy-mediated secretion (exophagy). In MVs extract we also identified α- and ß-C-terminal fragments (CTFs) of APP, and the peptidyl prolyl cis/trans isomerase Pin1.
Discussion: MVs represent an intercellular communication delivering multiple cargo with benefi cial or harmful messages [1]. Thus, differences in MVs contents after OGD/ogR suggest that ischemia leads to a robust upheaval with the autophagic mechanisms activation resultant in neuron-to-neuron transfer of material with neurodegenerative potential.
Conclusions: OGD treatment leads to mis-processed protein unconventional secretion also including exophagy pathway. These proteins represent seeds for protein aggregation according to the "prion-like" propagation mechanism [2] that may represent an early event in AD and proteinopathies.

References:
[1] Kalra H, Drummen GPC, Mathivanan S. Focus on extracellular vesicles: introducing the next small big thing. Int j Mol Sci. (2016);17(2):170
[2] Acquatella-Tran Van Ba I, Imberdis T, Perrier V. From prion diseases to prion-like propagation mechanisms of neurodegenerative diseases. Int J Cell Biol. (2013); 2013:975832