PRESENILINS REGULATE AUTOPHAGIC RESPONSE TO STRESS

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**Introduction:** Macroautophagy or autophagy, a lysosome-dependent degradative pathway for organelles and protein recycling, has been implicated in the Alzheimer's disease (AD) associated neurodegeneration. AD is characterized by the deposition of extracellular β-amyloid peptide (Aβ) plaques. Sequential cleavage of β-amyloid precursor protein by β- and γ- secretases generates Aβ. Mutations in presenilins (PS1/PS2), the catalytic subunits of γ-secretase complex, are major cause of early-onset familial AD. Recent studies show enrichment of PS1 in autophagic vacuoles and also indicate potential role of PS proteins in regulation of autophagic flux.

**Aims:** We address the role of PS in adaptive response to starvation - and oxidative stress - induced autophagy and possible involvement of Beclin-1 in this process.

**Methods:** We used genetic (PS wild-type and knowk-down cells) and pharmacological (γ-secretase inhibitor treatment: DAPT, L-685,458) as models of PS dependence. Autophagy was induced by EBSS medium and H₂O₂ treatment, and assessed via western blotting and immunocytochemistry.

**Results:** Autophagic induction was evaluated in Beclin-1 and Bcl-2 levels. Autophagic flux was shown by LC3 level. After induction of starvation and oxidative stress in cells, MTT (3-(4,5-Dimethythiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay showed enhanced stress-induced cell death in γ-secretase deficiency. Western blotting demonstrated increased Bcl-2 level, Beclin-1 cleavage and decreased phospho-Bcl-2 level in γ-secretase deficient and starvation-induced cells, and these changes were significantly enhanced in combination of both. Starvation-induced LC3-II clearance was promoted by γ-secretase deficiency.

**Conclusion:** Our data indicate that PS regulates autophagic degradation in response to stress. Increased beclin-1 cleavage in γ-secretase impaired cells contributes to enhanced cell death.