The association of palmitoylethanolamide with luteolin reduces neurotoxicity and apoptosis in amyloid-beta stressed hippocampal organotypic slice cultures

E. Esposito[1], I. Paterniti[1], M. Campolo[1], M. Cordaro[1], S. Cuzzocrea[1]

[1]Dept. of Biological and Environmental Sciences, University of Messina

Amyloid beta (Abeta)-induced neurotoxicity is a major pathological mechanism of Alzheimer disease (AD). Activation of glial cells and the consequent neuroinflammatory response is increasingly recognized as a prominent neuropathological feature of AD. In this study, we investigated the altered signal transduction associated with soluble amyloid beta-protein (Abeta) oligomer-mediated neurotoxicity in the hippocampus, which is primarily linked to cognitive dysfunction in AD, after treatment with the association of palmitoylethanolamide (PEA) with luteolin (Lut). In our *ex vivo* e *in vivo* studies we used a compound (PEALut,) obtained by a co-ultramicronization process where PEA, with well known anti-inflammatory effects, was associated with Lut, a widely distributed flavonoid with anti-inflammatory and antioxidant activities, to counteract the neuroinflammation of AD. In *in vitro* studies a human neuroblastoma cell line SH-SY5Y, opportunely differentiated with retinoic acid, was pretreated with PEALut at three different concentrations (0.1-1-10 μM) for 2h. The damage was induced by $A\beta_{1-42}$ (1μM). Twenty-four hrs after damage, we performed cell viability (MTT assay), western blot analysis for IκB-α and NF-κB, and double immunofluorescence staining of glial activation markers with pro-inflammatory cytokines. However, we used organotypic hippocampal slice cultures from P6 mice to assess the neuroprotective effect of PEALut. On day 21 of culturing, organotypic hippocampal slide cultures were pre-treated with PEALut (0.1-1-10 μM) and damaged with $A\beta_{1-42}$ (1μg/ml). After 24hrs, viability assay, NO production and western blot analysis for neurotrophic factors, glial activation, matrix metalloproteinases (MMPs), apoptotic process were performed. Moreover, mRNA expressions of GFAP, CD11beta, TNF-alpha, IL-1beta, iNOS, and COX-2 were investigated. Our data indicate that PEALut compound was able to blunt a $A\beta_{1-42}$ -induced neurotoxicity and to control glial activation. These results suggest that this association may provide an effective strategy for AD.