

The polyphenol oleuropein aglycone modulates PARP1 activity and its interplay with SIRT1

P. Nardiello^{1,2}, D. Pantano² I., Luccarini^{1,2}, C. Miceli¹, C. Nediani¹, M. Stefani¹ and F. Casamenti²

¹Dept. of Experimental and Clinical Biomedical Sciences, University of Florence, Italy

²Dept. of NEUROFARBA, University of Florence, Italy

Poly(ADP-ribose)polymerase-1 (PARP1) activation mediates amyloid- β (A β)-induced cell death in Alzheimer's disease brain. PARP1 is a DNA repair enzyme, normally activated by single strand breaks associated with oxidative stress. Our previous data have shown the beneficial effects of oleuropein aglycone (OLE) against protein/peptide aggregation *in vitro* and in TgCRND8 mice, a model of A β deposition. Recently, it has been reported that A β activates PARP1, that, similarly to SIRT1 requires NAD⁺ as a substrate; moreover, a crosstalk between SIRT1 and PARP1 does exist and PARP1 activity is blocked following SIRT1-dependent deacetylation.

In our study we investigated the state of activation of PARP1 and its network including SIRT1 in 6-month-old TgCRND8 mice treated for 8 weeks with a modified low-fat (5.0%) diet (10 g/day/mouse) either as such or supplemented with OLE (50 mg/kg of diet). N2a neuroblastoma cells (N2a) were also used. Both cultured cells and mouse brain samples were analyzed by immunohistochemistry and Western blotting.

We found a significant accumulation of PAR polymers and increased expression of PARP1 both in the cortex of TgCRND8 mice and in N2a cells exposed to N-methyl-N'-nitro-N-nitrosoguanidine (MNNG). PARP1 activation and the levels of its product, PAR, were rescued to control values by OLE in TgCRND8 mice (P<0.05). In N2a cells MNNG-induced PARP1 activation and PAR formation were abolished by pre-treatment for 24 h with either OLE (100 mM) (P<0.05) or PARP inhibitors 6(5H)-phenanthridinone (PHE, 30 mM) (P<0.001) or N-(6-oxo-5,6-dihydrophenanthridin-2-yl)-(N,N-dimethylamino) acetamide hydrochloride (PJ-34, 20 mM) (P<0.01). In parallel, OLE increased the immunoreactivity and protein levels of SIRT1, both *in vitro* and *in vivo*, (*in vivo*: P<0.001 vs Tg mice; *in vitro*: P<0.05 OLE treated cells vs control cells), reduced NF- κ B (P<0.05) and p53 (P<0.01) expression and levels *in vivo* and enhanced the MNNG-induced autophagy.

In conclusion these findings demonstrate that OLE interferes with the PARP1-mediated cascade of events leading both to apoptotic neuronal cell death via p53, to the inflammatory response via NF- κ B induction, and to increased autophagy via increased SIRT1 activity, further favoring cell survival. These findings implement and extend our previously reported data providing a new mechanism of neuroprotection by OLE with possible implications in AD.