ImmunoTools *special* Award 2013

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Poly(ADP-ribosylation) and Alzheimer disease

Poly(ADP-ribose) polymerase-1 (PARP-1) is the most abundant member of the family of PARP enzymes. It's activated by DNA strand breaks and is involved in the maintenance of genome stability, transcriptional regulation, and inflammation.

When high levels of DNA damage occur, such as during oxidative stress insults, PARP-1 activity increases considerably causing a drastic reduction of NAD+ levels, with consequences on the ATP production and impairment of cell functions. Moreover the excessive activation of PARP-1 under pathological conditions may lead to an accumulation of poly(ADP-ribose) polymers (PAR), a novel signaling molecule that induces cell death.

Alzheimer's disease (AD) is pathologically characterized by the presence of senile plaques of amyloid beta (Aβ) and neurofibrillary tangles in the brain. Aβ is know to induce oxidative stress leading to PARP-1 activation. Aβ is able to activate glial cells leading to an excessive release of pro-inflammatory mediators and cytokines, which in turn trigger a neurodegenerative cascades via neuroinflammation. Moreover, oxidative stress induced by Aβ modulates different cell signaling pathways inside the cells and a prominent signaling pathway is the NF-kB one.

The aim of my PhD project is to investigate the role of PARP-1 in inflammation and programmed cell death processes. To achieve this goal we will use human SH-SY5Y cells neuroblastoma-derived treated with $A\beta_{25-35}$ and 7PA2 cells carrying a double mutated APP that lead to amyloid peptide production. Moreover we will use the BV-2 murine microglia cells which are macrophagic cells within the central nervous system (CNS) able to quickly react to perturbing agents within the brain. All the experiments will be performed in the presence or absence of PARP-1 inhibitors.

The antibodies provided by ImmunoTools could be very useful for studying apoptosis events (Annexin V) and inflammatory events (IL-6, TNFα) through different experimental procedures: by *flow cytofluorimetry* is possible to quantify the fluorescence signals after treatment with $A\beta_{25-35}$; *Western Blot* analysis allow to analyse the protein levels; *immunoprecipitation* could be use to identify a phisycal interaction between two proteins; i*mmunofluorescence* allows to identify the localization of the proteins after the treatment. All the experiments will be carrying out in the presence or absence of PARP-1 inhibitors in order to verify if PARP-1 could prevent the toxic effects of $A\beta_{25-35}$.

ELISA assays will be very helpful to quantitatively measure the amount of cytokine as IL-6 and TNFa. Indeed ImmunoTools antibodies could be used to study the activation of microglia cells which become morphologically and functionally activated and produce a variety of proinflammatory factors, including neurotoxic reactive oxygen species (e.g. superoxide and nitric oxide), eicosanoids, and cytokines (e.g. tumor necrosis factor-α, interleukins 1α, 1β, and 6).

In conclusion recombinant cytokines can be used as control in western blot analysis.

ImmunoTools *special* AWARD for **Sara Martire** includes 25 reagents

FITC - conjugated anti-human IL-6, Control-IgG1, Annexin V,

PE - conjugated anti-human IL-6, Control-IgG1, Annexin V,

APC -conjugated anti-human IL-6, Control-IgG1, Annexin V,

recombinant human cytokines rh IL-1alpha, rh IL-6, rh Neuregulin, rh TNFα,

human IL-6 ELISA-set, human TNF alpha ELISA set

recombinant mouse cytokines rm IL-1alpha, rm IL-1beta, rm IL-6, rm TNFα,

recombinant rat cytokines rr IL-1alpha, rr IL-1beta, rr IL-6, rr TNFα,

DETAILS