



Seminarios internos del IBFG

Unravel the molecular mechanisms involved in ischemic tolerance and induced by preconditioning

Rebeca Vecino

Jueves 31 de Enero de 2019

Brain ischemic tolerance (IT) is a well-known phenomenon in which brief non-injurious preconditioning stimulus (preconditioning, PC) confer robust neuroprotection against a subsequent severe ischemic damage. Although the molecular mechanisms underlying IT are not yet fully clarified, the attenuation of the apoptotic cell death may be involved.

Tumor suppressor protein p53 accumulates in the ischemic areas of the brain and contributes to neuronal apoptotic cell death. Our recent results show that early PC prior ischemia provided neuroprotection by increasing E3-ubiquitin ligase MDM2 levels, which promotes its interaction with p53 and triggers p53 nuclear and cytosolic destabilization after ischemia [1].

Moreover, it is known that the cyclin-dependent kinase-5 (Cdk5) contributes to stabilization and activation of p53 promoting ischemia-associated neuronal apoptotic death [2-4]. Here, we aimed to study the possible role of p53, as well as its related proteins, in PC-promoted ischemic tolerance.

To do that primary cortical neurons were exposed to a validated *in vitro* pharmacological model of PC based on exposure of neurons with moderate subtoxic dose of NMDA (20 μ M) for 2 hours prior oxygen glucose deprivation (OGD) (preconditioning condition, NMDA-PC+OGD). In parallel, neurons were incubated in Normoxia (Nx) or preconditioning (NMDA-PC).

Our results demonstrate that NMDA-PC prevented the phosphorylation and the subsequent stabilization of p53 induced by OGD. By analyzing proteins involved in post-translational modifications of p53, we observe that PC decreased Cdk5 levels, both genetic and protein expression, just as the Cdk5/p25 complex formation and its kinase activity after ischemic damage. Thereby, pharmacological inhibition of Cdk5 activity with Roscovitine and Cdk5 knockdown with siRNA conferred neuroprotection against the ischemic insult by decreasing phosphorylated and active state of p53.

These findings demonstrate the key role of the MDM2-p53 pathway in neuroprotection induced by preconditioning against a subsequent ischemic insult and suggest that Cdk5 may be involved in PC-associated tolerance.

[1] Rebeca Vecino, María C. Burguete, Teresa Jover-Mengual, Jesús Agulla, Verónica Bobo-Jiménez, Juan B. Salom, Ángeles Almeida and María Delgado-Esteban. *The MDM2-p53 pathway is involved in preconditioning-induced neuronal tolerance to ischemia*. Sci Rep. 2018; 8: 1610. [2] Miguel Veas-Pérez de Tudela, Carolina Maestre, María Delgado-Esteban, Juan P. Bolaños and Ángeles Almeida. *Cdk5-mediated inhibition of APC/C-Cdh1 switches on the cyclin D1-Cdk4-pRb pathway causing aberrant S-phase entry of postmitotic neurons*. Scientific Reports 5: 18180 (2015) [3] Daniel Jimenez-Blasco, Patricia Santofimia-Castaño, Antonio Gonzalez, Ángeles Almeida and Juan P. Bolaños. *Astrocyte NMDA receptors' activity sustains neuronal survival through a Cdk5-Nrf2 pathway*. Cell Death and Differentiation 22(11): 1877-1889 (2015) [4] Rebeca Lapresa, Jesús Agulla, Irene Sánchez-Morán, Rubén Zamarreño, Estefanía Prieto, Juan P. Bolaños and Ángeles Almeida. *Amyloid- β promotes neurotoxicity by Cdk5-induced p53 stabilization*. Neuropharmacology. 2019 Mar 1;146:19-27