ROS-induced Sp1 upregulation promotes WRAP53 accumulation leading to neuroprotection after ischemia

S. Martínez-Peralta[I], I. Sánchez-Morán[II], C. Rodríguez[I], A. Almeida[I]

[I]Institute of Biomedical Research of Salamanca, University Hospital of Salamanca, University of Salamanca, CSIC, Salamanca, Spain, [II]Institute of Functional Biology and Genomics (IBFG), CSIC, University of Salamanca, Salamanca, Spain

Ischemia-induced oxidative stress compromises genome integrity, resulting in DNA damage and neuronal loss after stroke. Reactive oxygen species (ROS) generated after ischemia also activates molecular pathways to repair the damage and promote neuroprotection. We recently described that ischemia-induced ROS are necessary to accumulate WRAP53 (WD40 encoding RNA antisense to p53) in the nucleus, where it promotes DNA repair after stroke. However, the underlying mechanism remains unknown. Transcription factor Sp1 acts as a pleiotropic oxidative stress response protein in neurons. Interestingly, Wrap53 promoter contains putative consensus sequences (GC boxes) for Sp1. Hence, Sp1 might be a good candidate to modulate WRAP53-induced neuroprotection after ischemia.

Ischemia rapidly induced Sp1 expression in primary neurons subjected to a well-established in vitro model of ischemia (oxygen and glucose deprivation). WB and RT-PCR were performed to quantify protein and mRNA levels, showing that Sp1 induction preceded WRAP53 upregulation. CHIP analyses demonstrated that Wrap53 co-immunoprecipitated with anti-Sp1, proposing Sp1 as a modulator of WRAP53 upregulation after ischemia. Protein location was established by nuclear fractionation and confocal microscopy, revealing an increased nuclear/cytosolic ratio of WRAP53 in the presence of high levels of ROS. Moreover, Sp1 downregulation by siRNA prevented WRAP53 accumulation after ischemia.

In conclusion, Sp1 has been shown to modulate ROS-induced WRAP53 accumulation leading to neuroprotection after ischemia. This new ROS-Sp1-WRAP53 signaling pathway points towards novel therapeutic targets for stroke and identifies therapeutic windows based on the relevance of ROS as signaling molecules after ischemia.

Funding by ISCIII (PI21/00727, RD21/0006/0005 co-funded by the European Union); FEDER; JCYL (CSI151P20; Escalera de Excelencia CLU-2017-03 Cofinanciado por P.O.FEDER de Castilla y León 14-20)