Modulation of the Unfolded Protein Response and Antioxidant Response by S-glutathionylation

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Parkinson's disease (PD) is a neurodegenerative movement disorder characterized by the loss of dopaminergic neurons. Although the molecular mechanisms underlying dopaminergic neuronal death are still not completely understood, strong evidence has accumulated implicating mitochondrial dysfunction, oxidative stress and proteolytic pathways failure in the pathogenesis of the disease. These processes are associated with the generation of reactive oxygen species (ROS) and might lead to the accumulation of misfolded oxidized proteins causing endoplasmic reticulum (ER) stress. To restore the redox status cells are endowed with an antioxidant system coordinated by the transcription factor nuclear factor erythroid 2 related factor 2 (Nrf2). Glutathione S-Transferase pi (GSTP), a downstream target of Nrf2, protects cells from ROS by several mechanisms including the modulation of protein S-glutathionylation, a reversible post-translational modification that can protect proteins against irreversible oxidation. Homeostasis reestablishment upon ER stress is dependent on the unfolded protein response (UPR). The protein kinase R-like ER kinase (PERK), one of the main effectors of the UPR, is activated upon ER stress and triggers the inhibition of protein synthesis preventing an additional overload of the ER. In parallel, PERK phosphorylates Nrf2 initiating the antioxidant response. Herein we evaluated the expression levels of ER stress markers in C57BL/6 wild-type versus GSTP knockout mice upon treatment with the neurotoxin MPTP that mimic some of the pathological features of PD, with or without concomitant treatment with TUDCA, a chemical chaperone that is able to enhance ER adaptive capacity. We further evaluated the involvement of GSTP in the S-glutathionylation of antioxidant proteins and ER-stress related markers in vivo. Our results show that MPTP-induced oxidative stress leads to glutathione depletion, increased GSTP expression and altered glutathione-dependent reactions including S-glutathionylation. Moreover GSTP knockout mice display a different pattern of S-glutathionylation when compared to their wild-type counterparts. Expression levels of UPR effectors, namely PERK and ATF-6, are also altered in GSTP knockout mice. Furthermore, GSTP potentiates the S-glutathionylation of Kelch ECH associating protein 1 (Keap1), the endogenous negative regulator of Nrf2, in mice brain following MPTP administration with subsequent Nrf2 pathway activation and increased expression of GSTP, in a positive feedback regulatory loop. Overall our results demonstrate the involvement of ER stress in a mouse model of PD and provide new insights into the role of GSTP in the S-glutathionylation of antioxidant and ER-stress responsive proteins, unravelling a new mechanism contributing to GSTP-elicited neuronal protection. Keywords: Endoplasmic Reticulum Stress; S-glutathionylation; Antioxidant Response

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