

Prednisolone restores blood brain barrier damages in dystrophic MDX mouse

Roberto Tamma^a, Tiziana Annese^a, Vincenzo Benagiano^a, Vito Longo^b, Simona Ruggeri^b, Enrico Crivellato^c, Domenico Ribatti^a, Annamaria De Luca^d, Beatrice Nico^a

^a Department of Human Anatomy and Histology

^b Department of Internal Medicine and Clinical Oncology, University of Bari Medical School, Bari, Italy ;

^c Department of Medical and Morphological Research, Anatomy Section, University of Udine Medical School, Italy;

^d Department of Pharmaco-biology, Faculty of Pharmacy ,University of Bari.

Although the glucocorticoids delay the progression of Duchenne muscular dystrophy (DMD) their mechanism of action is unknown. In our previous studies we demonstrated that in the mdx mice, an animal model of DMD, besides the muscle degeneration, serious damages of the blood-brain barrier (BBB) occur taking to enhanced vessels permeability and brain edema (1). Moreover, we observed that the mdx mice after α -methyl-prednisolone (PDN) treatment ameliorated the histopathological profiles and the excitation-contraction of the myofibers (2).

In this study, we evaluated the effects of the PDN on the BBB of the mdx mice, by estimating the immunocytochemical and biochemical expression of endothelial ZO-1 and occludin, pericyte desmin, and glial GFAP and short dystrophin isoform Dp 71 proteins, used as BBB markers. In addition, we analyzed the expression of dystrophin associate proteins (DAPs) aquaporin-4 (AQP4) and α - β dystroglycan in parallel in both brain and muscles of PDN treated mdx as well as in control mice. Results showed in mdx PDN treated mice a significant increase of the mRNA and protein content of all the glial, pericyte and endothelial proteins as compared to untreated mdx. Moreover, by immunoprecipitation we demonstrated that the BBB alteration in the mdx mice were coupled with enhanced occludin and AQP4 phosphorylation degree which, instead, was reduced after PDN treatment. Finally we observed that AQP4 and α - β dystroglycan complex increases its mRNA and protein content in both PDN mdx brain and muscle fibers, compared with mdx mice where the perivascular glial membranes and the myofibers showed a light staining after immunofluorescence analysis . These data indicate that the PDN restores the BBB damages in the mdx mice by inducing in the glial cell the expression of GFAP, AQP4 and Dp71 proteins and in the pericytes and endothelial cells, of the desmin and ZO-1 proteins, which are deficient in the dystrophic mice. Moreover, the reduction in the AQP4 and occludin phosphorylation degree coupled with their anchoring to glial and endothelial membranes in the PDN mdx mice suggests that the glial and endothelial cells may be a cellular target of the drug. Finally, the enhanced expression of DAPs AQP4 and α - β dystroglycan in both brain and myofibers of PDN treated mdx mice compared to untreated mdx ones suggest the PDN might ameliorate the brain vessels and muscles functions of the dystrophic mice by a restoring a correct links between DAPs proteins and the extracellular matrix.

1. Nico B et al. *Glia*, 42: 235-251. (2003).

2. Cozzoli A. et al., *Neuropathol. Appl. Neurobiol.* 37, 243-256 (2011).

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