PREOPERATIVE PHARMACOLOGICAL CONDITIONING INFLUENCES CLINICAL AND MOLECULAR OUTCOME OF ISCHEMIA/REPERFUSION INJURY OF THE SPINAL CORD AFTER THORACOAORTIC CROSCLAMPING IN MICE.

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Patients, undergoing clamping procedure for thoracoabdominal aortic repair, may suffer postoperatively from the consequences of ischemia/reperfusion (I/R) injury, what can cause paraplegia. Studies show a central role of the endoplasmic reticulum (ER) stress as a result from the I/R and the resulting free radicals. Erythropoietin (Epo) and its carbamylated derivate (cEpo-Fc) are able to act antioxidative and antiapoptotic. It was shown that Epo and cEpo-Fc can improve the neurological outcome of animals after spinal cord injury. In this study, we examine the effects of pre-operative use of native erythropoietin (EPO) and its derivative cEPO-Fc on paraplegia and ER stress in vivo.

2) Method
An ischemia of the spinal cord was induced in male mice (C57BL/6J) by clamping the thoracic aorta and the left subclavian artery.

Three studygroups (Epo, cEpo-Fc and control) were observed for different periods of time (6h, 24h, 96h).

The clinical, neurological outcome of the mice was evaluated by using the Basso-Mouse-Scale (BMS).

The spinal cord was stained in Hematoxylin-Eosin (HE) and Luxol-Fast Blue (LFB). Immunohistochemical stainings for ER stress relevant proteins (GRP78 and Caspase 12) were performed as well.

Clamping-time: 7 minutes

5) Results of the Histopathology

5.1) Hematoxylin-Eosin staining (HE)
Significantly decreased necrosis in the Epo and cEpo-Fc groups in comparison to the control group.

5.2) Luxol-Fast-Blue staining (LFB)
Significantly higher amount of motoneurons in the Epo and cEpo-Fc groups in comparison to the control group.

5.3) Immunohistochemistry GRP78
Increased expression of GRP78 in the Epo and cEpo-Fc groups compared to the control group.

5.4) Immunohistochemistry Caspase 12
Decreased expression of Caspase12 in the Epo and cEpo-Fc groups compared to the control group.

The results show a significant positive effect of Epo and cEpo-Fc on the clinical neurological and histological outcome of the mice. The increased expression of Caspase12 (control) makes an increased apoptotic rate likely. The increased expression of GRP78 (Epo, cEpo-Fc) makes an increased formation of the UPR likely.

Native Epo and cEpo-Fc can significantly improve the clinical outcome of mice (C57BL/6J). Significant effects are also shown in histological stainings. The molecular mechanisms undlerlying these effects are being further evaluated.

6) Conclusion

The results show a significant positive effect of Epo and cEpo-Fc on the clinical neurological and histological outcome of the mice. The increased expression of Caspase12 (control) makes an increased apoptotic rate likely. The increased expression of GRP78 (Epo, cEpo-Fc) makes an increased formation of the UPR likely.

Native Epo and cEpo-Fc can significantly improve the clinical outcome of mice (C57BL/6J). Significant effects are also shown in histological stainings. The molecular mechanisms undlerlying these effects are being further evaluated.