Neurosteroids as neuroprotective, pro-myelinating and anti-inflammatory agents in Multiple Sclerosis (NBB project 793)

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Research question and background

Multiple Sclerosis (MS) occurs at least twice as often in women than in men. The clinical course of MS in women is, however, more benign. Pathologically, women show more inflammatory but less destructive lesions than men, in which atrophy and axonal damage are more evident. These differences strongly suggest an influence of gender steroid hormones on MS. We recently obtained the first evidence that the gender steroid hormones biosynthetic pathways and hormone receptors are indeed changed in MS tissue, and that these changes are partly gender and MS course-specific (NBB Project 650). In this project we propose to measure the actual levels of these hormones in MS lesions and normal appearing white matter (NAWM) from female and male patients using highly sensitive gas chromatography-mass spectrometry (GC/MS). We will also investigate functional effects of different steroids in post-mortem human primary glial cells from female and male MS and control brain donors.

Methods and tissues used

Human postmortem frozen and fresh tissue was provided by th NBB. Chronic active, chronic inactive lesions from female and male MS patients and sex/age subcortical white matter controls (n=10/group) isolated from the frozen tissue blocks as previously described (Luchetti et al 2014) as well as remyelinating areas from 7 females and 7 males along with matched control white matter. Samples were sent to the laboratory of Dr. Michael Schumacher/Dr. Philippe Liere in Paris where they will perform GC/MS quantification of steroids. Primary human microglia culture was isolated from fresh adult human post-mortem subcortical white matter (n=5 MS/CTR) as previously described (Luchetti et al 2014). Cells were treated with steroids (e.g. progesterone, dihydrotestosterone and estradiol) for 48hr and gene expression of neuroprotective molecules (e.g BDNF, GLAST, GS) and inflammatory markers (e.g. IL1b, IL-10, TNF) were analyzed by qPCR.

Results and conclusion

GC/MS analysis in remyelinating areas from females and males MS patients and white matter from matched control show a novel sex-dependent steroidogenic dysregulation. Specifically, estrone is increased in female MS patients, while in males 20a-dihydroprogesterone, dehydroepiandrosterone sulphate and testosterone levels are reduced. Effects of these steroids on myelination related molecules (e.g. MBP, PLP, MAG, MOG) are being tested in vitro in a human oligodendrocyte cell line treated with or without IL-1ß and TNF pro-inflammatory stimulus. Preliminary data suggest that testosterone increases gene expression of MBP when the cells are treated with IL-1ß and TNF, suggesting a promyelinating effect under inflammatory conditions.
In human primary microglia isolated from subcortical normal appearing white matter of 5 MS female patients, 2 non demented controls and 3 with other neurological diseases, P

treatment increases expression of BDNF (1,5 fold) in MS patients only. Interestingly in microglia isolated from a lesion of an MS patient, all steroid treatments increased BDNF (2-8 fold). Overall these data suggest that steroids may have some neuroprotective effects and modulate inflammation via regulation of gene expression in glial cells.

References:

Luchetti S, van Eden CG, Schuurman K, van Strien ME, Swaab DF, Huitinga I.Gender differences in multiple sclerosis: induction of estrogen signaling in male and progesterone signaling in female lesions. *J Neuropathol Exp Neurol. 2014 Feb;73(2):123-35.*