Intrathecal methotrexate reduces astrogliosis in a murine model of experimental demyelination

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Objective:

We hypothesize that the benefits of intrathecal methotrexate (ITMTX) on progressive MS is only partially related to its anti-inflammatory properties. Consequently, we investigated its influence on astrogliosis in a non-inflammatory CNS demyelination model.

Background:

MS patients with a progressive disease respond poorly to currently available anti-inflammatory disease modifying agents. We previously reported that the intrathecal administration of the antifolate drug methotrexate has a beneficial impact on the disease progression of severe primary and secondary progressive MS cases. Astrocytic activation and subsequent astrogliosis occur in progressive MS as well as in the cuprizone-induced model of corpus callosum demyelination and are considered to be hallmarks of scar formation in the CNS.

Methods:

To induce corpus callosum demyelination, male C57Bl/6 mice were fed with 0.2% cuprizone mixed into ground chow. Methotrexate was administered intracerebroventricularly by osmotic pumps (concentration 0.5µg/µL (diluted in PBS), flow rate 0.11µL/h). Brain tissue sections were stained for astrocytic markers and by DAPI. Three random sections were analysed per animal by a blinded investigator.

Results:

After four weeks of cuprizone feeding control mice were characterized by a strong increase in GFAP+ astrocytes within the corpus callosum as compared to naive mice. The number of GFAP+ astrocytes was significantly reduced by 40% in the animals which received methotrexate for the same period (p-value = 0.0125).

Discussion:

The pathophysiological basis of the cuprizone-induced demyelination model are not primarily related to inflammtory mechanisms. Similarly, the progressive forms of MS are less dependent on inflammatory events than its relapsing forms. We were able to establish an inhibition of astrogliosis by ITMTX in the corpus callosum of cuprizone fed mice corroborating that beneficial impact of ITMTX on disease progression is not solely mediated by its anti-inflammatory properties.