PhD thesis

The role of complement in Experimental Autoimmune Encephalomyelitis, the mouse modell of Multiple Sclerosis

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Introduction

Multiple sclerosis (MS) is the most common inflammatory autoimmune disease of the central nervous system (CNS). Immune response against the oligodendrocytes destroys their product, the myelin sheath. These lesions and the following axonal damage cause very diverse sensual and vegetative symptoms, depending on the site of the immune attack. Experimental autoimmune encephalomyelitis (EAE) is a widely used animal model of MS. It is believed that Th1 type regulatory T cells induce the inflammatory process and activate the effector cells, such as cytotoxic T cells, macrophages, resident astrocytes and microglia cells. More recently the possible role of Th17 cells was also described, but still remain some open questions regarding the pathogenesis of the disease. The complement system is known to play an important role not only in the effector phase, but also in the initiation and regulation of adaptive responses. The role of complement proteins in the pathogenesis of MS and EAE has been investigated for a long time; the results however are still contradictory.

So far no investigations of EAE have been carried out using cobra venom factor-treated mice. Cobra venom factor (CVF) is widely used for transient decomplementation of mice

and rats. The role of autoantibodies in MS is still controversial; yet one of the most important diagnostic hallmarks is the presence of oligoclonal Igs and plasma cells in cerebrospinal fluid.

In the present study our aim has been to reveal how lack of complement at the initiation of EAE influences the onset and course of the disease. As it is known for long that complement is involved in both T-dependent and T-independent antibody responses, we compared the amount of myelin oligodendrocyte glycoprotein (MOG)-specific antibodies in normal and CVF-treated mice, after induction of EAE. In addition to the levels of these autoantibodies, their complement activating ability was also assessed, using an antibody-complement (AbC) array system, developed recently at our department. Finally, the proliferative capacity of MOG-specific lymph node T-cells derived from normal and decomplemented animals has also been compared.

Our aims were to reveal

I. How lack of complement at the time of induction influences the pathogenesis of the acute, monophasic type of EAE.

II. How lack of complement at the time of induction influences the pathogenesis of the relapsing-remitting type of EAE.

III. How EAE develops in C3 KO animals

IV. How the amount of MOG-specific antibodies changes in complement-depleted animals and how T cells derived from diseased animals can be activated with the antigen, ex vivo. In addition, we aimed to determine whether the complement activating ability of MOG-specific antibodies is different in normal and C3-depleted animals.

V. The histological changes inside the CNS white matter using immuno-staining to identify different cell types and complement-products, involved in the pathogenesis of EAE.

Methods

- EAE mouse model
- Histology, immunohistology
- Sandwich ELISA
- Protein chip technology
- *In vitro* antigen presentation
- Tissue culture

Results

Our results show that in C57BL/6 animals with transiently depleted complement at the onset of the disease, development of EAE is significantly delayed, and severity is lower, compared to animals with normal complement activity. EAE induced in C3 KO animals showed earlier symptomes, but later less severe disease, than wild type mice. CVF administration was found to inhibit the relapses of EAE in SJL/J mice, and reduced mortality. We investigated the in vitro response of antigen-specific T cells isolated from the lymph nodes of MOG-immunized animals at the onset of the symptomes. Our results show that the proliferative capacity of antigen-specific T cells derived from CVF treated animals is significantly lower than in the control group. With a protein microarray system developed in our department we have detected lower MOG-specific antibody levels in the sera of CVF-treated mice. Moreover, we found lower numbers of $CD4^+$ T cells in the central nervous system of CVF-treated mice.

Our data prove, that complement has a modulatory effect in the pathogenesis of EAE. We have shown that lack of complement at the time of induction delays the onset of the disease and modulates the activity of MOG-specific T cells, reduces the level of MOG-specific antibodies and the number of infiltrating $CD4^+$ T cells in the central nervous system.

Publications

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