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## Glucocorticoids in the control of neuroinflammation

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**Abstract**

Glucocorticoids are a class of steroid hormones that are endowed with profound anti-inflammatory and immunosuppressive activities. Endogenous glucocorticoids are key players in the modulation of the immune system and establish an endocrine basis of many inflammatory diseases. In addition, synthetic glucocorticoids are amongst the most commonly prescribed drugs worldwide for the treatment of autoimmune disorders. In this review we summarize our present knowledge on the mechanisms by which glucocorticoids impact on multiple sclerosis (MS), a highly prevalent neuroinflammatory disease, and its animal model experimental autoimmune encephalomyelitis (EAE). In spite of the new methodologies that have become available during recent years, we are still far from a comprehensive picture of the mechanism by which glucocorticoids control neuroinflammation.

*Keywords:* Glucocorticoids; Multiple Sclerosis; Experimental Autoimmune Encephalomyelitis; Apoptosis; Blood-brain barrier

## 1. Introduction

Glucocorticoids (GCs) are a class of steroid hormones that are commonly used to treat acute and chronic inflammatory disorders (Tuckermann et al., 2005). They exert most of their functions by binding to the glucocorticoid receptor (GR), which controls gene expression and signalling cascades through genomic as well as non-genomic mechanisms. By this means, GCs modulate the survival, differentiation, migration and various effector functions of leukocytes and other cell types and thereby impact both, innate and adaptive immunity. Whilst GCs administered at pharmacological concentrations are considered purely immunosuppressive, fluctuations in the levels of endogenous GCs, e.g. during stress, result in more complex immunomodulatory effects (Elenkov and Chrousos, 1999).

One major application of GCs is the treatment of neuroinflammatory disorders, in particular multiple sclerosis (MS), the most prevalent chronic autoimmune disease of the central nervous system (CNS) in the Western world (Noseworthy et al., 2000). MS is characterized by infiltrating autoreactive T-cells in the CNS that initiate an inflammatory cascade involving leukocytes and humoral components (Sospedra and Martin, 2005). This causes demyelination and axonal loss, which eventually leads to severe functional deficits. While GCs have no positive effect on the long-term prognosis of MS, optic neuritis and other acute relapses are widely treated with high doses of methylprednisolone (Milligan et al., 1987). Still, such a therapy may lead to severe complications or incomplete recovery. Furthermore, it is known that the course of MS is influenced by the level of endogenous GCs (Elenkov and Chrousos, 1999). Women in the third trimester of pregnancy and Cushing syndrome patients often experience remission of the disease. This is explained by a polarization of the immune system towards  $T_H2$  dominated responses as a consequence of increased

cortisol synthesis. Thus, fluctuations in GC production impact the course of neuroinflammatory diseases such as MS.

Whilst the importance of GCs for the control of neuroinflammation is doubtless, the mechanism at work is still unclear. Presently the pro-apoptotic function of GCs, their ability to impact migration and extravasation through the blood-brain barrier (BBB) and various influences on the effector functions of immune cells are discussed (Reichardt et al., 2006). While clinical studies have made important contributions to solve these questions, new insight has mainly been achieved using experimental autoimmune encephalomyelitis (EAE), a widely recognized animal model for MS that reflects many hallmarks of the human disease (Gold et al., 2006). It will be important in the future to continue along this line to improve our understanding of the mechanism of GCs in the control of neuroinflammation.

## **2. Molecular basis of glucocorticoid actions**

The GR, a member of the nuclear receptor superfamily, is involved in the control of a plethora of physiological processes ranging from energy homeostasis and cognitive functions to the modulation of the immune system (Mangelsdorf et al., 1995). In the absence of GCs, the GR is sequestered in a multimeric complex in the cytoplasm. Once GCs have entered the cell, the GR is released and translocates into the nucleus (Beato et al., 1995). After binding to DNA, it drives transcription from so-called GC response elements (GREs), present in the promoter and enhancer regions of many genes (Luisi et al., 1991). Besides this mode of action, the GR also regulates gene expression by interfering with other transcription factors such as NF- $\kappa$ B, AP-1 or Stat5 without the need to directly bind DNA (Heck et al., 1997; Heck et

al., 1994; Stoecklin et al., 1997). Besides their established effects on transcription there is growing evidence for additional non-genomic effects of GCs (Song and Buttgereit, 2006). These comprise physicochemical interactions of the hormones with cellular membranes, the interference of the GR with cytosolic signal transduction components such as PI3K (Limbourg et al., 2002) or actions mediated through membrane-bound GRs (Bartholome et al., 2004). While cellular properties are modulated through different mechanisms, the GR itself is also subject to various modifications. These include the generation of multiple N-terminal isoforms by translational mechanisms (Lu and Cidlowski, 2005), differential phosphorylation of the receptor (Webster et al., 1997) and the use of distinct promoters by alternative mRNA splicing (Purton et al., 2004). Taken together, this forms the basis for a plethora of GC activities.

### **3. Multiple sclerosis and its animal model experimental autoimmune encephalomyelitis**

MS was first described in 1868 by the French neurologist Jean Martin Charcot who observed an accumulation of inflammatory cells in the CNS of patients suffering from periodic episodes of neurological dysfunction (Hafler, 2004). In 1933, Thomas Rivers showed that symptoms similar to the ones seen in MS patients could be induced by repeated injection of rabbit brain homogenate into primates without a need of infectious agents (Rivers et al., 1933). This marks the beginning of the long history of EAE, the most widely used animal model of MS.

MS is a neuroinflammatory disorder characterized by the infiltration of pathogenic T cells into the CNS reacting with myelin antigens (Figure 1). Although the site where the encephalitogenic cells are initially primed remains elusive, it is known that they cross the BBB followed by reactivation (Kawakami et al., 2005). In turn, effector cells release proinflammatory cytokines leading to the activation of the microglia and other local immune cells. Subsequently, additional autoreactive and conventional T cells, macrophages, granulocytes and B cells are attracted to the CNS accompanied by antibody production and complement deposition (Hemmer et al., 2006). This leads to the destruction of the myelin sheath, oligodendrocyte damage and axonal loss (Bjartmar and Trapp, 2001). For a long time MS and EAE were considered to be purely  $T_H1$ -mediated autoimmune disorders. However, the  $T_H17$  lineage (Komiyama et al., 2006) and  $CD8^+$  T cells (Frieze and Fugger, 2005) have recently gained attention and also appear to play an important role in the pathogenesis of neuroinflammatory diseases.

The majority of MS patients suffer from a relapsing-remitting form of the disease, which is characterized by attacks of neurological dysfunction followed by a gradual improvement. After ten years or more most of these patients develop secondary progressive MS where phases of quiescence are no longer observed. In contrast, up to 15% of all patients present with primary progressive MS that manifests in a continuous aggravation of the disease without remission. Whilst no treatment is available for the latter form, relapsing-remitting MS is most commonly treated with Interferon- $\beta$  and Glatiramer Acetate (Murray, 2006). In addition, new reagents such as Tysabri or FTY720 have proven to efficiently ameliorate MS in clinical trials (Fontoura et al., 2006). Whereas application of GCs has little impact on the long-term prognosis of MS, high-dose therapy still remains the standard regimen for the treatment of acute relapses and optic neuritis. Most commonly 0.5 - 2g



methylprednisolone are applied intravenously for five consecutive days followed by a rapid taper (Pozzilli et al., 2004). However, despite its unsurpassed efficacy, accompanying side effects still cannot be circumvented.

EAE has been instrumental in analysing the pathogenesis of MS for more than 70 years (Gold et al., 2006). EAE can be induced by injection of various myelin antigens in the presence of adjuvant (active EAE) or by adoptive transfer of encephalitogenic T cells (AT- EAE) (Ben-Nun et al., 1981; Swanborg, 2001). MBP-induced EAE in the Lewis rat results in a monophasic disease course, which mimics the inflammatory characteristics of MS whereas injection of myelin oligodendrocyte glycoprotein (MOG) into C57/Bl6 mice causes a chronic-progressive disease that is accompanied by demyelination and axonal damage (Reichardt et al., 2006). Moreover, DA rats or SJL mice are used to investigate a relapsing-remitting disease course, which is seen in the majority of MS patients (see above). The variety of EAE animal models highlights the fact that most characteristics of MS can be studied in rodents although none of them fully reflects the human pathology.

#### **4. Pivotal mechanisms of glucocorticoids in MS and EAE**

##### **4.1. Induction of apoptosis by glucocorticoids**

The phenomenon of GC-induced T cell apoptosis has been recognized almost 30 years ago but the exact mechanism is not yet fully understood (Screpanti et al., 1989). The presence of the GR, DNA-binding dependent transcription and *de novo* gene expression is required but none of the identified target genes such as the pro-apoptotic proteins Bim and PUMA has turned out to be essential (Erlacher et al.,

2005; Reichardt et al., 1998; Wang et al., 2006). Signal transduction events initiated in T cells after exposure to GCs include the activation of various caspases, the release of cathepsin B from lysosomes, increased  $H_2O_2$  levels, production of ceramides, altered  $Na^+ / K^+$  levels, interaction with kinases and many more (Herold et al., 2006).

Induction of T cell death in the CNS is widely observed in EAE models and MS patients (Figure 2). In the remission phase of MBP-EAE in Lewis rats, autoreactive T lymphocytes in the spinal cord undergo massive apoptosis (Tabi et al., 1994). Presumably, endogenous GCs are released as a consequence of proinflammatory cytokines produced during EAE and lead to the induction of T cell death (Pender et al., 1992). This notion is supported by the finding that adrenalectomized rats suffer from a fatal EAE accompanied by very low levels of apoptosis (Grauer et al., 2001; Smith et al., 1996). Furthermore, high-dose methylprednisolone pulse therapy of AT-EAE in Lewis rats augments T cell apoptosis in a dose-dependent manner (Schmidt et al., 2000). Similar results were obtained in clinical studies, demonstrating induction of apoptosis in peripheral blood leukocytes after high-dose GC therapy of MS patients (Leussink et al., 2001). In general, it appears that DNA fragmentation and the susceptibility to apoptosis after GC treatment is more pronounced in  $CD4^+$  T cells as compared to  $CD8^+$  T cells due to differences in Bcl-2 expression (Migita et al., 1997). Thus, both endogenous and therapeutically applied GCs cause T cell death during EAE and MS and are responsible for the amelioration of the disease. Although this is generally accepted, the true relevance for therapeutic efficacy in the treatment of EAE and MS is debatable. In particular, a marked repression of neurological symptoms could be achieved in Lewis rats after a single dose of 0,25mg/kg dexamethasone despite the absence of T cell apoptosis (Nguyen et al., 1997). Moreover, it is also not known whether autoreactive and conventional T cells are

differentially affected by GC-induced cell death (McCombe et al., 1996). Thus, administration of GCs leads to the induction of T cell apoptosis and ameliorates MS and EAE but the mechanism and the extent to which a causative relationship between both phenomena exists remains elusive.

#### **4.2. Effects of glucocorticoids on the blood-brain barrier and leukocyte migration**

The BBB mainly consists of endothelial cells, astrocyte end feet and pericytes and serves to protect the CNS from attacks by the systemic immune system. The endothelium shows specific characteristics in the sense that these cells lack fenestration, make more extensive tight junctions and contain a specific transport system (Ballabh et al., 2004). While the vascular endothelium does not express ICAM-1 and VCAM-1 under basal conditions, these adhesion molecules are upregulated in response to pro-inflammatory cytokines such as IFN- $\gamma$  and TNF- $\alpha$  (Sloka and Stefanelli, 2005). Consequently, integrines on encephalitogenic T cells interact with these adhesion molecules, which is required for the extravasation of additional effector T cells into the CNS (Engelhardt, 2006). Importantly, GCs inhibit the disruption of the BBB and thereby reduce leukocyte infiltration into the CNS (Paul and Bolton, 1995). Direct effects of GCs on the BBB were demonstrated *in vitro* (Figure 2). In cultured brain endothelial cells dexamethasone induced the reduction of pores, the concentration of F-Actin on the cell periphery and an increased expression of ZO-1 and occludin (Förster et al., 2005; Förster et al., 2006; Romero et al., 2003). How these mechanisms impact the integrity of the BBB *in vivo* requires

further investigation. Another possibility is the reduction of integrine levels on T cells as well as adhesion molecules on the endothelial cells (Figure 2). Following methylprednisolone treatment, one study described reduced amounts of soluble VCAM-1 and E-selectin in the serum of MS patients (Elovaara et al., 2000). Similarly, the amount of VLA-4 and LFA-1 on blood leukocytes was found to be decreased (Elovaara et al., 1998). High-dose methylprednisolone also reduced the TNF- $\alpha$  induced expression of ICAM-1 and VCAM-1 (Gelati et al., 2000). Studies in humans further demonstrated a down-regulation of gelatinase B and urokinase-type plasminogen activator and an increase of TIMPs after steroid therapy (Rosenberg et al., 1996). Also the cytokine-induced up-regulation of MMP-9 (Harkness et al., 2000) as well as the histamine H<sub>1</sub> and H<sub>2</sub> receptors (Karlstedt et al., 1999) in rat brain endothelial cells were diminished after dexamethasone treatment. Recently, it was shown that fibrinogen is deposited in MS plaques following BBB disruption. This leads to microglia activation through MAC-1 and thereby contributes to demyelination and an exaggerated EAE (Adams et al., 2007). Since GCs prevent disruption of the BBB, this should diminish fibrinogen leakage into the CNS and therefore contribute to the amelioration of the disease. In summary, these studies revealed cell-cell-interaction as well as extracellular matrix remodelling as potential targets of GCs in the context of the BBB (Figure 2).

Interpretation of GC effects on the BBB *in vivo* is somewhat problematic. In most studies encephalitogenic T cells cannot be distinguished from other T cells, which may disguise specific effects on the effector cells. In contrast, *in vitro* studies using cell lines do not allow addressing the complex interplay between infiltrating leukocytes and the endothelial cells of the BBB. Finally, the down-regulation of adhesion molecules on the vascular endothelium may either result from direct transcriptional regulation but could also be secondary to diminished cytokine release

by T effector cells in the CNS. Therefore, it is unclear to which extent GCs indeed directly modulate the function of the BBB or whether indirect effects leading to a diminished release of proinflammatory cytokines are dominating.

Chemokines and their receptors play a prominent role for the migration of leukocytes to the site of inflammation (Figure 2). In particular CXCR-3 seems to be crucial for neuroinflammation. The majority of T cells in MS patients express CXCR-3 within the CSF compartment (Sorensen et al., 2002) and almost all encephalitogenic T cells isolated from Lewis rats stain positive for this chemokine receptor (Tischner et al., 2006). High-dose methylprednisolone treatment of MS patients leads to a reduction of CXCR-3 and CCR5 expression on peripheral blood T cells (Elovaara et al., 2006; Wang et al., 2003). In addition, a diminished level of CXCL10/IP-10, the ligand of CXCR3, and an elevated level of CCL2 were observed in the CSF of MS patients after GC therapy (Moreira et al., 2006). It is noteworthy that the role of CCL2 in MS is controversial, although it appears that it is linked to the polarization of T cells towards a T<sub>H</sub>2 phenotype (Moreira et al., 2006). Thus, influences on the expression of chemokine receptors and their ligands may also result from a T<sub>H</sub>2 shift induced by GCs (see below). Finally, production of IL-8 by monocytes, which contributes to the disruption of the BBB by recruiting leukocytes to the CNS is also decreased in MS patients after GC treatment (Mirowska-Guzel et al., 2006).

In summary, administration of GCs interferes with the disruption of the BBB. This involves direct effects on endothelial cells, interactions between the vascular endothelium and infiltrating leukocytes, extracellular matrix remodelling and migration along chemokine gradients. However, direct effects on the BBB are hard to distinguish from indirect effects that stem from increased apoptosis or diminished pro-inflammatory cytokine production.

### 4.3. Immunosuppressive effects of glucocorticoids

Besides influences on apoptosis and the BBB, GCs are known for a variety of general immunosuppressive effects (Figure 2). GC treatment decreases the production and release of pro-inflammatory cytokines such as IFN- $\gamma$ , TNF- $\alpha$ , IL-2, IL-1 $\beta$  and IL-12 (Almawi et al., 1996), which is similarly observed in MS patients during steroid therapy. Methylprednisolone decreases the expression of TNF $\alpha$  and IFN $\gamma$  on the mRNA level in PBMCs isolated from MS patients (Gayo et al., 1998). This was also observed in cultured PLP-specific CD4<sup>+</sup> T cells in the presence of dexamethasone (Correale et al., 1998). Most of the pro-inflammatory cytokines that are down-regulated by GCs are T<sub>H</sub>1 cytokines, which are known to play an important role in the pathogenesis of MS. In contrast, the levels of T<sub>H</sub>2 cytokines such as IL-10 and TGF- $\beta$  are rather increased by GCs (Correale et al., 1998; Gayo et al., 1998). This observation led to the model that GCs favour the differentiation of T cells into the T<sub>H</sub>2 lineage and thereby modulate the susceptibility to EAE and MS (Figure 2). Such a notion would be in line with the finding that encephalitogenic T cells cultured in the presence of IL-4 and dexamethasone are unable to induce EAE. Furthermore, we have recently described a strain of transgenic rats (*superTGR*) characterized by enhanced GR signalling (van den Brandt et al., 2007). Intriguingly, these animals show increased resistance to EAE induction, presumably as a consequence of a T<sub>H</sub>2 bias of the immune response. Similarly, clinical studies show that MS patients often experience a remission of disease symptoms under conditions of chronically elevated cortisol levels (Elenkov and Chrousos, 1999). In summary, GCs impact on the

production of various cytokines, polarize the immune response towards the  $T_H2$  type and directly affect the effector functions of encephalitogenic T cells.

Besides the control of cytokine synthesis, also NO production by macrophages, neutrophilic granulocytes and microglial cells is inhibited by GCs (Kleinert et al., 1996) (Figure 2). NO is involved in the destruction of oligodendrocytes by microglial cells, and both IFN- $\gamma$  and TNF- $\alpha$  are able to induce iNOS (Redford et al., 1997; Sloka and Stefanelli, 2005). In a coculture system of rat primary neurons and activated microglia the addition of dexamethasone decreased the production of NO (Golde et al., 2003). However, it is also conceivable that this effect is secondary to the GC-mediated down-regulation of IFN- $\gamma$  and TNF- $\alpha$ , which are essential for the induction of iNOS (Redford et al., 1997; Sloka and Stefanelli, 2005). Another potentially important target for the immunosuppressive activity of GCs is the activation and priming of effector T cells, a process that requires the expression of MHC and costimulatory molecules on myeloid cells (Figure 2). Application of 1mg/kg dexamethasone in a Wistar rat EAE model reduced MHC class II expression on microglial cells, which presumably interferes with antigen presentation and thereby the activation of effector T cells (Kiefer and Kreutzberg, 1991). In contrast, the influence of GCs on CD80 and CD86 is controversial. Dexamethasone increased antigen uptake by DCs and decreased the basal expression of CD86 in cell culture (Vanderheyde et al., 1999). In contrast, the expression of B7 molecules in MS patients was not affected after high-dose GC therapy (Boylan et al., 1999).

The influence of GCs on regulatory T ( $T_{reg}$ ) cells was only recently recognized (Figure 2). Administration of dexamethasone to mice increases the relative number of  $CD4^+CD25^+$   $T_{reg}$  cells, which are more resistant to GC-induced apoptosis due to a higher expression of Bcl-2. After dexamethasone treatment  $T_{reg}$  cells express elevated levels of CTLA-4, which may enhance their suppressive capacity (Chen et

al., 2004). Importantly, similar observations were made in humans where the percentage of  $T_{reg}$  cells in MS patients was increased after short-term GC therapy (Navarro et al., 2006). Furthermore, concomitant administration of dexamethasone and IL-2 was more successful in treating EAE as compared to dexamethasone alone, which correlated with the expansion of  $T_{reg}$  cells *in vivo* (Chen et al., 2006). Thus, effects on  $T_{reg}$  cells presumably contribute to the therapeutic efficacy of GCs.

In summary, GCs exert immunosuppressive activities by acting on both T cells and myeloid cells, e.g. macrophages and microglia. This leads to a suppression of  $T_H1$  and an induction of  $T_H2$  cytokines, the repression of NO production, impaired T cell activation and a higher number of  $T_{reg}$  cells. In combination, these effects may significantly contribute to the beneficial clinical effect of GCs in the treatment of neuroinflammatory diseases.

## 5. Side effects of glucocorticoid therapy

Despite the unsurpassed efficacy of GCs in the treatment of MS, such therapies are accompanied by multiple side effects. These compromise mood disorders, gastrointestinal pain and headache, while chronic administration may additionally lead to osteoporosis, diabetes and an increased susceptibility to infectious diseases (Pozzilli et al., 2004). Importantly, these side effects depend on the duration, dose and combination with other therapeutic agents and complicate MS therapy to a not yet fully understood degree.

Although largely neglected in the context of MS, GCs exert a number of effects on neurons, which may be either protective as well as harmful. In one study it was shown that the addition of dexamethasone to cultured neurons accelerates the



myelination of axons. This process depends on the GR because it could be inhibited by RU486, an antagonist of the GR (Chan et al., 1998). In contrast, neurotoxic effects were also reported after high-dose GC therapy. In an optic neuritis model high-dose methylprednisolone administration augmented retinal ganglion cell apoptosis during chronic inflammation (Diem et al., 2003). This could be circumvented by a combination therapy with erythropoietin where the neurodegenerative effects were reduced (Diem et al., 2005). Moreover, it was observed that the fast withdrawal of GCs can lead to severe relapses (Reder et al., 1994), which can be prevented by slowly reducing GCs instead of a sudden withdrawal. This led to the current treatment protocol used in MS patients that includes an oral taper of methylprednisolone (Beck et al., 1992). Unfortunately, GC therapy has no beneficial long-term effect on disease progression in MS patients. Although GCs rapidly induce apoptosis and cause acute immunosuppression, this does not result in a persistent elimination or silencing of autoreactive lymphocytes. Furthermore, prolonged GC treatment often induces GC resistance by down-regulating GR levels or altering signal transduction pathways. Thus, GCs are an effective means to treat acute relapses while drugs such as Interferon- $\beta$ , Glatiramer Acetate or Tysabri are rather used to generally impact disease progression.

## **6. Conclusions and future directions**

Despite the long history of GC research and the wide usage of these drugs in the clinical management of MS there are still many open questions. Firstly, which molecular mechanisms underlie the effects of GCs in MS and how could this knowledge be exploited to develop new drugs with an improved therapeutic profile?

Also the cellular basis of GC effects continues to attract attention, e.g. which cell-types are the predominant targets of GCs in MS therapy and which of these effects are most crucial? Is it induction of apoptosis, the down-regulation of inflammatory mediators, compromised antigen presentation or effects on the BBB? Based on this knowledge, it will be interesting to see whether it becomes possible to improve the success of GC therapy by introducing different synthetic compounds. Whereas all these issues are related to the therapy of overt MS and EAE with GCs, the role of stress in the susceptibility to autoimmune diseases also remains debatable. How do endogenous GCs for example impact on the immune system and how does this modulate the development of autoimmune responses? Many of these questions can be addressed using improved animal models, especially some of the newly generated knock-out and transgenic mouse strains (Reichardt, 2004). Thus, basic and clinical research on the role that GCs play in MS will continue to be a fertile and fascinating ground that bears the potential to improve not only the understanding of this devastating autoimmune disease but, most importantly, also its clinical management.

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## References

- Adams, R.A., Bauer, J., Flick, M.J., Sikorski, S.L., Nuriel, T., Lassmann, H., Degen, J.L., Akassoglou, K., 2007. The fibrin-derived  $\gamma$ 377-395 peptide inhibits microglia activation and suppresses relapsing paralysis in central nervous system autoimmune disease. *J. Exp. Med.* 204, 571-582.
- Almawi, W.Y., Beyhum, H.N., Rahme, A.A., Rieder, M.J., 1996. Regulation of cytokine and cytokine receptor expression by glucocorticoids. *J. Leukoc. Biol.* 60, 563-572.
- Ballabh, P., Braun, A., Nedergaard, M., 2004. The blood-brain barrier: an overview: structure, regulation, and clinical implications. *Neurobiol. Dis.* 16, 1-13.
- Bartholome, B., Spies, C.M., Gaber, T., Schuchmann, S., Berki, T., Kunkel, D., Bienert, M., Radbruch, A., Burmester, G.R., Lauster, R., Scheffold, A., Buttgereit, F., 2004. Membrane glucocorticoid receptors (mGCR) are expressed in normal human peripheral blood mononuclear cells and up-regulated after in vitro stimulation and in patients with rheumatoid arthritis. *Faseb J.* 18, 70-80.
- Beato, M., Herrlich, P., Schütz, G., 1995. Steroid hormone receptors: many actors in search of a plot. *Cell* 83, 851-857.
- Beck, R.W., Cleary, P.A., Anderson, M.M., Jr., Keltner, J.L., Shults, W.T., Kaufman, D.I., Buckley, E.G., Corbett, J.J., Kupersmith, M.J., Miller, N.R., et al., 1992. A randomized, controlled trial of corticosteroids in the treatment of acute optic neuritis. The Optic Neuritis Study Group. *N. Engl. J. Med.* 326, 581-588.
- Ben-Nun, A., Wekerle, H., Cohen, I.R., 1981. The rapid isolation of clonable antigen-specific T lymphocyte lines capable of mediating autoimmune encephalomyelitis. *Eur. J. Immunol.* 11, 195-199.

- Bjartmar, C., Trapp, B.D., 2001. Axonal and neuronal degeneration in multiple sclerosis: mechanisms and functional consequences. *Curr. Opin. Neurol.* 14, 271-278.
- Boylan, M.T., Crockard, A.D., McDonnell, G.V., Armstrong, M.A., Hawkins, S.A., 1999. CD80 (B7-1) and CD86 (B7-2) expression in multiple sclerosis patients: clinical subtype specific variation in peripheral monocytes and B cells and lack of modulation by high dose methylprednisolone. *J. Neurol. Sci.* 167, 79-89.
- Chan, J.R., Phillips, L.J., 2nd, Glaser, M., 1998. Glucocorticoids and progestins signal the initiation and enhance the rate of myelin formation. *Proc Natl. Acad. Sci. U S A* 95, 10459-10464.
- Chen, X., Murakami, T., Oppenheim, J.J., Howard, O.M., 2004. Differential response of murine CD4+CD25+ and CD4+CD25- T cells to dexamethasone-induced cell death. *Eur. J. Immunol.* 34, 859-869.
- Chen, X., Oppenheim, J.J., Winkler-Pickett, R.T., Ortaldo, J.R., Howard, O.M., 2006. Glucocorticoid amplifies IL-2-dependent expansion of functional FoxP3(+)CD4(+)CD25(+) T regulatory cells in vivo and enhances their capacity to suppress EAE. *Eur. J. Immunol.* 36, 2139-2149.
- Correale, J., Arias, M., Gilmore, W., 1998. Steroid hormone regulation of cytokine secretion by proteolipid protein-specific CD4+ T cell clones isolated from multiple sclerosis patients and normal control subjects. *J. Immunol.* 161, 3365-3374.
- Diem, R., Hobom, M., Maier, K., Weissert, R., Storch, M.K., Meyer, R., Bähr, M., 2003. Methylprednisolone increases neuronal apoptosis during autoimmune CNS inflammation by inhibition of an endogenous neuroprotective pathway. *J. Neurosci.* 23, 6993-7000.

- Diem, R., Sattler, M.B., Merkler, D., Demmer, I., Maier, K., Stadelmann, C., Ehrenreich, H., Bähr, M., 2005. Combined therapy with methylprednisolone and erythropoietin in a model of multiple sclerosis. *Brain* 128, 375-385.
- Elenkov, I.J., Chrousos, G.P., 1999. Stress Hormones, Th1/Th2 patterns, Pro/Anti-inflammatory Cytokines and Susceptibility to Disease. *Trends Endocrinol. Metab.* 10, 359-368.
- Elovaara, I., Kuusisto, H., Paalavuo, R., Sarkijarvi, S., Lehtimaki, T., Huhtala, H., Vilpo, J., 2006. Effect of high-dose methylprednisolone treatment on CCR5 expression on blood cells in MS exacerbation. *Acta Neurol. Scand.* 113, 163-166.
- Elovaara, I., Lalla, M., Spare, E., Lehtimaki, T., Dastidar, P., 1998. Methylprednisolone reduces adhesion molecules in blood and cerebrospinal fluid in patients with MS. *Neurology* 51, 1703-1708.
- Elovaara, I., Ukkonen, M., Leppakynnas, M., Lehtimaki, T., Luomala, M., Peltola, J., Dastidar, P., 2000. Adhesion molecules in multiple sclerosis: relation to subtypes of disease and methylprednisolone therapy. *Arch. Neurol.* 57, 546-551.
- Engelhardt, B., 2006. Molecular mechanisms involved in T cell migration across the blood-brain barrier. *J. Neural. Transm.* 113, 477-485.
- Erlacher, M., Michalak, E.M., Kelly, P.N., Labi, V., Niederegger, H., Coultas, L., Adams, J.M., Strasser, A., Villunger, A., 2005. BH3-only proteins Puma and Bim are rate-limiting for gamma-radiation- and glucocorticoid-induced apoptosis of lymphoid cells in vivo. *Blood* 106, 4131-4138.
- Fontoura, P., Steinman, L., Miller, A., 2006. Emerging therapeutic targets in multiple sclerosis. *Curr. Opin. Neurol.* 19, 260-266.

- Förster, C., Silwedel, C., Golenhofen, N., Burek, M., Kietz, S., Mankertz, J., Drenckhan, D., 2005. Occludin as direct target for glucocorticoid-induced improvement of blood-brain barrier properties in a murine in vitro system. *J. Physiol.* 565, 475-486.
- Förster, C., Waschke, J., Burek, M., Leers, J., Drenckhan, D., 2006. Glucocorticoid effects on mouse microvascular endothelial barrier permeability are brain specific. *J. Physiol.* 573, 413-425.
- Friese, M.A., Fugger, L., 2005. Autoreactive CD8+ T cells in multiple sclerosis: a new target for therapy? *Brain* 128, 1747-1763.
- Gayo, A., Mozo, L., Suarez, A., Tunon, A., Lahoz, C., Gutierrez, C., 1998. Glucocorticoids increase IL-10 expression in multiple sclerosis patients with acute relapse. *J. Neuroimmunol.* 85, 122-130.
- Gelati, M., Corsini, E., Dufour, A., Massa, G., Giombini, S., Solero, C.L., Salmaggi, A., 2000. High-dose methylprednisolone reduces cytokine-induced adhesion molecules on human brain endothelium. *Can. J. Neurol. Sci.* 27, 241-244.
- Gold, R., Lington, C., Lassmann, H., 2006. Understanding pathogenesis and therapy of multiple sclerosis via animal models: 70 years of merits and culprits in experimental autoimmune encephalomyelitis research. *Brain* 129, 1953-1971.
- Golde, S., Coles, A., Lindquist, J.A., Compston, A., 2003. Decreased iNOS synthesis mediates dexamethasone-induced protection of neurons from inflammatory injury in vitro. *Eur. J. Neurosci.* 18, 2527-2537.
- Grauer, O., Offenhausser, M., Schmidt, J., Toyka, K.V., Gold, R., 2001. [Glucocorticosteroid therapy in optic neuritis and multiple sclerosis. Evidence from clinical studies and practical recommendations]. *Nervenarzt* 72, 577-589.
- Hafler, D.A., 2004. Multiple sclerosis. *J. Clin. Invest.* 113, 788-794.

- Harkness, K.A., Adamson, P., Sussman, J.D., Davies-Jones, G.A., Greenwood, J., Woodroffe, M.N., 2000. Dexamethasone regulation of matrix metalloproteinase expression in CNS vascular endothelium. *Brain* 123, 698-709.
- Heck, S., Bender, K., Kullmann, M., Gottlicher, M., Herrlich, P., Cato, A.C., 1997. I kappaB alpha-independent downregulation of NF-kappaB activity by glucocorticoid receptor. *Embo J.* 16, 4698-4707.
- Heck, S., Kullmann, M., Gast, A., Ponta, H., Rahmsdorf, H.J., Herrlich, P., Cato, A.C., 1994. A distinct modulating domain in glucocorticoid receptor monomers in the repression of activity of the transcription factor AP-1. *Embo J.* 13, 4087-4095.
- Hemmer, B., Nessler, S., Zhou, D., Kieseier, B., Hartung, H.P., 2006. Immunopathogenesis and immunotherapy of multiple sclerosis. *Nat. Clin. Pract. Neurol.* 2, 201-211.
- Herold, M.J., McPherson, K.G., Reichardt, H.M., 2006. Glucocorticoids in T cell apoptosis and function. *Cell. Mol. Life Sci.* 63, 60-72.
- Karlstedt, K., Sallmen, T., Eriksson, K.S., Lintunen, M., Couraud, P.O., Joo, F., Panula, P., 1999. Lack of histamine synthesis and down-regulation of H1 and H2 receptor mRNA levels by dexamethasone in cerebral endothelial cells. *J. Cereb. Blood Flow. Metab.* 19, 321-330.
- Kawakami, N., Nagerl, U.V., Odoardi, F., Bonhoeffer, T., Wekerle, H., Flügel, A., 2005. Live imaging of effector cell trafficking and autoantigen recognition within the unfolding autoimmune encephalomyelitis lesion. *J. Exp. Med.* 201, 1805-1814.



- Kiefer, R., Kreutzberg, G.W., 1991. Effects of dexamethasone on microglial activation in vivo: selective downregulation of major histocompatibility complex class II expression in regenerating facial nucleus. *J. Neuroimmunol.* 34, 99-108.
- Kleinert, H., Euchenhofer, C., Ihrig-Biedert, I., Forstermann, U., 1996. Glucocorticoids inhibit the induction of nitric oxide synthase II by down-regulating cytokine-induced activity of transcription factor nuclear factor-kappa B. *Mol. Pharmacol.* 49, 15-21.
- Komiyama, Y., Nakae, S., Matsuki, T., Nambu, A., Ishigame, H., Kakuta, S., Sudo, K., Iwakura, Y., 2006. IL-17 plays an important role in the development of experimental autoimmune encephalomyelitis. *J. Immunol.* 177, 566-573.
- Leussink, V.I., Jung, S., Merschdorf, U., Toyka, K.V., Gold, R., 2001. High-dose methylprednisolone therapy in multiple sclerosis induces apoptosis in peripheral blood leukocytes. *Arch. Neurol.* 58, 91-97.
- Limbourg, F.P., Huang, Z., Plumier, J.C., Simoncini, T., Fujioka, M., Tuckermann, J., Schütz, G., Moskowitz, M.A., Liao, J.K., 2002. Rapid nontranscriptional activation of endothelial nitric oxide synthase mediates increased cerebral blood flow and stroke protection by corticosteroids. *J. Clin. Invest.* 110, 1729-1738.
- Lu, N.Z., Cidlowski, J.A., 2005. Translational regulatory mechanisms generate N-terminal glucocorticoid receptor isoforms with unique transcriptional target genes. *Mol. Cell* 18, 331-342.
- Luisi, B.F., Xu, W.X., Otwinowski, Z., Freedman, L.P., Yamamoto, K.R., Sigler, P.B., 1991. Crystallographic analysis of the interaction of the glucocorticoid receptor with DNA. *Nature* 352, 497-505.

- Mangelsdorf, D.J., Thummel, C., Beato, M., Herrlich, P., Schütz, G., Umesono, K., Blumberg, B., Kastner, P., Mark, M., Chambon, P., et al., 1995. The nuclear receptor superfamily: the second decade. *Cell* 83, 835-839.
- McCombe, P.A., Nickson, I., Tabi, Z., Pender, M.P., 1996. Corticosteroid treatment of experimental autoimmune encephalomyelitis in the Lewis rat results in loss of V beta 8.2+ and myelin basic protein-reactive cells from the spinal cord, with increased total T-cell apoptosis but reduced apoptosis of V beta 8.2+ cells. *J. Neuroimmunol.* 70, 93-101.
- Migita, K., Eguchi, K., Kawabe, Y., Nakamura, T., Shirabe, S., Tsukada, T., Ichinose, Y., Nakamura, H., Nagataki, S., 1997. Apoptosis induction in human peripheral blood T lymphocytes by high-dose steroid therapy. *Transplantation* 63, 583-587.
- Milligan, N.M., Newcombe, R., Compston, D.A., 1987. A double-blind controlled trial of high dose methylprednisolone in patients with multiple sclerosis: 1. Clinical effects. *J. Neurol. Neurosurg. Psychiatry* 50, 511-516.
- Mirowska-Guzel, D.M., Kurowska, K., Skierski, J., Koronkiewicz, M., Wicha, W., Kruszewska, J., Czlonkowski, A., Czlonkowska, A., 2006. High dose of intravenously given glucocorticosteroids decrease IL-8 production by monocytes in multiple sclerosis patients treated during relapse. *J. Neuroimmunol.* 176, 134-140.
- Moreira, M.A., Tilbery, C.P., Monteiro, L.P., Teixeira, M.M., Teixeira, A.L., 2006. Effect of the treatment with methylprednisolone on the cerebrospinal fluid and serum levels of CCL2 and CXCL10 chemokines in patients with active multiple sclerosis. *Acta Neurol. Scand.* 114, 109-113.
- Murray, T.J., 2006. Diagnosis and treatment of multiple sclerosis. *Bmj* 332, 525-527.

- Navarro, J., Aristimuno, C., Sanchez-Ramon, S., Vigil, D., Martinez-Gines, M.L., Fernandez-Cruz, E., de Andres, C., 2006. Circulating dendritic cells subsets and regulatory T-cells at multiple sclerosis relapse: differential short-term changes on corticosteroids therapy. *J. Neuroimmunol.* 176, 153-161.
- Nguyen, K.B., McCombe, P.A., Pender, M.P., 1997. Increased apoptosis of T lymphocytes and macrophages in the central and peripheral nervous systems of Lewis rats with experimental autoimmune encephalomyelitis treated with dexamethasone. *J. Neuropathol. Exp. Neurol.* 56, 58-69.
- Noseworthy, J.H., Lucchinetti, C., Rodriguez, M., Weinshenker, B.G., 2000. Multiple sclerosis. *N. Engl. J. Med.* 343, 938-952.
- Paul, C., Bolton, C., 1995. Inhibition of blood-brain barrier disruption in experimental allergic encephalomyelitis by short-term therapy with dexamethasone or cyclosporin A. *Int. J. Immunopharmacol.* 17, 497-503.
- Pender, M.P., McCombe, P.A., Yoong, G., Nguyen, K.B., 1992. Apoptosis of alpha beta T lymphocytes in the nervous system in experimental autoimmune encephalomyelitis: its possible implications for recovery and acquired tolerance. *J. Autoimmun.* 5, 401-410.
- Pozzilli, C., Marinelli, F., Romano, S., Bagnato, F., 2004. Corticosteroids treatment. *J. Neurol. Sci.* 223, 47-51.
- Purton, J.F., Monk, J.A., Liddicoat, D.R., Kyparissoudis, K., Sakka, S., Richardson, S.J., Godfrey, D.I., Cole, T.J., 2004. Expression of the glucocorticoid receptor from the 1A promoter correlates with T lymphocyte sensitivity to glucocorticoid-induced cell death. *J. Immunol.* 173, 3816-3824.
- Reder, A.T., Thapar, M., Jensen, M.A., 1994. A reduction in serum glucocorticoids provokes experimental allergic encephalomyelitis: implications for treatment of inflammatory brain disease. *Neurology* 44, 2289-2294.

- Redford, E.J., Kapoor, R., Smith, K.J., 1997. Nitric oxide donors reversibly block axonal conduction: demyelinated axons are especially susceptible. *Brain* 120, 2149-2157.
- Reichardt, H.M., 2004. Immunomodulatory activities of glucocorticoids: insights from transgenesis and gene targeting. *Curr. Pharm. Des.* 10, 2797-2805.
- Reichardt, H.M., Gold, R., Lühder, F., 2006. Glucocorticoids in multiple sclerosis and experimental autoimmune encephalomyelitis. *Expert Rev. Neurother.* 6, 1657-1670.
- Reichardt, H.M., Kaestner, K.H., Tuckermann, J., Kretz, O., Wessely, O., Bock, R., Gass, P., Schmid, W., Herrlich, P., Angel, P., Schütz, G., 1998. DNA binding of the glucocorticoid receptor is not essential for survival. *Cell* 93, 531-541.
- Rivers, T.M., Sprunt, D.H., Berry, G.P., 1933. Observations on attempts to produce acute disseminated encephalomyelitis in monkeys. *J. Exp. Med.* 58.
- Romero, I.A., Radewicz, K., Jubin, E., Michel, C.C., Greenwood, J., Couraud, P.O., Adamson, P., 2003. Changes in cytoskeletal and tight junctional proteins correlate with decreased permeability induced by dexamethasone in cultured rat brain endothelial cells. *Neurosci. Lett.* 26, 112-116.
- Rosenberg, G.A., Dencoff, J.E., Correa, N., Jr., Reiners, M., Ford, C.C., 1996. Effect of steroids on CSF matrix metalloproteinases in multiple sclerosis: relation to blood-brain barrier injury. *Neurology* 46, 1626-1632.
- Schmidt, J., Gold, R., Schonrock, L., Zettl, U.K., Hartung, H.P., Toyka, K.V., 2000. T-cell apoptosis in situ in experimental autoimmune encephalomyelitis following methylprednisolone pulse therapy. *Brain* 123, 1431-1441.
- Screpanti, I., Morrone, S., Meco, D., Santoni, A., Gulino, A., Paolini, R., Crisanti, A., Mathieson, B.J., Frati, L., 1989. Steroid sensitivity of thymocyte subpopulations during intrathymic differentiation. Effects of 17 beta-estradiol

- and dexamethasone on subsets expressing T cell antigen receptor or IL-2 receptor. *J. Immunol.* 142, 3378-3383.
- Sloka, J.S., Stefanelli, M., 2005. The mechanism of action of methylprednisolone in the treatment of multiple sclerosis. *Mult. Scler.* 11, 425-432.
- Smith, T., Schmied, M., Hewson, A.K., Lassmann, H., Cuzner, M.L., 1996. Apoptosis of T cells and macrophages in the central nervous system of intact and adrenalectomized Lewis rats during experimental allergic encephalomyelitis. *J. Autoimmun.* 9, 167-174.
- Song, I.H., Buttgereit, F., 2006. Non-genomic glucocorticoid effects to provide the basis for new drug developments. *Mol. Cell. Endocrinol.* 246, 142-146.
- Sorensen, T.L., Trebst, C., Kivisakk, P., Klaege, K.L., Majmudar, A., Ravid, R., Lassmann, H., Olsen, D.B., Strieter, R.M., Ransohoff, R.M., Sellebjerg, F., 2002. Multiple sclerosis: a study of CXCL10 and CXCR3 co-localization in the inflamed central nervous system. *J. Neuroimmunol.* 127, 59-68.
- Sospedra, M., Martin, R., 2005. Immunology of multiple sclerosis. *Annu. Rev. Immunol.* 23, 683-747.
- Stoecklin, E., Wissler, M., Moriggi, R., Groner, B., 1997. Specific DNA binding of Stat5, but not of glucocorticoid receptor, is required for their functional cooperation in the regulation of gene transcription. *Mol. Cell. Biol.* 17, 6708-6716.
- Swanborg, R.H., 2001. Experimental autoimmune encephalomyelitis in the rat: lessons in T-cell immunology and autoreactivity. *Immunol. Rev.* 184, 129-135.
- Tabi, Z., McCombe, P.A., Pender, M.P., 1994. Apoptotic elimination of V beta 8.2+ cells from the central nervous system during recovery from experimental autoimmune encephalomyelitis induced by the passive transfer of V beta 8.2+ encephalitogenic T cells. *Eur. J. Immunol.* 24, 2609-2617.

- Tischner, D., Weishaupt, A., van den Brandt, J., Müller, N., Beyersdorf, N., Ip, C.W., Toyka, K.V., Hünig, T., Gold, R., Kerkau, T., Reichardt, H.M., 2006. Polyclonal expansion of regulatory T cells interferes with effector cell migration in a model of multiple sclerosis. *Brain* 129, 2635-2647.
- Tuckermann, J.P., Kleiman, A., McPherson, K.G., Reichardt, H.M., 2005. Molecular mechanisms of glucocorticoids in the control of inflammation and lymphocyte apoptosis. *Crit. Rev. Clin. Lab. Sci.* 42, 71-104.
- van den Brandt, J., Lühder, F., McPherson, K.G., de Graaf, K.L., Tischner, D., Wiehr, S., Herrmann, T., Weissert, R., Gold, R., Reichardt, H.M., 2007. Enhanced glucocorticoid receptor signalling in T cells impacts thymocyte apoptosis and adaptive immune responses. *Am. J. Pathol.* 170, 1041-1053.
- Vanderheyde, N., Verhasselt, V., Goldman, M., Willems, F., 1999. Inhibition of human dendritic cell functions by methylprednisolone. *Transplantation* 67, 1342-1347.
- Wang, D., Müller, N., McPherson, K.G., Reichardt, H.M., 2006. Glucocorticoids engage different signal transduction pathways to induce apoptosis in thymocytes and mature T cells. *J. Immunol.* 176, 1695-1702.
- Wang, H.Y., Matsui, M., Araya, S., Onai, N., Matsushima, K., Saida, T., 2003. Immune parameters associated with early treatment effects of high-dose intravenous methylprednisolone in multiple sclerosis. *J. Neurol. Sci.* 216, 61-66.
- Webster, J.C., Jewell, C.M., Bodwell, J.E., Munck, A., Sar, M., Cidlowski, J.A., 1997. Mouse glucocorticoid receptor phosphorylation status influences multiple functions of the receptor protein. *J. Biol. Chem.* 272, 9287-9293.

**Table 1: Beneficial effects of glucocorticoids in MS and EAE**

| <b>mechanism</b>  | <b>effects</b>  | <b>reference</b>   |
|---|---|--|
| <b>apoptosis</b>  | induce cell death (dose-dependent), more pronounced amongst CD4 <sup>+</sup> T cells, involved in remission of EAE  | Migita et al., 1997; Schmidt et al., 2000; Screpanti et al., 1989; Tabi et al., 1994             |
| <b>blood brain-barrier</b>  | prevent BBB disruption  | Engelhardt 2006  |
| direct effect on endothelial cells                                | accumulation of F-Actin in the cell periphery, upregulation of ZO-1 and occludin  | Förster et al., 2005; Förster et al., 2006; Romero et al., 2003                                  |
| interaction between endothelial cells and infiltrating leukocytes | down-regulation of integrines (LFA-1, VLA-4) on leukocytes and adhesion molecules (ICAM-1, VCAM-1, E-selectin) on endothelial cells                                       | Elovaara et al., 1998; Gelati et al., 2000   |
| extracellular matrix remodelling                                  | down-regulation of gelatinase B, urokinase-type plasminogen activator, increase of TIMPS, upregulation of MMP-9 and histamine H <sub>1</sub> and H <sub>2</sub> receptors | Harkness et al., 2000; Karlstedt et al., 1999; Rosenberg et al., 1996                            |
| migration along chemokine gradients                               | down-regulation of CXCR-3 and CCR-5 on blood leukocytes, diminished level of IP-10 and up-regulation of CCL2 in CSF, decreased production of IL-8                         | Elovaara et al., 2006; Mirowska-Guzel et al., 2006; Moreira et al., 2006; Wang et al., 2005      |
| <b>immunosuppression</b>  | cause a T <sub>H</sub> 2 bias   |  |
| T <sub>H</sub> 1 cytokines  | decreased production and release of pro-inflammatory cytokines such as IFN- $\gamma$ , TNF- $\alpha$ , IL-2, IL-1 $\beta$ and IL-12                                       | Almawi et al., 1996  |
| T <sub>H</sub> 2 cytokines  | increased levels of IL-10 and TGF- $\beta$  | Correale et al., 1998; Gayo et al., 1998   |
| NO production<br>antigen presentation                             | reduced NO levels<br>diminished MHC II expression on microglial cells, the influence on B7 molecules is controversial   | Golde et al., 2003<br>Boylan et al., 1999; Kiefer and Kreutzberg, 2001; Vanderheyde et al., 1999 |
| regulatory T cells  | increased relative number of T <sub>reg</sub> cells, up-regulation of CTLA-4  | Chen et al., 2004; Chen et al., 2006   |

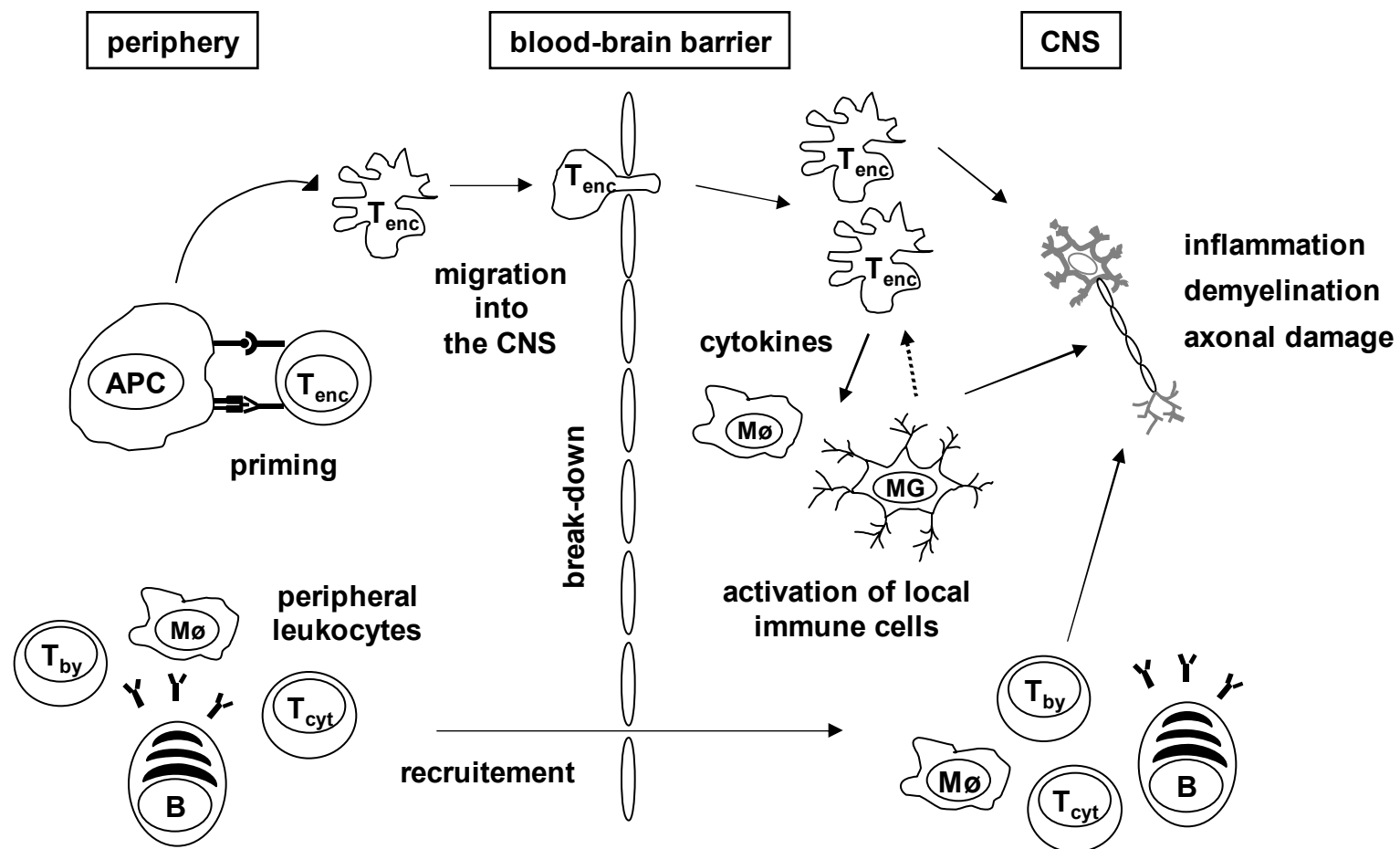
## Figure legends

**Figure 1. Pathomechanisms of multiple sclerosis.** Encephalitogenic T cells ( $T_{enc}$ ) are primed by antigen presenting cells (APS) followed by the migration into the CNS. The release of pro-inflammatory cytokines leads to the activation of macrophages ( $M\emptyset$ ) and microglial cells (MG). Peripheral leukocytes such as macrophages, B cells,  $CD8^+$  T cells ( $T_{cyt}$ ) and  $CD4^+$  bystander T cells ( $T_{by}$ ) are recruited to the CNS. In conjunction, this leads to inflammation, demyelination and axonal damage.

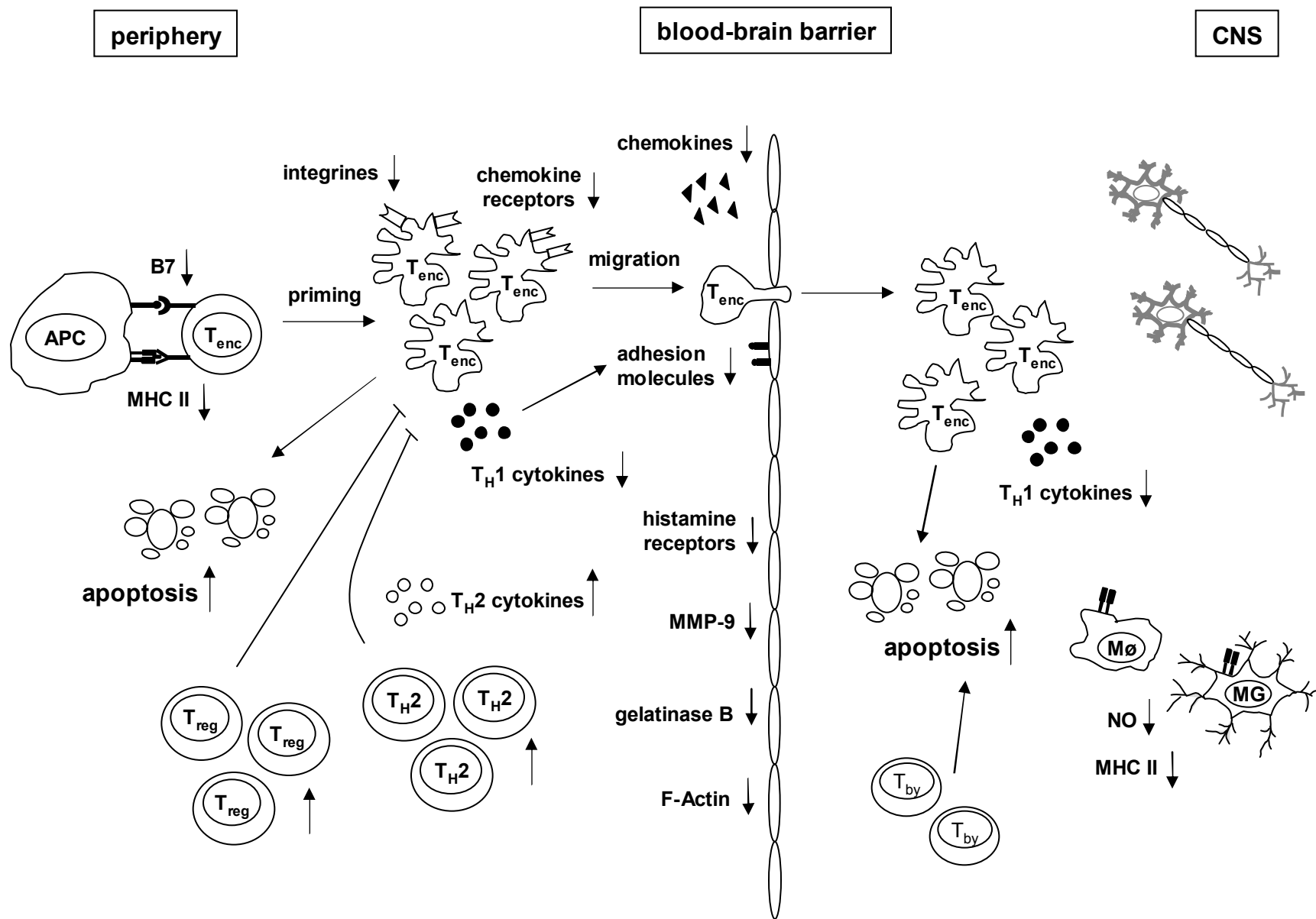
**Figure 2. Glucocorticoid effects on neuroinflammation.** GCs induce or repress the expression of secreted effector molecules and surface receptors on leukocytes and endothelial cells of the BBB. In addition, GCs causes T cell apoptosis in peripheral lymphoid organs and the CNS.  $T_{reg}$  = regulatory T cells;  $T_H2$  = T helper 2 cells.

**Figure 3. Side effects of GCs in MS and EAE.** GCs induce rapid and chronic systemic effects, impact on neurons and may cause relapses upon withdrawal.

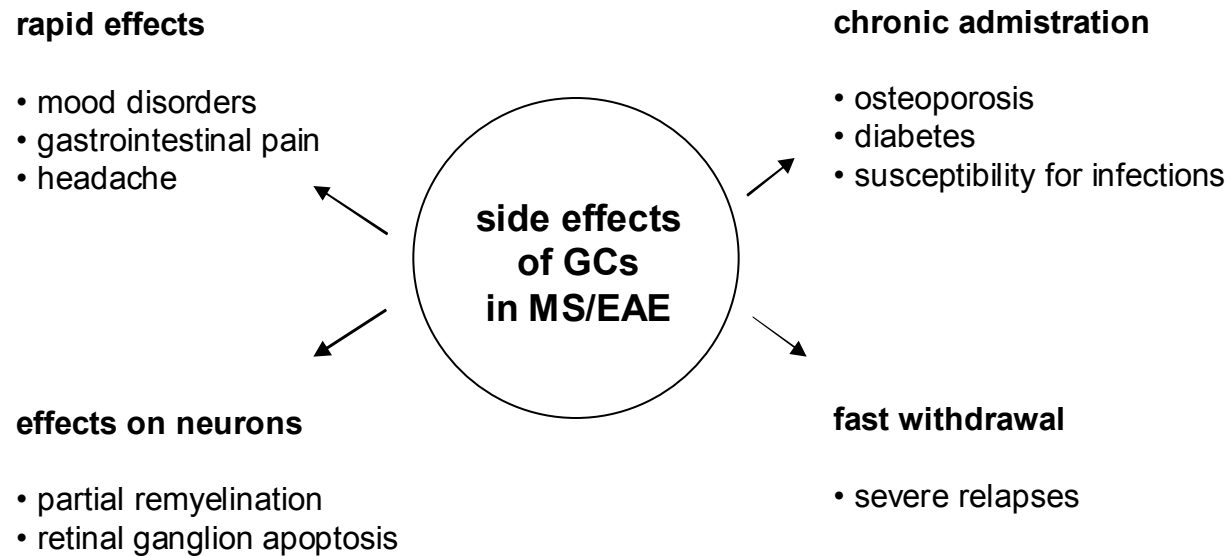




Tischner and Reichardt, Figure 1



Tischner and Reichardt, Figure 2



Tischner and Reichardt, Figure 3