Summary

The brain is protected from invaders by the presence of an endothelial blood-brain barrier (BBB). This implies that the bloodvessels of the brain are different from the rest of the body; they are less permeable for pathogens and large proteins by the presence of tight junctions between endothelial cells surrounding these vessels. Around these bloodvessels perivascular macrophages (PVM) are located. PVM might play a role in the recognition and uptake of pathogens and their degradation products present in the bloodstream or from the brain parenchyma (Kida et al., 1993; Mato et al., 1996), and as such may contribute significantly to the initiation of both innate as well as adaptive immune responses in the brain. The PVM express a variety of receptors enabling pathogen recognition and uptake, including members of the scavenger receptor family. These receptors have a relatively broad ligand binding specificity (Resnick et al., 1994; Krieger, 1997) and can mediate the recognition and uptake of a variety of pathogens, including viruses, bacteria and fungi. The aim of this thesis was to investigate the role of the PVM and the PVM-associated scavenger receptor CD163 in the development of CNS inflammation.

Functional evidence suggests that PVM play a supportive role during experimental autoimmune encephalomyelitis (EAE) in rodents (Polfliet et al., 2002), an animal model for Multiple Sclerosis (MS). However, PVM in the human CNS were still poorly characterized. We investigated CD163 expression in the normal human brain and in the brain of MS patients (Chapter 4) and showed expression of CD163, primarily on PVM and to a lesser extent on myelin-containing ‘foamy’ macrophages within MS lesions. To gain more insight into the function of PVM in antigen recognition and presentation we studied the co-expression of DC-SIGN, mannose receptor, MHC class II, and several costimulatory molecules by PVM in the normal and inflamed human CNS (MS brain lesions). A subpopulation of the CD163-positive PVM in the human brain express several molecules involved in antigen recognition, presentation, and costimulation. Therefore PVM are equipped to recognize antigen and present it to T cells, supporting a role in the regulation of perivascular inflammation in the human CNS.

The cervical lymph nodes (CLN) are the first draining site of the brain and therefore possibly reflect the first site of encounter between myelin antigens and naïve T lymphocytes (Weller, 1998). Non-human primates with EAE showed the presence of myelin components in cells expressing dendritic cell and macrophage markers in the CLN (De Vos et al., 2002). By means of ultrasound guided fine needle aspiration cytology (USgFNAC) we obtained cells, in vivo, from non-enlarged CLN of MS patients and healthy controls (HC), and found macrophages containing myelin proteins in the CLN of all MS patients, whereas these could only be detected in a minority of HC (Chapter 5). These findings are consistent with antigen transport from the brain to the CLN. Understanding of the mechanisms behind this may be relevant for induction and/or maintenance of autoimmunity in the CNS. Antigen transport in the CNS could either be cell-mediated, cell-independent, or both. PVM are particularly effective scavengers of the perivascular space in the CNS (Kida et al., 1993; Angelov et al.,
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1996; Mato et al., 1996) and may thus play a major role in antigen uptake, but the precise route of the ingested particles and antigens remains unclear.

After studying PVM in the human brain and the expression of CD163 on these cells, we investigated the regulation of the scavenger receptor CD163 on monocytes in MS. CD163 is a tissue macrophage marker (Van den Heuvel et al., 1999), but is also expressed on a small percentage of blood monocytes in humans. CD163 has been implicated to play a role in both homeostatic and inflammatory processes (Moestrup and Moller, 2004) and proteolytic shedding of CD163 from the cell membrane results in a soluble form of the receptor, sCD163 (Droste et al., 1999; Moller et al., 2002; Timmermann and Hogger, 2005). Increased sCD163 levels have been found elevated in a wide range of pathologies (Moestrup and Moller, 2004). In MS, plasma sCD163 was found increased and monocyte membrane CD163 decreased (Chapter 6). sCD163 levels appear dependent not only on the surface expression of CD163, but also on the presence of proteases and protease inhibitors that regulate CD163 shedding (Chapter 6) (Droste et al., 1999; Hintz et al., 2002; Timmermann and Hogger, 2005).

As indicated above CD163 can be shed from the cell membrane by metalloproteinase activity in response to LPS and phorbol esters in vitro (Droste et al., 1999; Hintz et al., 2002; Timmermann and Hogger, 2005). However, CD163 can be potently upregulated by glucocorticoids (GC) in vitro (Chapter 9) (Hogger et al., 1998; Van den Heuvel et al., 1999) and in vivo (Chapter 7). Although a higher production of CD163 can be found in response to GC in vivo, this does not hold true for sCD163 levels after GC. Moreover, we provided evidence that measuring membrane CD163 response in vitro might have prognostic value for predicting individual in vivo glucocorticoid responsiveness of MS patients.

Although a role for CD163 in inflammatory diseases has been postulated the only two well described functions of CD163 are its binding to hemoglobin (Hb)-haptoglobin (Hp) complexes resulting in endocytosis (Kristiansen et al., 2001; Graversen et al., 2002; Madsen et al., 2004) and the capacity of CD163 to trigger cytokine production (Van den Heuvel et al., 1999). In this thesis we identified the rat macrophage ED2 surface antigen as the ortholog of human CD163 (Chapter 9). Moreover, triggering of rat CD163 was shown to induce cytokine and nitric oxide production in macrophages (Chapter 8). We also studied other potential functional aspects of the CD163 molecule in more detail. In particular, our findings implicate CD163 on macrophages as a receptor for erythroblast and suggest a regulatory role for CD163 in erythropoiesis (Chapter 9). This is the first demonstration of a role for CD163 in cell-cell interactions between macrophages and hematopoietic cells and this could also be relevant during (CNS) inflammation.

Two secreted molecules of the scavenger receptor cysteine-rich superfamily class B (SRCR-B), i.e. the salivary agglutinin gp-340 and Spu, have been shown to bind Gram-positive and Gram-negative bacteria (Sarrias et al., 2005). We demonstrated that CD163, also a member of the SRCR-B family, can mediate bacterial recognition and that the second SRCR domain of the molecule mediates this. Furthermore, we demonstrate that bacterial recognition triggers CD163-dependent cytokine production by macrophages. This demonstrates for the first time that CD163 on macrophages can
act as a microbial receptor and suggest a role as an innate sensor in the host response against bacterial infection (Chapter 10).

Collectively, these observations support the idea that the PVM strategically located at the BBB acts as a sensor during inflammatory responses within or outside the CNS. CD163 may be one of the important receptors on PVM by which it may sense inflammation or infection. We have now established that cellular and bacterial ligands of CD163 result in the production of pro-inflammatory cytokines or other effector functions, which might have implications for the development and pathogenesis of MS and meningitis, whether this will contribute to leukocyte infiltration and bacterial clearance in the CNS during inflammation remains to be established.
Reference List


