Blood Brain Barrier Compromise with Endothelial Inflammation may Lead to Autoimmune Loss of Myelin during Multiple Sclerosis

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Abstract: Multiple sclerosis is an autoimmune disease characterized by multifocal areas of inflammation and demyelination within the central nervous system. The mechanism that triggers the disease remains elusive. However, recent findings may indicate that multiple sclerosis, at its source, could be a hemodynamic disorder. It has been found that multiple sclerosis patients exhibit significant stenoses in extracranial veins draining the central nervous system (in azygous and internal jugular veins), which are associated with significant pressure gradients measured across strictures. Such anatomic venous abnormalities were not found in the control group of healthy subjects. In this review, it is hypothesized that pathological refluxing venous flow in the cerebral and spinal veins increases the expression of adhesion molecules, particularly intercellular adhesion molecule-1 (ICAM-1), by the cerebrovascular endothelium. This, in turn, could lead to the increased permeability of the blood-brain barrier. Inflamed and activated endothelium could secrete proinflammatory cytokines, including GM-CSF and TGF-beta . In these settings, monocytes could transform into antigen-presenting cells and initiate an autoimmune attack against myelin-containing cells. Consequently, a potential therapeutic option for multiple sclerosis could be pharmacotherapy with either substances that strengthen the tight-junctions barrier, or with agents that reduce the expression of adhesion molecules. In addition, surgical correction could be an option in some anatomical variants of pathologic venous outflow. We are optimistic that a hemodynamic approach to the multiple sclerosis pathogenesis can open a new chapter of investigations and treatment of this debilitating neurologic disease.

Keywords: Adhesion Molecules, Blood-Brain Barrier, Multiple Sclerosis, Venous Insufficiency.

INTRODUCTION

Multiple sclerosis is an autoimmune disease characterized by multifocal areas of inflammation and demyelination within the central nervous system. This autoimmune process is primarily due to the myelin-reactive $CD4^+$ and $CD8^+$ T cells. In addition, B cells also contribute to the progression of the disease (Alter *et al.*, 2003; Frohman *et al.*, 2005), and it is suspected that interleukin-17-secreting T lymphocytes (Th17 cells) are responsible for the initiation of the inflammatory process (Ifergan *et al.*, 2008). Therapeutic strategies, which are more or less efficient, predominantly target the inflammatory cascade. Although several potential mechanisms (mainly inflammatory demyelination and axonal degeneration) could be involved in the progression of multiple sclerosis, the mechanism triggering the disease remains elusive. In addition, an efficient therapy that can protect a patient from relapses of the disease does not currently exist.

The pathogenesis of multiple sclerosis and its animal model, experimental allergic encephalomyelitis, are attributable to the presence of T cells within the nervous tissue. However, leukocytes can enter the brain and spinal cord parenchyma only after a breakdown of the integrity of the blood-brain barrier. Endotheliocytes of the brain and spinal cord, which differ greatly from those in the periphery (Man *et al.*, 2008), are characterized by the presence of tight junctions, low expression of adhesion molecules, lack of fenestration, and minimal pinocytotic activity. These cells make up the blood-brain barrier that is responsible for maintaining the homeostasis of the central nervous system. The function of the blood-brain barrier is primarily supported by tight-junction complexes between adjacent endothelial cells and a lack of frequent interactions between the endothelium and circulating cells (Hawkins *et al.*, 2005). Therefore, the central nervous system, under normal conditions, is an immunoprivileged organ, and antigens of the nervous tissue are not fully accessible to the immune system, since immune cells cannot easily penetrate the cerebral and spinal parenchyma. Although the loss of integrity of the blood-brain barrier is a hallmark of multiple sclerosis and its animal model disease, experimental allergic encephalomyelitis, it remains unclear whether this disruption is secondary to the inflammation, or, alternatively, if the weakening of tight junctions building this barrier precedes the inflammatory process. In experimental allergic encephalomyelitis, the inflammation of the nervous tissue occurs because of the injection of myelin antigens. Thus, the disassembly of the blood-brain barrier is likely to be a secondary event; still, this is not necessarily the case for multiple sclerosis in humans. The other option is that a not-yet-known factor disassembles the tight junctions building the blood-brain barrier, allows the penetration of brain parenchyma by lymphocytes and, in this way, initiates an autoimmune attack against the nervous tissue antigens (primarily myelin). This review focuses on a potential role for pathological hemodynamics in the cerebral and spinal circulation that could trigger this autoimmune reaction.

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INTRACRANIAL PATHOLOGICAL VENOUS HEMODYNAMICS IN THE MULTIPLE SCLEROSIS PATIENTS

Physicians have known of the close relationship between multiple sclerosis lesions and intracranial veins since 1916, when *JW Dawson* described the periventricular lesions (so-called "Dawson's fingers") that extend along large cerebral veins (Dawson, 1916)(Schelling, 1986)(Charcot, 1868). Multiple sclerosis plaques localize predominantly around small veins; they can also extend along the axis of these vessels (Tan *et al.*, 2000). In the case of acute multiple sclerosis lesion, these veins are characterized by lymphocytic and macrophage perivascular infiltrate (Zamboni, 2006). Although veins in the area of chronic multiple sclerosis foci often lack this perivascular inflammation, they exhibit thickening and hyalinization of their walls (Law *et al.*, 2004; Ge *et al.*, 2005; Adams, 1988). Recent *in vivo* studies with ultrahigh-field MR imaging have confirmed the relationship of multiple sclerosis lesions with small intracranial veins (Ge *et al.*, 2008). For a long time, the association of multiple sclerosis plaques with cerebral veins has been regarded as a secondary phenomenon, since any inflammatory process routinely begins from extravasation of immune cells in the postcapillary venules (Andrian *et al.*, 2000). Recent findings, however, indicate that, in addition to the localization of multiple sclerosis plaques in close contact with intracranial veins, these veins in multiple sclerosis patients are characterized by pathological flow patterns (Zamboni *et al.*, 2007; Zamboni *et al.*, 2008; Zamboni *et al.*, 2009). This pathological venous hemodynamics could hardly be explained on the basis of an inflammatory process. Moreover, improper cerebral flow (observed *via* an MR examination) has even been detected in patients with minimal demyelination (Law *et al.* 2004). In addition, retinal periphlebitis that is present in 10-20% of multiple sclerosis patients (Rucker, 1947; Kerrison *et al.*, 1994) has been found to precede the disease (Lightman *et al.*, 1987), and to be a risk factor for having a new relapse (Sepulcre *et al.*, 2007). Therefore, pathological venous hemodynamics could be a primary event that subsequently leads to the initiation of an autoimmune process.

Using perfusion MR imaging, *Law et al.* found a significantly decreased cerebral blood flow in patients with multiple sclerosis. In addition, they demonstrated that healthy subjects exhibited a higher cerebral blood flow in the periventricular regions of their brains (the area that is routinely affected by multiple sclerosis), while there were no significant differences in the cerebral blood flow between various regions of the brains of the multiple sclerosis patients (Law *et al.*, 2004).

The internal jugular vein has one anatomical valve that is situated about 0.5 cm above its junction with the subclavian vein (Chung *et al.*, 2008). Similar valves have been also found in the terminal segments of vertebral veins (Scapinelli, 2000) and in the azygous arch (Yeh *et al.*, 2004; Ichikawa *et al.*, 2008). These proximally situated valves may play a role in avoiding or limiting the venous refluxes. Yet, distal internal jugular and cranial veins, as well as vertebral venous plexus, lack venous valves; still, little is known about the mechanisms that rule the intracranial venous hemodynamics, as intracranial venous refluxes, which can be present in cancer patients with obstructed pathways of venous return in the upper mediastinum (Peart *et al.*, 1975), are uncommon findings.

Under normal conditions in the supine position venous outflow from the brain is maintained primarily through internal jugular veins. On the contrary, in the upright position, jugular veins are collapsed and blood outflow is mainly through the vertebral venous plexus and deep cervical veins. Yet, the internal jugular veins could be opened in an upright position after a Valsalva maneuver (Baumgartner *et al.*, 1997; Menegatti *et al.*, 2008; Stolz *et al.*, 1999; Gisolf *et al.*, 2004; Chung *et al.*, 2008). *Zamboni et al.*, using transcranial color-coded duplex sonography, found pathological venous refluxes within the intracranial veins (deep middle cerebral veins and transverse sinus) during the activation of a thoracic pump in about 50% of the multiple sclerosis patients. Such refluxes were absent in the deep middle cerebral veins in healthy subjects, and only 7% of healthy individuals exhibited venous refluxes in the transverse sinus (Zamboni *et al.*, 2007). Moreover, further studies have shown that multiple sclerosis patients exhibited at least two of five patterns of pathologic cerebral venous outflow: 1) reflux in the internal jugular and/or vertebral veins over 0.88 s; 2) reflux in the deep cerebral veins over 0.5 s; 3) at least 50% stenosis of the proximal internal jugular vein/veins; 4) no detectable flow in the internal jugular and/or vertebral veins; 5) no position-dependent change in diameter of the internal jugular vein/veins (Zamboni *et al.*, 2009).

Until recently, the exact cause of these pathological intracranial venous refluxes and pathologic cerebral venous outflows remained undetermined. But in the currently-published study by *Zamboni*, it was found that multiple sclerosis patients exhibited significant stenoses in extracranial veins that drained the central nervous system (Zamboni *et al.*, 2008). In addition, these lesions were associated with significant pressure gradients measured across a stricture. It is important to note that such anatomic venous abnormalities were not found in the control group of healthy subjects. Venous stenoses or occlusions have been found in the azygous vein and/or internal jugular veins. Four distinct patterns of pathologic venous outflows associated with multiple sclerosis have been described: type A – characterized by an obstruction of the proximal azygous vein accompanied by a stenosis of one of the internal jugular veins; type B - characterized by an obstruction of the proximal azygous vein and bilateral stenoses of the internal jugular veins; type C - characterized by bilateral stenoses of the internal jugular veins and the normal azygous vein; and type D - characterized by multiple stenoses and occlusions in the azygous vein system. Interestingly, these distinct flow patterns were accompanied by different clinical manifestations of multiple sclerosis. Types A, B and C were predominantly seen in patients with relapsing-remitting and secondary progressive clinical courses, while type D was associated with the primary progressive course (Zamboni *et al.*, 2008). Thus, the localization of venous obstructions could significantly influence the clinical picture and prognosis of the disease. However, these preliminary findings undoubtedly should be proven by other investigators in order to establish venous abnormalities as the primary cause of multiple sclerosis.

Table 1. Findings Favoring the Idea of Venous Background of Multiple Sclerosis

Year	Examination	MS Patients	Author; Journal
1868	authopsy	vasocentric localization of MS plaques	JM Charcot; *Gazette Hosp Paris*
1916	authopsy	periventricular lesions extending along large cerebral veins ("Dawson's fingers")	JW Dawson; *Trans Roy Soc Edinb*
1947	ophtalmoscopic examination	signs of retinal periphlebitis	CW Rucker; *Trans Am Ophthalmol Soc*
1986	skull radiography	widening of the main venous passageways of the skull	F Schelling; *Med Hypothes*
1994	histological examination of cadaveric eyes	retinal periphlebitis	JB Kerrison; *Retina*
2000	MR venography	perivenous distribution of MS lesions in the brain	IL Tan; *Am J Neuroradiol*
2004	perfusion MR imaging	decreased perfusion of the periventricular white matter	M Law; *Radiology*
2007	transcranial doppler sonography	pathological venous refluxes within the intracranial veins	P Zamboni; *Curr Neurovasc Res*
2008	ultrahigh-field MR imaging	close relationship of MS plaques with small intracranial veins	Y Ge; *Am J Neuroradiol*
2008	selective phlebography	significant stenoses of extracranial veins (azygous and internal jugular veins) draining the central nervous system	P Zamboni; *J Neurol Neurosurg Psychiatry*
2009	doppler sonography of extra- and intracranial veins draining the brain and spinal cord	patterns of pathologic cerebral venous outflow (reflux in internal jugular vein, vertebral veins or deep cerebral veins, stenosis of the proximal internal jugular vein, no detectable flow in the internal jugular vein or vertebral veins, no position-dependent change in diameter of the internal jugular vein)	P Zamboni; *J Neurol Sci*

TIGHT JUNCTIONS OF THE CEREBROVASCULAR ENDOTHELIUM AND INTEGRITY OF THE BLOOD-BRAIN BARRIER

Tight junctions between endothelial cells in the central nervous system are responsible for the integrity of the blood-brain barrier. The major components of tight junctions are proteins, such as occludin, claudin and JAMs (junctional adhesion molecules), that link to the intracellular cytoskeleton, primarily actin fibers, through ZO (zonula occludens) and other accessory proteins. It has been demonstrated that inhibition of the peroxynitrite-dependent loss of the blood-brain barrier integrity can prevent the development of experimental allergic encephalomyelitis in mice. Thus, the loss of integrity of the blood-brain barrier is the fundamental event in the development of the animal

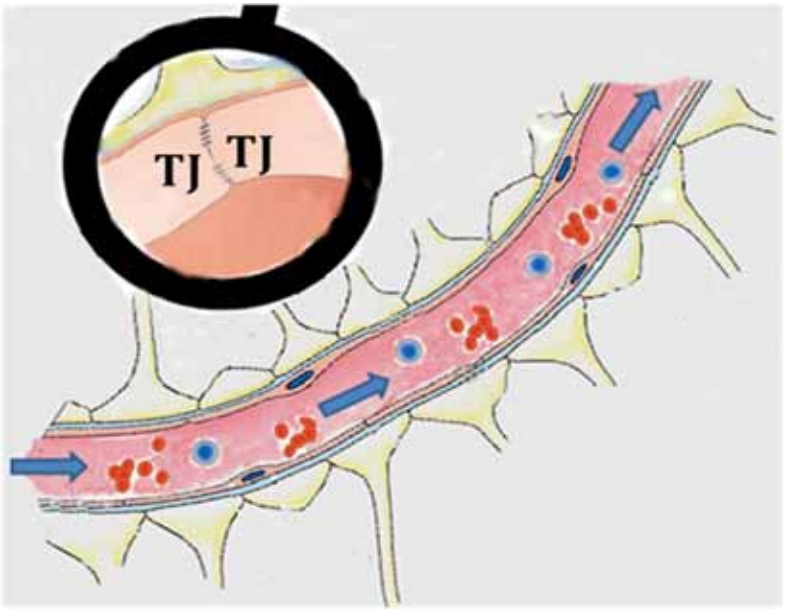

Fig. (**1**). Under normal conditions (monodirectional flow) cerebrovascular endothelium does not express adhesion molecules such as ICAM-1 or VCAM-1, consequently leukocytes do not adhere to the vascular wall; in addition, a high expression of tight junction (TJ) proteins strengthen the blood-brain barrier.

model disease (Fabis *et al.*, 2007) and, perhaps, also of multiple sclerosis in humans (Kermode *et al.*, 1990). It has been found that a subtle blood-brain barrier leakage that was detected *via* MRI is an early finding in multiple sclerosis patients (Soon *et al.*, 2007). The exact mechanism of this blood-brain barrier leakage remains elusive; still, an increase in ICAM-1 expression by the cerebral endothelium may be, at least partially, responsible for the blood-brain barrier disruption (Trojano *et al.*, 2000)(Dietrich *et al.*, 2002). The integrity of the blood-brain barrier is regulated by signals coming from glial cells (Colgan *et al.*, 2008) and from the blood. In addition to this chemical signaling (for instance, the level of cytosolic Ca^{2+}, histamine, thrombin, eicosanoids, and cytokines TNF-α, IL-1, IL-6 and VEGF), the integrity of tight junctions can be regulated by mechanical forces (mainly by the level and character of the shear stress). The influence of shear forces, though, on the behavior of tight junctions is highly dependent on the type of endothelium, and we are still far away from a full understanding of this problem (especially as far as the cerebrovascular endothelium is concerned) (Pearson *et al.*, 2006; de Vries *et al.*, 1997; Brown *et al.*, 2002; Li *et al.*, 1999; Demaio *et al.*, 2001).

Yet, it was recently demonstrated that the blood-flow-associated forces, predominantly the level of shear stress, can profoundly affect the expression of tight junction proteins, thus regulating the strength of the endothelial barrier. It has been shown that increased shear stress, especially with pulsatile flow characteristics, upregulated pivotal tight junction proteins, such as occludin and ZO-1 in the cerebrovascular endothelium. Consequently, increased expression of these proteins was associated with reduced transendothelial permeability (Colgan *et al.*, 2007). In parallel, loss of shear stress after flow cessation enhanced the blood-brain barrier permeability. (Krizanac-Bengez *et al.*, 2006a; Krizanac-Bengez *et al.*, 2006b) Therefore, a reduced shear, for example due to the refluxing venous blood flow, could potentially result in the weakening of the blood-brain barrier. In addition, it has been found that steady shear stress upregulated the activity of the Na-K-Cl cotransporter in cerebral microvascular endothelium. Although it remains unclear whether this protein could control the integrity of the blood-brain barrier, it is suspected that it plays a role in the regulation of endothelial cell volume (Chang *et al.*, 2008; Suvatne *et al.*, 2001).

Leukocytes (T and B cells, monocytes, and others) exert their immunologic functions mainly through direct contact with antigens. This, however, requires precise and active navigation through blood vessels across the endothelial barrier to the target organ. This process, which is called homing and takes place in specialized microvessels, is a multi-step process that includes rolling of a cell, its firm adhesion to the endothelium, and migration across the endothelial barrier toward the gradient of chemokines. Leukocytes precisely home to their targets because of a specialized sets of adhesion molecules, chemokines, and their receptors. Correspondingly, endothelial cells express adhesion molecules that specifically bind their ligands and receptors on target leukocytes (Andrian *et al.*, 2000).

In a manner similar to the immune reactions in other tissues, under inflammatory conditions, activated lymphocytes travel through blood vessels of the brain and spinal cord, are captured by adhesion molecules expressed

Table 2. Flow Characteristics Influencing the Integrity of Endothelium that might be Responsible for the Disruption of the Blood-Brain Barrier

Flow characteristics	Endothelium examined	Molecule, structure or property studied	Effect	Author; Journal
steady or pulsatile shear stress	*in vitro* flow-perfused blood-brain barrier model	expression of occludin and ZO-1 (proteins building tight junctions)	increased expression	OC Colgan; *Am J Physiol Heart Circ Physiol 2007*
steady or pulsatile shear stress	*in vitro* flow-perfused blood-brain barrier model	blood-brain barrier permeability	reduced transendothelial permeability	OC Colgan; *Am J Physiol Heart Circ Physiol 2007*
loss of flow	*in vitro* flow-perfused blood-brain barrier model	blood-brain barrier permeability	increased transendothelial permeability	L Krizanac-Bengez; *Am J Physiol Cell Physiol 2006; J Cell Physiol 2006*
chronic venous insufficiency (venous reflux)	dermal microvessels – patients with mild clinical symptoms	ICAM-1 and VCAM-1 (adhesion molecules responsible for adhesion of leukocytes to the endothelium)	increased expression of both proteins in comparison with healthy individuals	M Peschen; *Acta Derm Venereol 1999*
chronic venous insufficiency (venous reflux)	dermal microvessels – patients with venous ulcers	ICAM-1 and VCAM-1 (adhesion molecules responsible for adhesion of leukocytes to the endothelium)	increased expression of ICAM-1 at the ulcer center	K Rosner; *Acta Derm Venereol 2001*
turbulent flow	*in vitro* human $\alpha_4\beta_1$ integrin–expressing leukocytes ($\alpha_4\beta_1$ integrin is the ligand of VCAM-1 adhesion molecule)	avidity of $\alpha_4\beta_1$ integrin	increased avidity	GJ Zwartz; *J Biol Chem 2004*
steady shear stress	*in vitro* cerebral microvascular endothelium	expression of Na-K-Cl cotransporter	increased expression	E Chang; *Am J Physiol Cell Physiol 2008*

by endotheliocytes, and finally transmigrate across the endothelial barrier. Expression of adhesion molecules by brain and spinal cord endothelial cells is likely to be an essential step in the initiation of this inflammatory process (as, under normal conditions, cerebrovascular endothelium express very low levels of these proteins) (Elovaara *et al.*, 2000).

In the central nervous system, lymphocyte recruitment and transmigration across the endothelial barrier depend primarily on the interactions between ICAM-1 (Intercellular Adhesion Molecule-1), which is expressed by endothelial cells, and its ligand, integrin $\alpha_L\beta_2$, which is expressed by lymphocytes (Adamson *et al.*, 2002; Etienne *et al.*, 1998; Lyck *et al.*, 2003). In addition, an exposure to chemotactic stimulus, which is dependent on chemokines and the G-protein-coupled receptors, seems to be an indispensable step in the process of extravasation of a lymphocyte (in analogy to the case of other microvessels) (Adamson *et al.*, 2002; Alt *et al.*, 2002; Man *et al.*, 2008; Andrian *et al.*, 2000). Interaction between adhesion molecule VCAM-1 (Vascular Cell Adhesion Molecule-1) and integrin $\alpha_4\beta_1$ plays an additional and rather minor role in the lymphocyte transmigration across cerebrovascular endothelium although, in the inflamed spinal cord, T cells can interact with the endothelial VCAM-1 without previous rolling and independent of G-protein-dependent adhesion (Engelhardt, 2006).

Contrary to inflammatory pathologies, expression of adhesion molecules in the cerebral endothelium in healthy individuals is very low. Under normal conditions, cerebral endothelium expresses only low levels of ICAM-1, while other adhesion molecules, such as VCAM-1 and ALCAM (Activated Leukocyte Cell Adhesion Molecule), are not constitutively expressed (Elovaara *et al.*, 2000). Consequently, immune cells very infrequently adhere to endotheliocytes and transmigrate across the blood-brain barrier. This situation, however, changes dramatically during brain inflammation, infection, or ischemia. For example, during infection of the brain, the expression of adhesion molecules in the cerebral microvessels increases, leukocytes adhere to the endothelium, and transmigrate across the blood-brain barrier, as part of the cellular reaction to the microbes.

A similar overexpression of adhesion molecules ICAM-1, VCAM-1, and ALCAM (Lee *et al.*, 2008; Cayrol *et al.*, 2008; Bowen *et al.*, 1995) is observed in multiple sclerosis. Enhanced expression of adhesion molecules in cerebral endothelium consequently leads to increased interactions between endotheliocytes and leukocytes (de Vries *et al.* 1997; Minagar *et al.*, 2003) and to the disruption of the blood-brain barrier. The crosslinking of ICAM-1, as expressed by the cerebrovascular endothelium with its counterpart integrin $\alpha_L\beta_2$ on the lymphocytic surface, not only allows a lymphocyte to adhere to the endothelium but also leads to a weakening of the blood-brain barrier and facilitates the transendothelial lymphocyte migration (Etienne *et al.*, 1998; Walters *et al.*, 2002). Therefore, ICAM-1 expression by the cerebrovascular endothelium is potentially a critical point in the disassembling of this barrier (Lyck *et al.*, 2003; Huber *et al.*, 2006; Huber *et al.*, 2002; Brooks *et al.*, 2005; Walters *et al.*, 2002; Sobel *et al.*,1990). Unfortunately, in this case, normal antigens of the nervous tissue, for instance myelin, can be the targets of an autoimmune attack.

According to recent findings by *Ifergan*, the autoimmune process within the central nervous system is initiated primarily by perivascular dendritic (antigen presenting) cells. These dendritic cells arise from the migration of blood monocytes ($CD11c^+CD14^+$ subpopulation) across the inflamed cerebrovascular endothelium. This process is dependent on the secretion of cytokines GM-CSF (Granulocyte-Monocyte Colony Stimulating Factor) and TGF-β (Transforming Growth Factor-beta) by endotheliocytes and, moreover, is associated with the adhesion of monocytes to the cerebral endothelium (since treatment with anti-ICAM-1 or anti-VCAM-1 antibodies prevented this transformation of monocytes into dendritic cells) (Ifergan *et al.*, 2008). Consequently, the expression of adhesion molecules (ICAM-1 and VCAM-1) by the cerebrovascular endothelium is likely to be

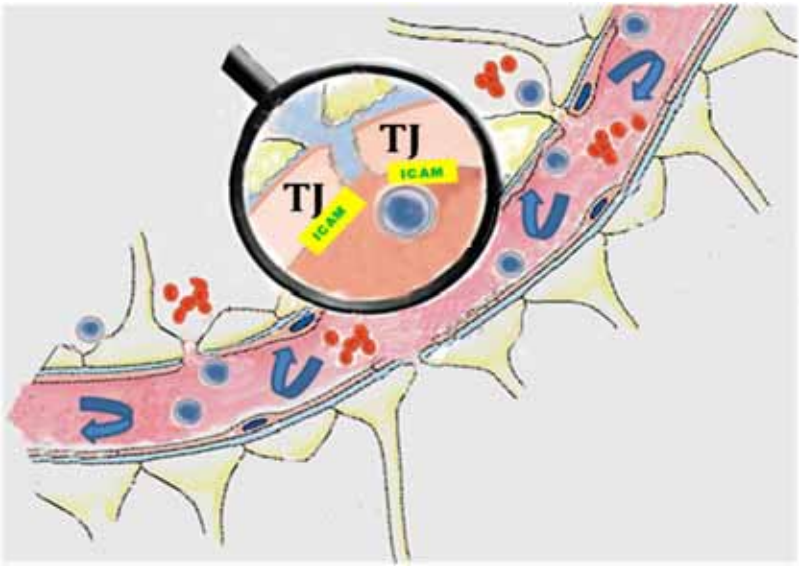

Fig. (**2**). Refluxing blood flow results in overexpression of adhesion molecules (primarily ICAM-1) by cerebrovascular endothelium that leads to adhesion of leukocytes, disruption of tight junctions (TJ), loss of integrity of the blood-brain barrier and extravasation of blood cells into the brain parenchyma.

an important step leading to the initiation of a multiple sclerosis plaque.

HYPOTHETICAL MODEL OF MULTIPLE SCLEROSIS INITIATION

Although not all of the pieces of this puzzle are in their proper place, the following model of the disease could provide a useful framework for future research. Recent findings by *Zamboni* and his team (Zamboni *et al.,* 2008) indicate that multiple sclerosis could fundamentally be a hemodynamic disorder. In addition, some parallels regarding iron metabolism and genetics between multiple sclerosis and chronic venous insufficiency of the lower extremities (Zamboni, 2006; Simka *et al.*, 2008) suggest that further detailed investigations of the extra- and intracranial hemodynamic disturbances could be a promising avenue.

It could be speculated that pathological venous flow in the cerebral and spinal veins increases the expression of adhesion molecules, primarily ICAM-1, by the cerebrovascular endothelium in a manner that is similar to the case of the chronic venous insufficiency. This, in turn, could lead to the increased permeability of the blood-brain barrier, allowing the adhesion of several subpopulations of leukocytes to the endothelium and permitting their extra-vasation to the perivascular interstitium. Inflamed and activated endothelia secrete proinflammatory cytokines, including GM-CSF and TGF-β. In these settings, monocytes could transform into antigen-presenting cells and could initiate the autoimmune attack against myelin-containing cells. It has not yet been determined whether the pathological flow in cerebral and spinal veins could increase the ICAM-1 (and perhaps also VCAM-1) expression by endotheliocytes, but it is possible that such an abnormal flow could trigger overexpression of these adhesion molecules (in analogy with the case of dermal vessels) (Peschen *et al.*, 1999; Rosner *et al.*, 2001; Takase *et al.*, 2004). Moreover, it has been found that vortical and turbulent flow enhance leukocyte/endothelium interactions due to the increased avidity of $\alpha_4\beta_1$ integrin - the ligand of VCAM-1 adhesion molecule (Zwartz *et al.*, 2004).

Consequently, a potential therapeutic option could be pharmacotherapy, either with substances that strengthen the tight-junctions barrier or with agents that reduce the expression of ICAM-1 and other adhesion molecules. For example, luteolin (a flavonoid) was found to suppress the clinical symptoms of experimental allergic encephalomyelitis, prevent monocyte transmigration across the blood-brain barrier, and to modulate the activity of Rho GTPases (the signal transducers), which are involved in the process of migration across the endothelium (Hendriks *et al.*, 2004). Flavonoids are a large group of polyphenolic compounds, some of which (diosmin, hesperidin and others) are currently used in the treatment of chronic venous insufficiency (Simka *et al.*, 2003). However, flavonoids have been found to delay recovery from experimental autoimmune encephalomyelitis (Verbeek *et al.*, 2005). Thus, no data currently exist to clearly support the idea of pharmacotherapy of multiple sclerosis with flavonoids in humans, although the other blood-brain barrier-targeted agents may be useful in the treatment of multiple sclerosis.

Perhaps, in addition to or instead of pharmacotherapy, surgical correction could be an option in some anatomical variants of pathological venous outflow, such as in the localized occlusion of the azygous vein.

Hopefully, a hemodynamic approach to the multiple sclerosis pathogenesis can open a new chapter of investigations and treatment of this debilitating neurologic disease.

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