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CRITICAL REVIEW

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Central nervous system lymphatic unit, immunity, and epilepsy: Is there a link?

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Summary

The recent definition of a network of lymphatic vessels in the meninges surrounding the brain and the spinal cord has advanced our knowledge on the functional anatomy of fluid movement within the central nervous system (CNS). Meningeal lymphatic vessels along dural sinuses and main nerves contribute to cerebrospinal fluid (CSF) drainage, integrating the cerebrovascular and periventricular routes, and forming a circuit that we here define as the CNS-lymphatic unit. The latter unit is important for parenchymal waste clearance, brain homeostasis, and the regulation of immune or inflammatory processes within the brain. Disruption of fluid drain mechanisms may promote or sustain CNS disease, conceivably applicable to epilepsy where extracellular accumulation of macromolecules and metabolic by-products occur in the interstitial and perivascular spaces. Herein we address an emerging concept and propose a theoretical framework on: (a) how a defect of brain clearance of macromolecules could favor neuronal hyperexcitability and seizures, and (b) whether meningeal lymphatic vessel dysfunction contributes to the neuroimmune cross-talk in epileptic pathophysiology. We propose possible molecular interventions targeting meningeal lymphatic dysfunctions, a potential target for immune-mediated epilepsy.

KEYWORDS

acquired epilepsy, central nervous system immune surveillance, immune epilepsy, meningeal lymphatic vessels, parenchymal clearance

1 | INTRODUCTION

The brain has been considered as an immune-privileged organ mainly due to the presence of brain barriers that restrict the relocation of immune cells and the uncertain existence, or relevance, of a central nervous system (CNS) lymphatic drainage.[1,2] This notion was recently amended because of studies describing the presence of functional lymphatic vessels in the meninges surrounding the brain and the spinal cord.[3,4] Experimental evidence has demonstrated that the meningeal lymphatic vessels (MLVs) play a role in the drainage of macromolecules in the brain parenchyma[3,4], and were proposed as a route of communication between the CNS and the immune system.[5]

We here examine MLVs as a contributor of fluid drainage in the CNS, integrating the interstitial fluid (ISF) and the cerebrospinal fluid (CSF) paths. We define this system as a CNS-lymphatic unit, discussing the potential association between flawed MLVs, CSF-ISF drainage, and the generation

of a pro-ictogenic brain environment. We examine the participation of MLVs in the neuroimmune interaction, in response to brain-derived antigens.

1.1 | Lymphatic vessels: basic functions

New knowledge of MLVs is emerging,[3,4,6–8] including the anatomic localization and the implication for draining of solutes and immune cells. However, the exact functions of MLVs in both healthy and pathological conditions remain to be characterized. Due to shared anatomical and functional aspects between meningeal and peripheral lymphatic vessels, we here refer to the latter to revise the lymphatic system fundamental aspects. In the periphery, lymphatic vessels develop in close association with veins in the subcutaneous tissues and alongside arteries in the viscera. Lymphatic capillaries are constituted of a thin wall of endothelial cells, with smooth muscle cells and an adventitia layer present in larger vessels. The existence of openings in the endothelium and specialized valves allows for the collection of interstitial fluid, molecules, and proteins that have leaked from adjacent blood vessels due to damage or pressure changes, cleaning the tissue from the accumulating by-products.[9] Anatomic and functional defects of the peripheral lymphatic system result in the disruption of drainage and the development of lymphedema (accumulation of protein-rich fluid).[10,11] Primary lymphedema is caused by congenital mutations in the genes involved in lymphatic vessel development (eg, Vascular endothelial growth factor receptor 3 [VEGFR-3]). Secondary lymphedema is a consequence of increased tissue pressure following trauma or tumors compressing the vessels, surgeries (eg, the removal of lymph nodes), scar tissue, chronic venous insufficiency, obesity, and infections (eg, filariasis, first cause of lymphedema in developing countries).[11] Each of these conditions can result in the overload of lymphatic transport capacity due to the obstruction or interruption of lymphatic vessels, favoring edema formation.

The lymphatic system is also a key player in immune surveillance.[12] Lymphatic vessels drain soluble and cell-associated antigens from the tissues into regional lymph nodes, where they are presented to T and B lymphocytes via specialized antigen-presenting cells (APCs). The interaction between APCs, lymphocytes, and the lymph node environment establishes whether naive lymphocytes will mount an effector response, become tolerant, or undergo apoptosis to avoid autoimmunity. Therefore, lymphatic vessels play a central role in immune-cell activation and differentiation.[13,14]

1.2 | The meningeal lymphatic vessels

In the CNS, the lymphatic vessels are located in the dura mater facing the subarachnoid space, lining the dural sinuses (the sinuses on the calvarium and the pterygopalatine and the middle meningeal arteries on the cranial base; Figure 1), or along the cranial nerves (trigeminal, optic, and spinal nerves).[3,4,15] Experimental evidence suggests that MLVs are important for the collection of the interstitial fluid solutes from the brain parenchyma, draining into lymph nodes located in the neck (deep and superficial cervical lymph nodes, dcLNs and scLNs, respectively), and participate to the transport of T cells, dendritic cells, and macrophages.[3] The dcLNs are the primary collectors of the MLVs constituting the draining lymph nodes of the CNS[3,4], and are indicated as the principal lymph nodes involved in the immune response to CNS-derived antigens[16]. MLVs are involved in: (a) CNS fluid movement, (b) drainage of solutes from the brain parenchyma, and (c) modulation of the immune response to CNS-derived antigens. MLV dysfunction could participate in the pathogenesis of neurodegenerative diseases, where accumulation of macromolecules in brain parenchyma and a neuro- immune cross-talk occur.[17,18]

Key Points

- Meningeal lymphatic vessels are functionally connected to CSF-ISF drainage pathways, constituting the CNS-lymphatic unit
- The CNS-lymphatic unit contributes to brain interstitial clearance and impacts the neuroimmune interactions
- Functional alterations of the CNS-lymphatic unit may contribute to the pathogenesis of acquired and immune epilepsies

2 | THE CNS-LYMPHATIC UNIT AND PARENCHYMAL WASTE ACCUMULATION: IMPLICATIONS FOR SEIZURES AND EPILEPSY

2.1 | Blood-brain barrier impairment, macromolecule accumulation, and neuronal hyperexcitability

The blood-brain barrier (BBB) is a functional-anatomic unit and a fundamental segment of the cerebrovascular tree. The BBB consists of a multicellular assembly of endothelial cells, astrocytes, and pericytes,[30] with a main function of separating the circulating blood solutes and cells from the ISF and the brain parenchyma.[31]

BBB damage and dysfunction play an important role in generating and sustaining ictal activity.[32–34] Neuronal hyperexcitability can be induced following BBB damage through different mechanisms, including: (a) rapid disequilibrium in parenchymal

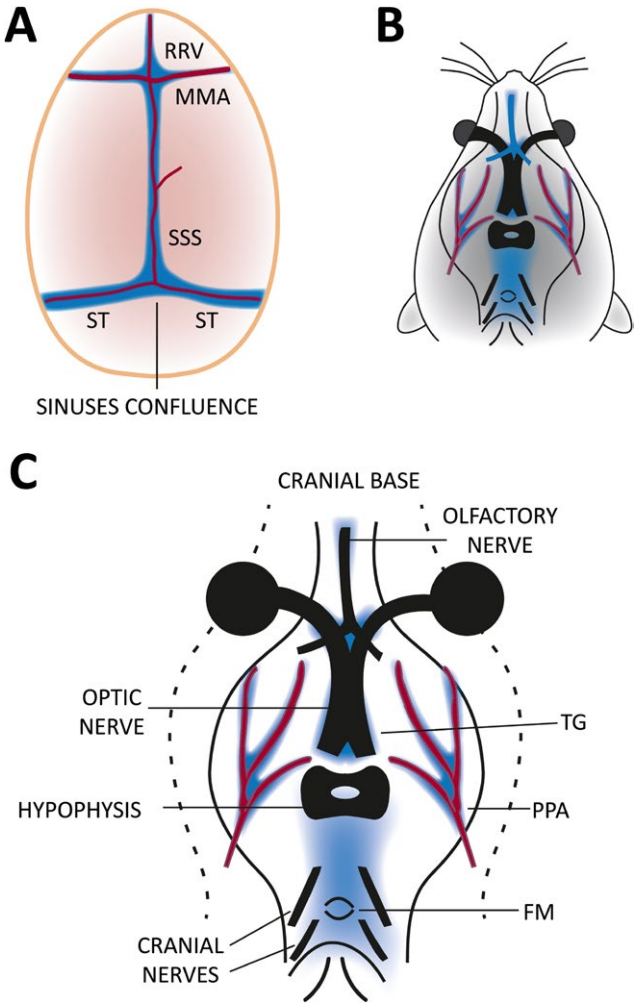

FIGURE 1 Schematic representation of lymphatic flow (light blue) located in the meninges, at the calvarium and cranial fossa (dorsal and ventral side of the cranium). A, In the calvarium, MLVs are located along the dural sinuses and the middle meningeal artery (red), as well as along the rostral rhinal vein and the tentorium around the pineal gland. B, C In the cranial fossa, MLVs are along the pterygopalatine artery (red), the optic and trigeminal nerves, the hypophysis, and the IX-XI cranial nerves. MLVs cover the spinal canal, exiting together with the cranial nerves toward the lymph nodes (C). FM, foramen magnum; MLVs, meningeal lymphatic vessels; MMA, middle meningeal artery; PPA, pterygopalatine artery; RRV, rostral rhinal vein; SSS, sinus sagittalis superior; ST, sinus transversus; TG, trigeminal nerve

ionic concentrations (eg, K^+), impacting the initiation and propagation of action potentials[32]; (b) perivascular and parenchymal accumulation of serum proteins,[35–37] which can promote neuronal damage and hyperexcitability[38] (Box 2); (c) the setting up of a self-sustaining cycle between seizure activity and BBB permeability, driven by increased interstitial glutamate levels,[39] metabolic mismatch (hypometabolism, hypoxia), and/or edema, all resulting in or perpetrating neuroinflammation.[40–42] BBB abnormalities are associated with transient vasogenic or cytotoxic edema in cortical and subcortical ictal regions.[32,43,44] Following BBB damage, interstitial protein accumulation promotes water entry into the brain as well as changes in the lipophilicity of the perivascular space.[45] It is therefore plausible to assume that BBB damage occurring during seizures will interfere with ISF formation and its movement along the arteriole-capillary routes (Box 1). Disrupted ISF circulation during seizures could, in turn, favor the interstitial accumulation of waste products (eg, hyperphosphorylated tubulin-associated protein, pTau,[46,47]), sustaining astrocytes and microglia activation, neuroinflammation and ictal activity.[48]

2.2 | Macromolecule clearance and the meningeal lymphatic vessels (MLVs)

The MLVs contribute to the clearance of solutes from the brain parenchyma.[4] Clearance of cortically injected ovalbumin (45 kDa) was significantly reduced in K14-VEGFR3-Ig mice (K14flt4-tg, a model of congenital lymphedema lacking a functional meningeal lymphatic drainage[49]) as compared to control. By measuring the intensity of the fluorescent signal, Aspelund et al[4] demonstrated that, in physiologic conditions, ovalbumin is cleared from the brain and transported to the dcLNs, presumably through the MLVs located at the base of the skull. In the absence of a functional MLVs (K14flt4-tg mice) ovalbumin accumulates in the brain parenchyma. Similar results were obtained by Louveau et al[8] (identifying 5 "hotspots" of lymphatic drainage in the meninges), and using mice undergoing surgical ligation of the lymphatic vessels afferent to the dcLNs.[3,4] These data demonstrate that MLVs play an important role in the clearance of interstitial accumulating molecules, strengthening the notion of dcLNs as collectors of brain drainage pathways.

Plog et al[50] demonstrated that ISF draining along the perivascular space ends in the dcLNs. By impairing the CSF-ISF exchange (using pharmacologic, surgical, and physical manipulations[24]) the authors observed a reduced clearance of tracers (including ovalbumin) from the brain and a defect in drainage toward the dcLNs. These results suggest that CSF, ISF, and the meningeal lymphatic flows are functionally connected and contribute as a whole to interstitial clearance. Building from this evidence, here we specify a CNS-lymphatic unit, constituted by the structures allowing ISF and CSF movement (ventricles, perivascular space, and basement membrane of capillaries) and the MLVs. Impaired clearance of toxic molecules (eg, amyloid beta or pTau) is a trait of neurodegenerative diseases contributing to neuronal hyperexcitability (Box 2). Therefore, a functional modification of the CNS-lymphatic unit could be pathologic.

3 | ROLE OF MENINGEAL LYMPHATIC VESSELS IN CNS IMMUNE SURVEILLANCE

3.1 | T cells in the CNS lymph nodes

The role of adaptive immunity in the pathophysiology of CNS diseases is emerging.[71–73] Here we address the mechanisms

Box 1: CNS fluids

The cerebrospinal (CSF) and the interstitial fluid (ISF) are the principal fluid components of the CNS. The CSF is an ultrafiltrate of blood plasma, with a low protein content in the ventricles. At spinal cord level, protein concentration in the CSF is higher and includes a component of white blood cells. Its main functions are to protect the brain (buoyancy and shock absorption), to maintain brain homeostasis, and to accumulate waste products, (eg, brain cell metabolites). The CSF is produced by the choroid plexus (up to 80%) filling the lateral, third, and fourth ventricles, while the remaining 20% may derive from the ependymal cells lining the ventricles and from the subarachnoid space.[19,20] The CSF circulates through the ventricles, the cisterns, and fills the subarachnoid space, and may re-enter the cortex via dispersion along large caliber arteries/arterioles.[19,21] A component of the CSF flows along the Virchow-Robin space and in the perivascular space (pia and the glia limitans). The CSF is also assumed to enter the periventricular organs directly from the ventricles[22] (Figure 2). The CSF has a pulsatile flow (along the antero-posterior axis), as systolic and diastolic pressure changes impact CSF flow velocity and direction[21]. The CSF exits the CNS via the arachnoid villi into the sinus sagittalis superior or flows to the nasal lymphatics through the cribriform plate and along principal nerve routes (olfactory, optic, and spinal nerves, where MLVs are also located; Figure 3).
The ISF fills the extracellular space within the brain parenchyma (15%-20% of total brain volume).[23] The ISF has a unique composition of ions, proteins, peptides, and neurotransmitters, essential to maintain the isotonicity of the brain cellular microenvironment.[20] The ISF derives at the BBB from secretion processes, where water follows ionic transport into the brain and across the endothelium (reviewed in Brinker et al[19]). Starling's forces (oncotic vs hydrostatic pressure) control the production of BBB exudate in disease conditions, when serum proteins can access the brain. Movement of ISF in the extracellular space may follow diffusion and convection mechanisms. However, the relative contribution of the two remains to be defined.[21,22] The interchange and mixing between the CSF and ISF is difficult to estimate, as it may vary depending on brain region (eg, depth of the cortical layers or proximity to ventricles where the CSF can diffuse).
The ISF drains along 3 potential pathways[23]: (a) the ventricle wall through ependymal cells, (b) the perivascular (and the Virchow-Robin) space at the surface of the brain,[24] and (c) the blood vessel wall (basement membrane)[20,25,26] (Figure 2). The first 2 pathways allow for ISF-CSF interchange, whereas the third one assumes a direct flow of ISF to the MLVs.
Clearance of molecules from the CNS was proposed to be compartmentalized: solute drainage from the brain parenchyma occurs along the perivascular pathways into the dcLNs,[27] whereas CSF from ventricles and subarachnoid spaces drain to both scLNs and dcLNs[27–29] (Figure 3). Modifications of CSF-ISF drainage due to congenital malformations or as result of lymphatic vessel obstruction could generate proinflammatory conditions due to solute and cell accumulation, promoting neuroimmune reactions.

of adaptive neuroimmunity, focusing on the link between MLVs, dcLNs, and T-cell activation. Available studies point to a pivotal role of dcLNs in CNS immune surveillance.[16,74–76] As previously demonstrated,[16,77] the immune response to CNS-derived antigens is regional. Antigens drained from the CSF or present in the meninges trigger a T-cell response,[77] whereas antigens expressed in the brain parenchyma induce preferably a humoral immune reaction.[16] CSF-ISF clearance (Box 1 and Figure 3) follows distinct pathways (ventricles, periventricular organs, subarachnoid and parenchyma space, or cortical and subcortical regions) determining specific antigen-draining routes toward secondary lymphoid organs, perhaps influencing the immune response (Figure 4). ISF drains mainly to the dcLNs,[4,27] while solutes present in the CSF flow into both scLNs and dcLNs, as well as to lumbar LNs.[19,78–80] In the dcLNs, brain-derived antigens elicit a CNS-specific T-helper immune response (Section 3.2), whereas immune response triggered in the superficial or lumbar lymph nodes has been proposed to be skewed toward CD8+ T-cell activation.[81]

3.2 | Role of deep cervical lymph nodes in brain immune tolerance and response

By injecting immunogenic tumor-derived antigen directly into the brain parenchyma, Harling-Berg and colleagues demonstrated that, in the dcLNs, the evoked immune response is T-helper type 2 (Th2) and B-cell mediated, resulting in antibody production.[16] Injuries to the CNS (eg, optic nerve injury) promote a similar immune response associated with the up-regulation increase of regulatory T cells (T_{reg}, a cell subpopulation pivotal in maintaining tolerance to self-antigens and in preventing autoimmune disease,[82] Figure 4). Dissimilarly, in the peripheral lymphatic organs, CNS-derived antigens elicit a cytotoxic immune response (CD8+ T-cell mediated), without activation of the T_{reg} subpopulation.[82,83] The source of the CNS-derived antigens (parenchymal vs meningeal) may determine the lymph nodes to which the antigens drain to, eventually influencing the immune response. This was proposed as a mechanism to provide brain protection from pathogen infection, at the same time preserving neurons from autoimmune attacks.[84] Of

Box 2: pTau accumulation and neuronal hyperexcitability

Deposits of hyperphosphorylated tubulin-associated protein (pTau) are correlated with neurodegeneration and axonal injury in patients with epilepsy and in experimental models (for a comprehensive review see Ali et al,[51] Saletti et al,[52] and Zheng et al[53]). Accumulation of pTau was reported in brain specimens obtained from patients with focal cortical dysplasia or acquired epilepsy (eg, post-traumatic),[47,54–56] as well as in temporal lobe epilepsy patients with no history of TBI.[57,58] Results were corroborated by using experimental models of epilepsy[59] or of TBI associated with the development of seizures.[60,61] pTau has been implicated in the regulation of neuronal network synchronization[62,63] and in neuroplasticity changes[64,65] resulting in hyperexcitability.[63] In a murine model of Alzheimer disease, the reduction of pTau levels corresponded to decreased electroencephalographic seizures.[62] From a pharmacologic point of view,[66,67] the administration of sodium selenate (a potent activator of tau phosphatase PP2A) resulted in the decrease of pTau and in the reduction of network hyperexcitability or seizure susceptibility,[68] as well as in the inhibition of epileptogenesis.[69] These results support pTau as a common component of neurodegenerative diseases, including acquired epilepsies.[70] As accumulation of pTau is associated with neuronal network excitability, favoring pTau clearance could result in an antiepileptic effect.

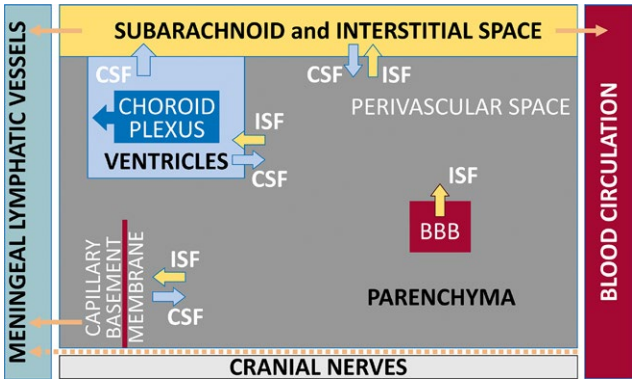

FIGURE 2 Schematic representation of cerebrospinal fluid (CSF) and interstitial fluid (ISF) production and circulation in the brain. CSF is mainly produced by the choroid plexus, whereas ISF derives from secretion at the level of the blood brain barrier (BBB). CSF and ISF interchange and mix at the level of the ventricles, and along the perivascular space or the capillary basement membrane. Arrows show direction and relative contribution of CSF and ISF to the net fluid circulation

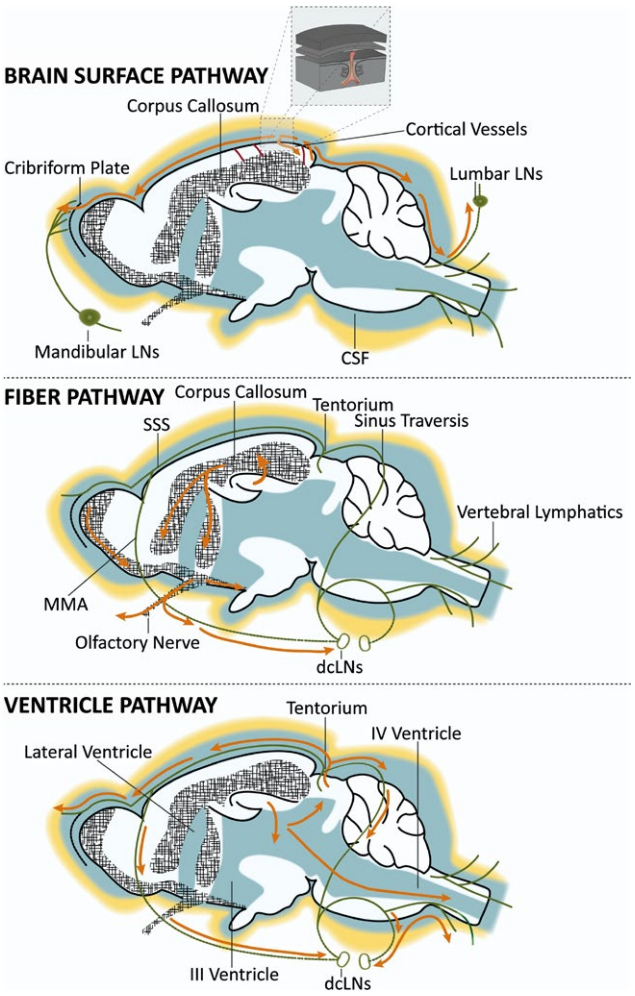

FIGURE 3 Schematic representation of solute drainage in the CNS. A, Cerebrospinal fluid (CSF) in the subarachnoid space is drained through the cribriform plate and collected by the lymphatic vessels present in the nasal cavity (afferent of the mandibular lymph nodes) or is reabsorbed into sinuses via the arachnoid villi. Alternatively, a part of the CSF recirculates from the subarachnoid space into the brain parenchyma along the perivascular spaces surrounding penetrating arteries (Box 1), and exchange with the interstitial fluid in the superficial layers of the neocortex. CSF flowing along the spinal canal is drained though the MLVs and allegedly transported to the lumbar lymph nodes. B, One main route for ISF and solute movement within the brain is along the white matter tracts (eg, corpus callosum, anterior commissures, and stria terminalis), and along the olfactory and optic nerve projections. Here solutes can be collected by the MLVs present in the dura mater running along the intracranial surface of the nerves and transported to the deep cervical lymph nodes (dcLNs). C, Alternatively, solutes can be transported to the ventricular system drained with the CSF. MLVs present in the tentorium and around sinus confluence, as well as the one along the rostral rhinal vein are putative collectors of the solutes drained through this pathway. CSF, cerebrospinal fluid; dcLNs, deep cervical lymph nodes; MLVs, meningeal lymphatic vessels; SSS, sinus sagittalis superior; MMA, middle meningeal artery

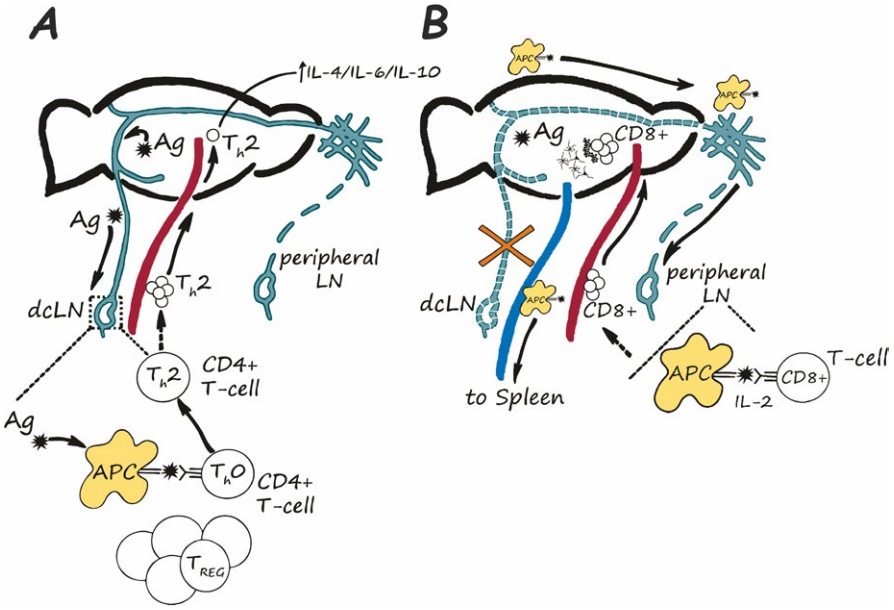

FIGURE 4 Cartoon schematizing alternative immune responses toward brain-derived antigens. A, Soluble antigens from the brain parenchyma are transported along the interstitial fluid (ISF) route and the MLVs (blue-green) to the dcLNs. Here, depending on the inflammatory milieu, they can elicit immune tolerance mechanisms or a noncytotoxic immune reaction (Th2 mediated under T_{reg} regulation), protecting neurons and astrocytes from degeneration. B, A functional defect in one or more elements of the CNS lymphatic unit (eg, MLV congenital malformation or obstruction secondary to brain trauma) could result in drainage of brain-derived antigens to secondary lymphoid organs other thatn the dcLNs (eg, to the spleen via arachnoid villi and the venous system (blue) or to peripheral LNs via the cribriform plate), bypassing the specific neuroimmune response elicited in the dcLNs. Cytotoxic CD8+ T cells could be activated against neuronal or astrocytic self-antigens, homing to the brain, where kill targeted cells (ie, neurons and/or astrocytes). Ag, antigen; APC, antigen-presenting cell; dcLNs, deep cervical lymph nodes; MLVs, meningeal lymphatic vessels; Th0, naive T cell; Th2, type 2 CD4+ T helper; T_{reg}, regulatory T cell

interest, pharmacologic depletion of T_{reg} in the dcLNs resulted in neurodegeneration in a model of optic nerve lesion.[82]

The MLVs are afferent to the dcLNs.[4] We speculate that functional obstruction of MLVs could result in a detour of brain-derived antigens toward alternative secondary lymphatic organs (eg, scLNs, lumbar lymph nodes, or spleen), circumventing the regulation of the neuroimmune response provided by the dcLNs. As a result, the antigens drained from the brain could promote a cytotoxic CD8-mediated auto immune reaction. Our preliminary data obtained using K14flt4-tg mice (lacking MLVs and dcLNs) support this hypothesis showing CD8+ T-cell immune response specifically in the cortical areas surrounding the lesion in a model of traumatic brain injury (TBI; controlled cortical injury [CCI] delivered unilaterally to the somatosensory cortex).

4 | MENINGEAL LYMPHATIC VESSELS AND THE DEVELOPMENT OF AUTOIMMUNE ENCEPHALITIDES

The International League Against Epilepsy (ILAE) has included autoimmunity among the etiologies of epilepsy: "immune epilepsy is the direct result of an immune disorder, in which seizures are a core symptom, and the hallmark is the presence of autoimmune-mediated brain inflammation".[85] Autoimmune encephalitides are classified as follows: (a) encephalitides with pathogenic antibodies against cell surface proteins (eg, anti-NMDA [*N*-methyl-D-aspartate] receptor, anti-LGI1, anti-VGKC complex); (b) T-cell diseases against intracellular antigens (eg, GAD65); and (c) encephalitides associated with other autoimmune disorders (eg, lupus cerebritis).[86] Seizures and status epilepticus are common symptoms in autoimmune encephalitides,[87] which can be resistant to antiepileptic drugs (AEDs) and respond better to immune therapies.[88] Autoimmune encephalitides can relapse,[86] suggesting the presence of a functional defect in the immune surveillance of the CNS.

4.1 | CNS-lymphatic unit and Rasmussen encephalitis pathophysiology: a proposed link

Here we focus on Rasmussen encephalitis (RE), described as focal seizures due to chronic localized encephalitis of probable viral origin.[89] RE is a slow-progressing neurologic disorder, characterized by unilateral brain atrophy and the presence of active microglia/macrophage nodules.[90,91] RE is associated with focal aware or focal impaired awareness seizures with motor onset, or with focal to bilateral tonic-clonic

seizures, and poor response to AEDs.[92] Studies performed using brain specimens obtained from RE patients have indicated the presence of brain-infiltrating cytotoxic CD8+ T cells undergoing clonal local expansion.[93–95] The infiltrating CD8+ T cells are juxtaposed to neurons and astrocytes, with granzyme-B–containing granules polarized toward neuronal or astrocytic membranes.

In his original paper,[89] Rasmussen proposed a brain viral infection as the initiating event eliciting the CD8+ T-cell immune response. This would explain the clonal composition of the T-cell receptor repertoire found in the brain of RE patients[94] and the observed hemispheric distribution with centrifugal expansion,[96] suggestive of a focal infection. However, no sign of viral infection has been found in brain specimens obtained from RE patients.[96]

Here we propose the hypothesis (Figure 4) that the CD8+ -mediated immune response observed in RE may be the result of insufficient lymphatic drainage, either congenital (as in primary lymphedema) or consequent to the obstruction of the lymphatic flow. Under this condition, the control of the neuroimmune response provided by the MLVs-dcLNs may fail and brain-derived antigens could reach the peripheral lymph nodes, where a cytotoxic CD8+ T-cell mediated response occurs. Activated CD8+ T cells could home back to the brain and selectively target those cells (ie, neurons or glia) expressing the self-antigen. This could result in the specific neuronal and astrocytic cell loss observed in RE brains.[97,98]

A possible objection to our hypothesis is that autoimmune responses are usually not focal, whereas RE is. However, localized brain infiltration of activated CD8+ T cells may be facilitated in areas of BBB dysfunction and ongoing neuroinflammation. The latter could be the consequence of a cellular imprint of precedent insults and of a regional damage following head trauma or hypoxic events.[99] Under these conditions, proinflammatory cytokines can upregulate the expression of adhesion molecules (ICAM-1, VCAM-1, and E-selectin) on endothelial cells.[100] These factors bind to specific ligands expressed by the activated leukocytes allowing the adhesion, rolling, and migration of activated T cells across the brain endothelium[79]. In summary, RE could therefore be the result of a double-hit, specifically a, reduced CNS-lymphatic unit efficiency (activating autoimmune T cells) and a brain insult, inducing regional neuroinflammation and BBB dysfunction, that promotes focal lymphocyte CNS recruitment.

5 | CNS-LYMPHATIC UNIT IMPAIRMENT AND MODULATORY APPROACHES

Strategies aimed at regenerating the lymphatic system may represent a supporting therapeutic intervention. It is known that inflammation can directly promote lymphangiogenesis, an extensive and localized growth of lymphatic vessels.[101] Tissue-infiltrating inflammatory cells (eg, CD11b+/Gr-1+ macrophages) are capable of forming tube-like structures displaying lymphatic markers (ie, Lymphatic vessel endothelial hyaluronic acid receptor [Lyve-1], Prospero homeobox protein 1 [Prox1], and podoplanin)[102] and producing the vascular endothelial growth factors VEGF-C and VEGF-D, promoting the genesis of new lymphatic vessels via VEGFR-3 signaling.[102] The newly-formed lymphatic vessels contribute to restore the fluid drainage and counteract the inflammatory processes.[102–105] It is therefore possible to exploit lymphangiogenic mechanisms to restore a compromised lymphatic system. For instance, the lymphangiogenesis-inducing factor VEGF-C can be administered locally to recover lymphatic drainage. The administration of the soluble form of the human recombinant (hr)VEGF-C[106] or its localized viral vectors-mediated over-expression[107,108] resulted in growth of functional and mature lymphatic vessels in animal models of peripheral lymphedema. Similarly, intracerebroventricular injections of adenoviral VEGF-C vector induced the growth of lymphatic capillaries in the meningeal compartment.[109] However, the functionality of these newly generated MLVs is uncertain, and further studies are required to decipher the ability of the new lymphatic vessels to clear parenchymal solutes and to control neuroimmunity.

6 | CONCLUDING REMARKS

Experimental evidence points to MLVs as a structural component of the CNS-lymphatic unit, impacting brain homeostasis, solute interstitial clearance, immune surveillance or inflammation. We have reviewed how alterations of the physiologic drainage of brain fluids could determine the accumulation of macromolecules within the brain parenchyma, resulting in the alteration of the extracellular ionic equilibrium, ultimately impacting neuronal excitability. The correct drainage of brain-derived antigens could be important for the allostasis of the neuroimmune cross-talk. We updated the hypothesis supporting the involvement of dcLNs in immune CNS surveillance and proposed that functional alterations of the MLVs (primary afferent vessels of the dcLNs) could result in autoimmune reactions. We suggested that a dysfunction of the CNS-lymphatic unit could be implicated in the pathophysiology of specific forms of epilepsy, as in situations where the primary cause is unknown (eg, Rasmussen encephalitis).

Moreover, functional of the CNS-lymphatic unit due to congenital defects or as a result of brain trauma, tumors, or infections could contribute to acquired or immune epilepsies. Addressing the dynamics of the CNS-lymphatic unit in the context of ictal activity could be important to disclose new therapeutic targets.

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DISCLOSURE

None of the authors has any conflict of interest to disclose. The authors confirm that have read the Journal's position on issues involved in ethical publication and affirm that this report is consistent with those guidelines.

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