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EDITORIAL

Lymphatic system and gut microbiota affect immunopathology of neuroinflammatory diseases, including multiple sclerosis, neuromyelitis optica and Alzheimer's disease

Infections with microbes, such as viruses and bacteria, result in damage of the peripheral nervous system or the central nervous system either by microbial replication in the nervous tissues (microbial pathology) or by uncontrolled immune responses (immunopathology). For example, as reviewed by Dr Kutsuna, Zika virus infection has been associated with two neurological complications: microcephaly and Guillain–Barré syndrome (GBS).[1] Microcephaly can be caused by direct microbial infection (viral replication) in the brain (viral pathology), while GBS is an antibody-mediated immunopathology (Zika virus induced-GBS might be heterogenous; a group of patients had a "parainfectious" onset, not a postinfectious onset typically seen in GBS).[2]

Two systems affect microbial pathology and immunopathology

Although a variety of factors can affect immunopathology triggered by microbial infections, two essential systems maintaining whole-body homeostasis have long been neglected in the field: the lymphatic system and microbiota. Although the lymphatic system and microbiota have been described in most medical textbooks of anatomy, immunology and microbiology, their roles in immunopathology associated with microbial infections had not been investigated until recently. Although involvement of the lymphatics and microbiota in microbial immunopathology is intuitively plausible, the reason why they have long been neglected by pathologists might due to their invisibility in routine hematoxylin–eosin-stained sections. While most mucosal microbes are washed out during tissue processing, lymphatics are either indistinguishable with blood vessels or invisible in traditional histology staining; lymphatic-specific immunohistochemistry and/or gene targeting have become available only recently.

Although regional lymph nodes have been used to study immune responses in health and diseases, lymphatic vessels have not been investigated by most immunologists. In medical education and clinical settings, only some disease conditions (e.g. filariasis, chylothorax, lymph node metastasis in cancer and lymph node swelling in infections) remind us of the presence of the lymphatic vessels. This is partly due to the fact that lymphadenectomy does not necessarily result in expected adverse effects, such as lymphedema and local infection. In many immunology experiments, even in the situation where some immunologists misunderstand that lymph nodes are connected with blood vessels, the misconception affects neither experimental results nor their interpretation in most cases.

Lymphatic system in immunopathology

Dr Al-Kofahi and colleagues from Dr J. Steven Alexander's group excellently reviewed the anatomy and functions of the lymphatic system, particularly in the gastrointestinal tract, the heart and the central nervous system.[3] During acute microbial infection, lymphatic flow is increased in the infected tissue, limiting edema as well as providing more soluble microbial antigens and antigen-laden dendritic cells into the regional lymph nodes for antigen-presentation. The authors have proposed that dysfunction of the lymphatics leads to the persistence of immune cells and mediators in the tissue (Fig. 1). This leads to chronic inflammation and tissue damage by immunopathology, whereas the lymphostasis might confine pathogens locally, limiting systemic spread of the microbes. This theory is based on experimental findings by Dr Alexander's group and others; for example, Al-Kofahi et al. have shown that dysfunction of lymphatic vessels can contribute to prolonged inflammation in inflammatory bowel diseases, such as Crohn's disease and ulcerative colitis, as well as myocarditis using a mouse model induced with Theiler's murine encephalomyelitis virus.[4] Furthermore, Dr Alexander's group previously showed that downregulation of a set of serum lymphatic markers, such as prospero homeobox 1 and angiopoietin-2, might be characteristic of secondary progressive multiple sclerosis (MS); here, lymphatic-specific molecules can be biomarkers to distinguish between secondary progressive MS and relapsing–remitting MS.[5]

Gut microbiota in immunopathology and neuropathology

In biomedical education, the gut microbiota has also been taught in association with only limited subjects, such as production of vitamin K and *Clostridium difficile*-induced pseudomembranous colitis. Recently, however, the gut microbiota has been investigated in a variety of health and disease conditions, where there seems to be a myth that "good bacteria" are beneficial for everything from aging, obesity and infections, to brain diseases, whereas "bad bacteria" are bad for anything. As reviewed by Dr Park et al., this is likely not the case in MS and two "bad bacteria," *Clostridium perfringens* type A and *Helicobacter pylori*.[6] While the former causes food poisoning and gas gangrene, and the latter is associated with gastritis, gastric cancer and idiopathic thrombocytopenic purpura, the presence of both bacteria are lower in MS patients than controls. In contrast, *H. pylori* is associated with exacerbation of neuromyelitis optica (NMO) and Alzheimer's disease (AD).

The above contrasting roles of *H. pylori* can be explained when comparing and contrasting: (i) antimicrobial immune responses versus immunopathology; and (ii) cellular immunity/pro-inflammatory T helper (Th)1 and Th17 cells versus humoral immunity/anti-inflammatory Th2 and regulatory T cells. For eradication of *H. pylori*, cellular immunity plays a key role. However, when hosts failed to eradicate *H. pylori*, to prevent uncontrolled pro-inflammatory responses that can cause gastritis, the immune response is skewed to anti-inflammatory, which protects immunopathology at the expense of bacterial persistence. This might be the case in asymptomatic carriers of *H. pylori*. Here, suppression of Th1/Th17 is protective against MS, whereas enhanced humoral responses play no role in eradicating *H. pylori*, but might lead to antibody-mediated autoimmune diseases, such as idiopathic

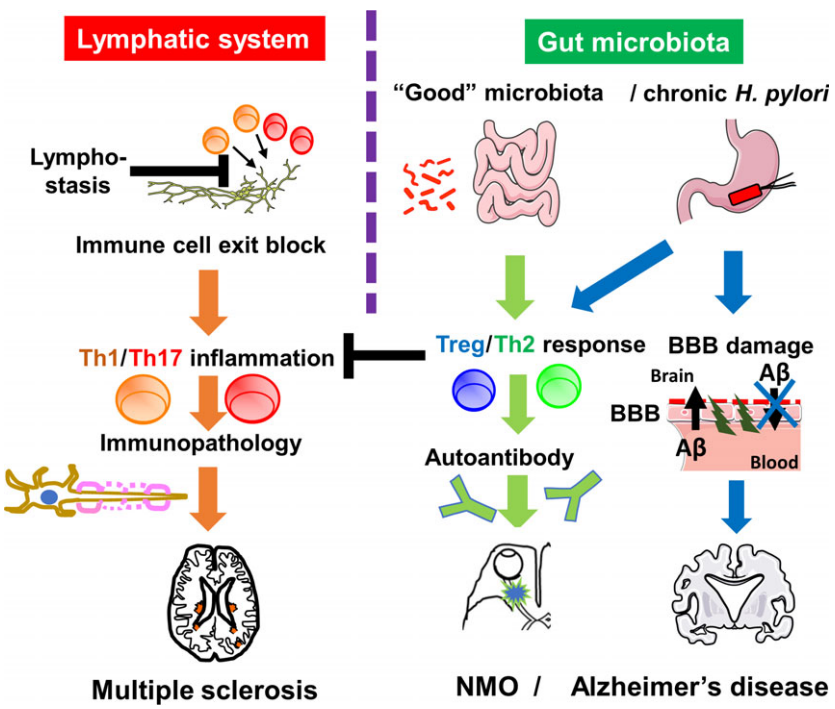

Figure 1 Lymphatic system and gut microbiota in neuroinflammatory diseases. (Left) Lymphatic vessels drain immune cells and edema fluid from inflamed tissues into regional lymph nodes. Dysfunction of lymphatics blocks the exit of inflammatory cells from the tissue, leading to chronic inflammation and/or immunopathology, such as multiple sclerosis, in the brain (where the presence of the lymphatic system has been proposed recently). (Right) "Good bacteria" in the gut microbiota as well as chronic *Helicobacter pylori* infection change the T helper (Th) cell subset balance toward regulatory T cells (Tregs)/Th2 responses. Tregs and Th2 cells can suppress pro-inflammatory Th1/Th17 inflammation, preventing immune-mediated tissue damage; for example, gastritis in the stomach and multiple sclerosis in the brain. In contrast, increased Th2 cytokines can enhance autoantibody production, exacerbating antibody-mediated disease, including neuromyelitis optica (NMO). As the antibody against *H. pylori* has no role in bacterial clearance, the suppression of anti-bacterial Th1/Th17 immunity leads to *H. pylori* persistence, which has been associated with blood–brain barrier (BBB) dysfunction. BBB dysfunction can not only increase the accumulation of amyloid-β (Aβ) from the periphery, but can also decrease the clearance of Aβ from the brain, contributing to the progression of Alzheimer's disease.

thrombocytopenic purpura and NMO. Suppression of pro-inflammatory T cells in chronic *H. pylori* infection might explain a lack of T-cell infiltration in the brain lesions of AD, despite activation of resident innate cells, including microglia, whereas *H. pylori* infection can also contribute to dysfunction of the blood–brain barrier observed in AD.

"10 pitfalls of microbiota studies"

Oversimplification and/or overestimation of the roles of the gut bacterial community (bacteriome) in microbiota studies can be explained by "10 pitfalls of microbiome studies" proposed by Dr Park et al.: (i) the presence of fungal (mycobiome) and viral communities (virome); (ii) microbial taxonomy/classification; (iii) fecal bacteria ratio underrepresentation; (iv) "dysbiosis" as the outcome; (v) discrepancy with primary immunodeficiency diseases; (vi) age, sex and country; (vii) good bacteria is not always good; (viii) antibiotics affect systemic microbiota and immunity; (ix) fecal microbiome transplantation methodology and safety; and (x) tailor-made therapy.[6] The proposal is useful to plan and evaluate microbiota studies, clinically and experimentally.

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Conflict of interest

None declared.

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Ikuo Tsunoda
Department of Microbiology, Kindai University Faculty of Medicine, Osakasayama, Osaka, Japan

