The gut microbiome molecular mimicry piece in the multiple sclerosis puzzle

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The etiological complexity of multiple sclerosis, an immune-mediated, neurodegenerative disease with multifactorial etiology is still elusive because of an incomplete understanding of the complex synergy between contributing factors such as genetic susceptibility and aberrant immune response. Recently, the disease phenotypes have also been shown to be associated with dysbiosis of the gut microbiome, a dynamic reservoir of billions of microbes, their proteins and metabolites capable of mimicking the autoantigens. Microbial factors could potentially trigger the neuroinflammation and symptoms of MS. In this perspective article, we discussed how microbial molecules resulting from a leaky gut might mimic a host’s autoantigen, potentially contributing to the disease disequilibrium. It further highlights the importance of targeting the gut microbiome for alternate therapeutic options for the treatment of MS.

KEYWORDS
multiple sclerosis, gut microbiome, molecular mimicry, microbial metabolites, aberrant immune response, leaky gut, HLA genes, Epstein-Barr virus

Introduction

Multiple sclerosis (MS) is an immune-mediated, chronic, debilitating, demyelinating neurodegenerative disease. It is estimated to be affecting 2.8 million people worldwide in 2020, a 30% increase since 2013, and in the United States, MS patients number roughly 400,000 people (1–3). MS is a complex autoimmune disease in which the combined roles of genetic susceptibility and aberrant immune response are largely established. However, an understanding of how environmental or microbial factors, infectious or not, initiate, trigger, or maintain the different stages or phenotypes of MS remains to be established. Despite the availability of several therapeutic treatments over the last few years, mainly to stall the disease’s progression, still there is no cure for MS. Failure to develop a cure stems partially from an incomplete understanding of MS pathophysiology and the factors that trigger its symptoms. This article attempts to piece together these pieces of the MS jigsaw puzzle.
The pathogenesis of MS is considered secondary to the autoreactive lymphocytes entering the central nervous system and causing neuroinflammation and subsequent demyelination of the axons (4, 5). The hallmark of this disease is axon demyelination where muscle weakness, blurred vision, and malfunctioned urinary and gastrointestinal systems are its visible consequences (6). Clinically, the disease has four phenotypes: clinically isolated syndrome (CIS), relapsing remitting MS (RRMS), secondary progressive MS (SPMS), and primary-progressive MS (PPMS), the latter of which can be further classified into active and inactive/remission (7). A majority of patients (85%) suffer from RRMS, but others can convert to a progressive course with an accumulation of disabilities leading to the loss of mobility (8). PPMS is a steady progressive form of MS from its onset (9). The heterogeneity of the MS symptoms, its undefined pathogenesis, unknown triggering factors, and its different phenotypes pose a challenge for the diagnosis and treatment of this debilitating disease.

Genetics of MS

There is a strong genetic component to MS as evidenced by its higher concordance rate in monozygotic twins (25-30%). However, it has a lower rate in dizygotic twins (3-7%) and first-degree relatives (3%). Several familial studies supported the presence of a genetic component in MS, but the rapid decrease in risk from monozygotic twins to other family members illustrates a polygenic nature (10–12). The polygenic risk of MS is both concentrated and dispersed consistent with its spectrum of symptoms. Indeed, no single genetic variant can predict effectively the susceptibility of patients (13). Genetic risk (11) increases with the increasing number of risk alleles in a patient (14). Many studies have investigated the genetic architecture of MS disease; however, the results were not always replicated. This is consistent with the common disease common variant theory which implies that common diseases are caused by hundreds of common variants, each of which has a small effect (11, 15). The most definitive genetic association to MS disease is the human leukocyte antigen genes (HLA) class II or more narrowly DRBI*15:01 allele (11, 16, 17) on chromosome 6p21 (18) which links the adaptive immune system to MS disease pathogenesis. However, the DRB*15:01 allele has a moderate effect in predicting disease susceptibility (19) and seems to be associated with both the disease’s early onset in general and with a high disease rate in women (17, 18). Other studies reported an association between HLA-DRBI*04 and PPMS clinical phenotype (20, 21). However, this association with PPMS has not been confirmed by other studies (18, 22, 23). Other HLA alleles such as HLA-A*02:01 and HLA-DRBI*11 showed a protective role for MS (24, 25). Roughly, 200 genetic loci residing in the non-HLA polymorphic genes have also been implicated, accounting for 20% of the MS genetic associations. The majority of these non-HLA alleles are related to immunogenic pathways such as interleukin 7 receptor (IL7R) (26), interleukin 2 receptor (IL2RA) (27), TAGAP (28), and vitamin D receptor (VDR) (29). These variants associated with non-HLA genes affect the function of their respective genes, such as how the creation of soluble protein of both IL2 and IL7 genes leads to downstream signaling inhibition (11, 30).

Immunology of MS

MS is a disease of aberrant immune signaling pathways modulated by hundreds of genetic variants regulating immune cells. In terms of immuno-pathophysiology, MS is linked to the imbalance between T regulatory (Treg), and T helper (Th) cells (31). Th cells usually recognize the presented peptides by major histocompatibility complex (MHC) class II on the antigen presenting cells (32). Th cells differentiate into Th1, Th2, and Th17, and these differentiated cells secrete different cytokines such as IL17, TNF-α, IFN-γ, IL-4 and CCL4 (33). Treg cells are the only self-reactive T-cells to maintain immune homeostasis. They normally express interleukin (IL2) receptor α chain (CD25) and its function depends on the transcription factor FOXP3 (34). Treg cells produce inhibitory cytokines such as IL10 and transforming growth factor (TGF)-β. These cytokines inhibit the proliferation of inflammatory Th cells and promote immune tolerance which protects against demyelination (35).

MS patients have abundant inflammatory Th17 cells and low levels of the anti-inflammatory Treg cells. This immune imbalance enhances the infiltration of monocytes and macrophages (35, 36) in central nervous system (CNS) which increases the reactive oxygen species, triggers the lesion formation, disturbs the tight junctions of the blood brain barrier (BBB), and attracts more monocytes towards the BBB. This eventually leads to phagocytosis of the myelin and the loss of the action potential (37, 38). In summary, overactive T cells start the RRMS and the progressive state is maintained by the infiltrating monocytes and macrophages in the CNS (39). Interestingly, Venken et al. (2006) showed that there was no difference in Tregs count between RRMS and secondary progressive, the two phenotypes of MS disease (40). Lately, the role of B cells was studied for MS pathology where the cells were displaying a proinflammatory phenotype in patients, exacerbating the Th17 cell response (41–43). The host genetics comprises one piece of the puzzle, but leaves questions regarding possible links between genetic susceptibility, MS immune-pathophysiology, microbial agents or their products from gut and/or outside environments.

These questions linger despite several studies reporting associations or interactions between the host’s genetic susceptibility and environmental factors, such as smoking (44), obesity (45), heavy metal poisoning (46), low vitamin D, high salt intake (33, 47), and Epstein-Barr virus infection (48) - all have a reported association with MS. Of particular note, it has been suggested that a related low exposure to infectious agents in childhood helps prime the immune system (49). An emerging
theme for microbial triggers comes from the rich microbial reservoir of the human gut. Indeed, the importance of human gut microbiome cannot be overstated as any one or more of its thousands of viruses or bacterial species could potentially house one of MS’s triggers. This enormous repertoire of microbial factors and metabolites has been shown to possess a path connecting the gut-brain nexus (50). Furthermore, the gut microbiome dysbiosis has demonstrated associations with other complex autoimmune and neurological diseases such as Alzheimer’s disease, Parkinson’s, and autism spectrum disorder (51–53). If microbial triggers from gut microbiome, especially its microbial peptides, can be a source of molecular mimicry with the myelin sheath, then this could potentially explain, in part, how the MS is triggered in genetically susceptible individuals.

**Gut microbiome and MS**

The search for an elusive, consistent infectious agent with a triggering or contributing role in causing MS has led to a much-needed investigation of dysbiosis in human gut microbiota. The plausibility of such an agent lies in the availability of billions of microbes whose proteins or metabolites could interact with a host’s signaling pathways to determine the overall health of a person. Certainly, gut microbe(s) could trigger the neuroinflammation and the symptoms of MS. Indeed, numerous studies have reported dysbiosis of the gut microbiome in MS cohorts capable of either a triggering or a facilitating infectious agent mechanism for developing MS in genetically susceptible individuals compared to non-MS subjects (54–57). Moreover, a relapsing-remitting, mouse MS model (experimental autoimmune encephalomyelitis (EAE)) suggests a triggering of MS disease upon exposure to commensal, but not pathogenic, gut bacterial flora (58). Even individual bacterium from the gut microbiome (specifically, segmented filamentous bacteria) was capable of inducing EAE in germ-free mice (55). Consistent with a role for gut microbiota in MS, treatment of the EAE experimental mice with non-absorbing oral antibiotics, kanamycin, colistin, and vancomycin for reducing the bacterial load in the gut led to the improvement of EAE development in those mice (33). The administration of antimicrobials and resulting changes in the gut microbiota further lead to the reduction of proinflammatory cytokines and mesenteric Th17 cells (59), which has been reported to play a role in MS disease development. Despite this evidence, MS risk cannot be efficiently reduced by antibiotics alone (60); many pieces of the gut microbiome puzzle related to MS disease are still missing.

In MS patients, gut microbiome dysbiosis has been reported at different taxa level (61–63) (Table 1). Understanding these perturbations in MS patient’s vis-à-vis non-MS healthy controls from a similar environment can shed light on the function of one or more taxa in MS. For instance, *Methanobrevibacter* has been reported to be in higher relative abundance in the gut microbiome of MS patients as displayed in Table 1 (56) as well as in patients with other inflammatory autoimmune diseases (73). It has been proposed that *Methanobrevibacter* may play a complex role, activating the dendritic cells and recruiting other inflammatory cells, as well as producing methane gas, which delays the gut transit time leading to constipation, one of the MS symptoms (63, 74).

**TABLE 1** Reported changes in gut microbiome of MS patients cohorts.

| Organisms | Increase | Decrease | References |
|-----------|----------|----------|------------|
| Phylum    | Firmicutes | Bacteroidetes | (56, 64, 65) |
|           | Proteobacteria | Ascomycota* |          |
|           | Euryarchaeota |          |          |
|           | Verrucomicrobia |          |          |
|           | *Banditomycota* |          |          |
| Family    | Lachnospiraceae | Butyrivimonas | (54, 56, 57, 64, 65, 67–71) |
|           | Desulfovibrionaceae |         |          |
| Genus     | *Methanobrevibacter Akkermansia* | Clostridia | (56, 64, 65) |
|           | Streptococcus | Preveotella |          |
|           | Acestobacter | Bacteroides |          |
|           | Blautia | Lactobacillus |          |
|           | Bifidobacterium | Sutterella |          |
|           | Adlercreutzia | Collinsella |          |
|           | Flavobacterium | Caprobacillus |          |
|           | Pseudomonas | Anaerostipes |          |
|           | Mycoplana |         |          |
|           | Sacciachromyces* |       |          |
|           | Aspergillus | Prepostella |          |
|           | Candida* | Bacteroides fragilis | (57, 62, 63, 72) |
|           | Epiconicum* | Parabacteroides distasonis |          |

*Fungi.
the other hand, the depletion of _Bacteroides fragilis_ and _Clostridoides_ in gut microbiota of MS patients as shown in Table 1 suggests a protective function in healthy individuals (63). For instance, the polysaccharide A of _B. fragilis_ protected against axon demyelination in mice (75), and the _Clostridoides_ produce butyrate, which increases the Treg differentiation favoring the anti-inflammatory state (54, 76). Then, why not there is a MS gut microbiome biosignature identified from the dysbiosis from MS patients? It is because not all studies have observed major changes between the gut microbiome in MS and healthy control except that _Akkermansia_ and _Methanobrevibacter_ showed consistent increase in MS patients while _Prevotella_ and _Bacteroides_ were reduced in MS patients (67). Furthermore, Knight et al. (2014) reported that predicting discrete clusters as disease biomarkers is not highly effective because the human microbiome is a continuous gradient of relative abundance of species. Medications. In addition to bacterial dysbiosis, changes in the relative abundance of specific fungi have also been reported (Table 1). In summary, detection of precise microbial signature for MS is still elusive and needs to be evaluated longitudinally during the disease course and from multi-ethnic patients living in different environmental conditions.

### Microbiome metabolites and MS

Could microbiome-produced metabolites be identified as biomarkers in MS? Lower concentration of lipid 654 produced by _Bacteroidetes_ species is being explored as a biomarker as this compound showed decrease in concentration in both MS and Alzheimer patients in comparison to healthy controls (78). Jangi et al. (2016) reported an increase in breath methane in MS patients, which may be corresponding to the increase of _Methanobrevibacter_ in gut microbiome (56). During dysbiosis, the microbiota either sequester important nutrients from the host or alter the production of metabolites. The former mechanism is shown with the taxa _Desulfovibrionaceae_, which sequester cysteine. This amino acid is mostly used for the synthesis of glutathione, a tripeptide of cysteine and glutamate, protect against reactive oxygen species in the CNS (37). As a consequence, a low concentration of glutathione has been encountered in MS patients in comparison to healthy individuals (79). The latter mechanism for dysbiosis is the altered metabolite production in favor of low beneficial compounds or high detrimental ones. Another example of beneficial metabolites are the short chain fatty acids (SCFAs) like propionate. Collectively, the amount and type of SCFAs produced depend on the microbiota composition and the substrates available (80). SCFAs are produced by a wide variety of gut bacterial species such as _Bacteroides, Bifidobacterium, Lactobacillus, and Clostridium_ (35). Bacteroidetes produce acetate and propionate, while firmicutes produce butyrate (33). Many studies pointed out the beneficial effects of SCFA for MS. SCFAs maintain the integrity of both the intestinal barrier through increasing the expression of tight junction proteins (35, 76) and the BBB (81). Moreover, SCFAs increase the differentiation of the anti-inflammatory Treg, which can relieve the axonal damage to some extent (35). Additionally, SCFAs could inhibit histone deacetylases in a concentration-dependent manner as it helps maintain immune homeostasis (82). Furthermore, propionate has been suggested as a potent immunomodulatory for MS patients which decreases the relapse episodes and brain atrophy because it increases Treg and decreases Th17 cells (83). In MS patients, both stool and plasma revealed a decrease in SCFAs concentration which implies a reduction in SCFAs-producing bacteria of the gut microbiome (7, 54, 68). Additionally, the higher level of _Prevotella_ in MS patients undergoing disease-modifying therapy was accompanied by higher concentration of butyrate (84). In conclusion, SCFAs are crucial for establishing an anti-inflammatory state, a state usually deficient in MS patients.

Gut microbiota such as firmicutes, _Lactobacillus_, and _Enterobacteriaceae_ (to name a few) can metabolize aromatic amino acids such as tyrosine, phenylalanine, and tryptophan. Mostly the genera in phylum firmicutes can produce P-cresol from tyrosine and phenolic compounds from phenylalanine (35). Through the kynurenine pathway, they can also metabolize tryptophan into different metabolites known as “TRYP-6.” These metabolites can be kynurenine, quinolinolate, indole, indole acetic acid (IAA), indole propionic acid (IPA) and tryptamine (35). Some of these compounds, namely indole derivatives, can cross the BBB and act on aryl hydrogen receptor (AHR), further decreasing reactive oxygen species and controlling the neuroinflammation processes (85). Moreover, other tryptophan metabolites, after binding to AHR receptors, exhibit anti-inflammatory effect on astrocytes. The astrocytes are the main component of neurovascular units, which preserve the integrity of BBB under normal conditions (86, 87). However in MS, it was observed that astrocytes upregulate 4-galactosyltransferase enzymes, which boost inflammation of the CNS (88). Thus, the anti-inflammatory effect of tryptophan metabolites helps delay the progression of MS disease (89). Other studies reported variations in the kynurenine level in different stages of MS disease (active/inactive) and clinical phenotypes (90). Considering all these complex interactions, the challenge remains to fully understand the exact mechanism of tryptophan metabolism in MS. Other metabolites such as _Bacillus_ derived poly-gamma-glutamic acid favor the imbalance of Th cells towards Th1 rather than Th17 cells (91). Phytoestrogens metabolites have anti-inflammatory effect and a significant amelioration of EAE in mouse model (92). Bacteria metabolizing phytoestrogens such as _Prevotella_ and _Aldercreutzia_ are decreased in MS patients, which would favor the inflammatory state in the MS patients. Thus, the metabolites produced by gut microbiome could affect the intestinal barrier integrity, the immune tolerance, and the neuroinflammation encountered in the brain.
Could microbial molecular mimicry trigger MS?

Microbes-derived molecular mimicry emanating from the homology between microbial- and human-derived antigens has been known to induce autoimmunity (93–95). In rheumatic fever, molecular mimicry is displayed between M proteins of *Streptococcus pyogenes* and the cardiac myosin (96). Mechanistically, a cross reactivity could happen when T cells recognize antigens through major histocompatibility complex (MHC) class II via a sequence of 8-10 amino acids (97), a small enough number of amino acids that can be shared between microbial and host peptides and/or due to the similarity of anchor proteins that bind specifically to MHC molecules, a phenomenon known as polyspecificity. The MHC usually binds to these anchor amino acids residues but has flexibility in the remaining residues, a flexibility that increases the responsiveness of MHC class to variety of pathogens and xenobiotics (98). In the case of MS, amino acid changes encountered in the MHC binding peptides affect those antigens binding to the HLA-DRB1 (11). Thus, the molecular mimicry of bacterial peptides to the myelin protein are worth experimental exploration. Hundreds of thousands of proteins from thousands of bacterial species and their serovars from gut microbiota could potentially produce autoantigenic peptides in genetically susceptible individuals or under certain immunodeficient conditions. For instance, different bacterial species of the gut microbiome can induce imbalance in Th and Treg cells (55, 58). Moreover, for T cells to attack the myelin sheath inside the CNS, they should be activated peripherally by maybe a bacterial peptide (99). In the brain of MS patients, it has been reported that bacterial peptidoglycan in antigen-presenting cells suggests a triggering of the pathophysiology through bacterial products (100). Similarity between bacterial products and autoantigens capable of inducing MS has also been studied. Mostly, the primary candidates autoantigens were myelin basic protein (MBP), proteolipid protein, myelin-associated glycoprotein, and myelin oligodendrocyte glycoprotein (101). However, MBP was thoroughly investigated because of its induction of MS symptoms in mice and genetically susceptible primates in presence of an adjuvant (102, 103). The MBP sequence was divided into the tryptophan, midpeptide, and hyperacute regions (103, 104). Some proteins of *Bacteroides* and *Bifidobacterium* species showed some similarity with different MBP regions (103). Interestingly, even the adjuvant required for induction of the immune response is found in bacteria like *N*-acetylglucosamine 1-dipeptide (103). Still the question remains: why does MS happen in certain patients when these antigens and adjuvants are available in every human gut?

Furthermore, CD4 T cells can be activated by another candidate, GDP-L-fucose synthase, in a manner quite similar to the myelin sheath especially in HLA-DRB3* positive patients. Fucose synthase peptides have homology with bacterial fucose synthase of *Akkermansia*, and *Prevotella* (99). Not only with specific bacterium, cross reactivity between the myelin sheath and the Epstein-Barr virus nuclear antigen 1 has also been shown suggesting Epstein-Barr virus as a possible inducer of MS especially in genetically susceptible people (48, 105, 106). Surprisingly, when *Acinetobacter* and *Pseudomonas* act as infectious agents they use mucuno-decarboxylase enzyme, which shares a similar sequence with myelin protein (107). These in-silico and experimental studies present a plausible relationship between MS autoimmune targets and molecular mimicry emanating from microbes, possibly gut microbes. The molecular mimicry emanating from the gut microbiome has also been investigated in another CNS demyelinating disease, neuromyelitis optica (108), an autoimmune disease causing optic neuritis and active myelitis. In neuromyelitis optica, autoantibodies are produced against the astrocyte water channel protein aquaporin 4 (AQP4) (109). Interestingly the commensal bacterium, *Clostridium perfringens* showed increased relative abundance in the patients of neuromyelitis optica (110) than the healthy controls and the adenosine triphosphate-binding cassette (ABC) transporter permease sequences of *C. perfringens* shared homology with T cell epitope with the Aquaporin 4 with the possibility of mimicry from the gut (108). However, it should be noted that induction of autoimmune disease by molecular mimicry is complex and bacterial or viral peptide(s) acting alone are expected to be insufficient to initiate a complex disease like MS. There ought to be accompanying host genetic susceptibility linked with aberrant immune response triggered by one or more microbial antigens or molecules. And just like the dispersed genetic variants associated host genetic susceptibility, peptides from more than one microbe could be capable of mimicry in different MS patients.

The leaky gut and translocation of gut microbial peptides

The MS-gut microbiome association operates with an underlying assumption that one or more microbial triggers from the gut can cross the intestinal barrier due to a phenomenon called leaky gut which allows dissemination of the bacteria to distal organs and enables immune system exposure to such bacteria (76). Disruption of the intestinal barrier could be due to dysbiosis. Indeed, *Akkermansia* species could feed on the mucin layer in the mucous layer of the intestine, exposing the gut microbiome to systemic circulation (56, 111). Alternatively, MS patients exhibit high secretion of INF-γ which would stimulate the differentiation of Th cells to Th17 cells. Th17 cells produce IL17, which in combination with INF-γ would reorganize the intestinal tight junctions leading to a compromised barrier (112). Again, this barrier disruption would expose the already hidden gut microbiome to the immune system. A disturbed intestinal barrier is usually evaluated by the lactulose/mannitol permeability test, and
in case of MS patients, abnormal permeability has been reported in about 73% of MS patients (113). In animal models, the severity of EAE increased with compromised intestinal barrier but improved with treatment with *Escherichia coli* strain Nissle 1917 as a probiotic (114) suggesting ameliorating intestinal barrier could be one of the potential goals for innovative treatment to attenuate the MS disease.

**Immunologic tolerance**

The gut microbiota are important for achieving immunologic tolerance and establishing good immune response against pathogens. This was partly confirmed by the upregulation of CD +4 T cells and development of gut-associated lymphoid tissues in the underdeveloped immune system of germ-free mice after the transfer of commensal bacteria (115). The gut immune cells do not respond to commensal bacteria because of a special phenotype of macrophage called “inflammation anergy” (116). Moreover, any inflammatory response in the gut is usually suppressed by cytokines produced by lymphocytes in the gut-associated lymphoid tissues (86). As mentioned before, the imbalance between Th17 and Treg is one of the studied causes for MS disease. Interestingly, Th17 and Treg cells are found in the intestine where gut microbiome is assumed to enhance the differentiation of Th17 (117). The germ-free mice do not have Th17 except after induction of microbial colonization (118). On the other hand, no significant difference in Treg counts between MS patients and healthy controls was observed (40, 119). However, the Tregs showed lowered antinflammatory function in MS patients through reduction in IL10 production in germ free mice transplanted with fecal samples of MS patients in comparison to that of healthy control (57). The colonic Treg cells also depend on some microbial signals for efficient function (120). For instance, CD41FOXP31 Treg cells are upregulated by *Clostridia* (*Clostridoides*) strains (121) and by *P. histicola* in mice which led to downregulation of IL-17 and IFNγ (62). Moreover, the gut microbiota do not affect only other immune cells numbers but their function. For example, the capacity to kill pathogens of neutrophils was reduced in germ free mice (122).

![Figure 1](https://example.com/figure1.png)

**FIGURE 1**

A potential mechanism to link the role of gut microbiome to trigger and maintain MS symptoms in genetically susceptible patients. Specific microbes (e.g., *Methanobrevibacter* and *Akkermansia*), microbial peptides (RNA polymerase B (103)), or metabolites (short chain fatty acids and phytoestrogens) from the gut could enter the circulatory system due to leaky gut and induce aberrant immune response, particularly the imbalance of Th and Treg cells. The T cells imbalance where Th17 cells outnumber Tregs impairing immune tolerance and subsequently lead to inflammation and MS lesions in the brain.
Discussion

As a heterogeneous, complicated, immune-mediated disease, MS has complex etiology. Several studies have provided evidence that the gut microbiome plays a role in MS pathogenesis, therefore it could help in its diagnosis, and targeted therapeutic intervention with further research. In this article, we highlighted the current understanding of the potential roles of microbial agents or their products from the gut microbiota in MS as displayed in Figure 1. We speculated that either molecular mimicry or microbial metabolites in the gut microbiome could contribute to the known aberrant immune response of Th17 and Treg imbalances. Many studies have reported compositional alteration of different microbial species of gut microbiome in MS. Indeed, the gut microbiome with its rich microbial reservoir and their protein could be a source for faulty cross-recognition of Th cells, leading to this aberrant immune response. Moreover, the plentiful anti-inflammatory metabolites produced by the gut microbiome suffer from a reduction in MS diseases potentiating the inflammatory state encountered in this disease. With the puzzle thus far presented, there is a need to further understand these complex relationships between the genetics, immune response and the gut microbiome to understand the pathophysiology of the disease and develop better therapeutic options for this debilitating disease.

Author contributions

NSE: conceptualization, writing the original draft article, and final editing. PA: review and editing, VRB: conceptualization and review, SKS: conceptualization, writing, reviewing, and final editing. All authors contributed to the article and approved the submitted version.

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Conflict of interest

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References

1. Walton C, King R, Rightmear L, Kaye W, Leray E, Marrie RA, et al. Raising prevalence of multiple sclerosis worldwide: Insights from the atlas of MS, third edition. Mult Scler Houndmills Basingstoke Engl (2020) 26(14):1816–21. doi: 10.1177/1352458520970841
2. Dilkohorskasak P, Valuck RJ, Nair KV, Corboy JR, Allen RR, Campbell JD. Multiple sclerosis prevalence in the united states commercially insured population. Neurology (2016) 86(11):1014–21. doi: 10.1212/ WNL.0000000000002469
3. Paul A, Comabella M, Gandhi R. Biomarkers in multiple sclerosis. Cold Spring Harb Perspect Med (2019) 9(3):a029058. doi: 10.1101/cshperspect.a029058
4. Compton A, Coles A. Multiple sclerosis. Lancet (2008) 372(9648):1502–17. doi: 10.1016/S0140-6736(08)61620-7
5. Trapp RD, New KA. Multiple sclerosis: An immune or neurodegenerative disorder? Annu Rev Neurosci (2008) 31(1):247–69. doi: 10.1146/ annurev.neuro.30.051606.094313
6. Kohl HM, Castillo AR, Ochoa-Reparaz J. The microbiome as a therapeutic target for multiple sclerosis: Can genetically engineered probiotics treat the disease? Dis Basel Switz (2020) 8(3):33. doi: 10.3390/disease8030033
7. Park J, Wang Q, Wu Q, Mao-Draayer Y, Kim CH. Bidirectional regulatory potentials of short-chain fatty acids and their G-protein-coupled receptors in autoimmune neuroinflammation. Sci Rep (2019) 9(1):8837–7. doi: 10.1038/s41598-019-45311-y
8. Fissner S, Gold R. Progressive multiple sclerosis: latest therapeutic developments and future directions. Ther Adv Neurol Disord (2019) 12:175628441987323. doi: 10.1177/175628441987323
9. Mills EA, Beggy JA, Fisher C, Mao-Draayer Y. Impact of trial design and patient heterogeneity on the identification of clinically effective therapies for progressive MS. Mult Scler Houndmills Basingstoke Engl (2018) 24(14):1795–807. doi: 10.1177/1352458518800800
10. Dyment DA, Sadovnick AD, Ebers GC. Genetics of multiple sclerosis. Hum Mol Genet (1997) 6(10):1693–8. doi: 10.1093/hmg/6.10.1693
11. Patsopoulos NA. Genetics of multiple sclerosis: An overview and new directions. Cold Spring Harb Perspect Med (2018) 8(7):a028951. doi: 10.1101/ cshperspect.a028951
12. International Multiple Sclerosis Genetics Consortium. Multiple sclerosis genomic map implicates peripheral immune cells and microbiota in susceptibility. Science (2019) 365(6460):eaav7788. doi: 10.1126/science.aav7788
13. Sawcer S, Franklin RM, Ban M. Multiple sclerosis genetics. Lancet Neurol (2014) 13(7):700–9. doi: 10.1016/S1474-4422(14)70041-9
Primarily chronic progressive and relapsing/remitting multiple sclerosis: two outcome of MS.

Immune-related loci identified by genome-wide association studies are associated with gut microbiota at immune, metabolomic, and neuroactive level. 33. Haase SL, The chart locates | beating MS: a story of b cells, with turns and twists. Mult Scler 2018;24(11):1293–305.

The role of gut microbiota with multiple sclerosis. 34. Lacki SJ, Kucey JM, Kowalski JR, et al. Th1, Th17, and Th9 effector cells induce experimental autoimmune encephalomyelitis with disease progression. J Neuroinflammation (2016) 13(1):74.

It is associated with gut microbiota at immune, metabolomic, and neuroactive level. 35. Riccio P, Rossano R. Diet, gut microbiota, and vitamins d + a in multiple sclerosis. Mult Scler 2016;22(9):1167–74.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis. Frontiers in Immunology (2017) 8:2164–6.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The role of gut microbiota with multiple sclerosis. 36. Kastow BJ, Baecher-Allan C. Effector T cells in multiple sclerosis.

Altered expression of the gut microbiome in multiple sclerosis: a systematic review and meta-analysis. Steroids (2020) 158:108615. doi:10.1016/j.steroids.2020.108615.

The gut microbiome and multiple sclerosis. 37. Dopkins N, Nagarkatti PS, Nagarkatti M. The role of gut microbiota with multiple sclerosis.

The gut microbiome and multiple sclerosis. 38. Nalbandian L, Frouin F, Scher J, et al. Diet, gut microbiota, and vitamins d + a in multiple sclerosis. J Neuroinflammation (2016) 13(1):74.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The role of gut microbiota with multiple sclerosis. 39. Lim CK, Bilgin A, Lovejoy DB, Tan V, Bustamante S, Taylor BV, et al. Kynurenic pathway metabolomics predict and provides mechanistic insight into multiple sclerosis progression. Sci Rep (2017) 7:44173–3. doi:10.1038/srep44173.

The role of gut microbiota with multiple sclerosis. 40. Venken K, Hellings N, Hensken K, Rummens JL, Medar O, D’hooghe MB, et al. Secondary progression in contrast to relapsing-remitting multiple sclerosis patients show a normal CD4+CD25+ regulatory T cell population. J Neuroimmunol (2009) 214(1–2):136–46.

The role of gut microbiota with multiple sclerosis. 41. Bar-Or A, Fawaz L, Fan B, Darlington PJ, Rüeger A, Ghoraby C, et al. Abnormal b-cell cytokine responses a trigger of t-cell-mediated disease in MS? Ann Neurol (2010) 67(4):452–61. doi:10.1002/ana.21939.

The role of gut microbiota with multiple sclerosis. 42. Hauser SL. The chart locates | beating MS: a story of b cells, with turns and twists. Mult Scler 2018;24(11):1293–305.

The role of gut microbiota with multiple sclerosis. 43. Riccio P, Rossano R. Diet, gut microbiota, and vitamins d + a in multiple sclerosis. Neurotheraphy (2017) 15:1–75.

The role of gut microbiota with multiple sclerosis. 44. Voogt NJ, Kooij TO, Kasper LH. The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The role of gut microbiota with multiple sclerosis. 45. Gianfrancesco MA, Barcellos LF. Obesity and multiple sclerosis susceptibility: A review. J Neuroinflammation (2016) 13(1):5.

The role of gut microbiota with multiple sclerosis. 46. Riccio P, Rossano R. Diet, gut microbiota, and vitamins d + a in multiple sclerosis. Neurotheraphy (2017) 15:1–75.

The role of gut microbiota with multiple sclerosis. 47. Zostawa J, Adamczyk J, Sowa P, Adamczyk-Sowa M. The influence of sodium on pathophysiology of multiple sclerosis. Neurosci Ethiopia (2018) 5(1):13–22. doi:10.1093/brain/awp070.

The role of gut microbiota with multiple sclerosis. 48. Ngirachuk DM. Smoking: effects on multiple sclerosis susceptibility and disease progression. Ther Adv Neurol Disord (2012) 5(1):13–22. doi:10.1177/175625156429694.

The role of gut microbiota with multiple sclerosis. 49. Wu Z, Wang L, Sun X. Parasite-derived proteins for the treatment of multiple sclerosis. Frontiers in Immunology (2011) 2:221.

The role of gut microbiota with multiple sclerosis. 50. Carabotti M, Scirocco A, Maselli MA, Severi C. The gut-brain axis: development, aging and disease. Frontiers in Neurology (2020) 11:5.

The role of gut microbiota with multiple sclerosis. 51. Kastow BJ, Baecher-Allan C. Effector T cells in multiple sclerosis.

The role of gut microbiota with multiple sclerosis. 52. Garcia-Gutierrez E, Narbad A, Rodriguez JM. Autism spectrum disorder associated with gut microbiota at immune, metabolomic, and neuroactive level. Front Neurosci (2020) 14:578666. doi:10.3389/fnins.2020.578666.

The role of gut microbiota with multiple sclerosis. 53. Vogt NM, Kerby RL, Dill-McFarland KA, Harding SJ, Merluzzi AP, Johnson SC. The gut microbiome and multiple sclerosis: Cold Spring Harb Perspect Med (2018) 9.e029017.

The role of gut microbiota with multiple sclerosis. 54. Miyake S, Kim S, Suda W, Oshima K, Nakamura M, Matsuoka T, et al. Dying in the gut microbiota with patients with multiple sclerosis, with a striking depletion of species belonging to clostridia XIVa and IV clusters. PloS One (2015) 10(9):e0137429. doi:10.1371/journal.pone.0137429.

The role of gut microbiota with multiple sclerosis. 55. Lee YK, Menezes JS, Umeshki Y, Mazmanian SK. Proinflammatory T-cell responses to gut microbiota promote experimental autoimmune encephalomyelitis. Proc Natl Acad Sci USA (2011) 108(Suppl 1):4615–22. doi:10.1073/pnas.1000882107.

The role of gut microbiota with multiple sclerosis. 56. Jung S, Gandhi R, Cox LM, Li N, von Glehn F, Yan R, et al. Alterations of the gut microbiome in multiple sclerosis. Nat Commun (2016) 7:12015–5. doi:10.1038/ncomms12015.
75. Ochoa-Repa L, Carelli G, Galiotte S, Bennett A, Harabagiu A, Parslow R, et al. Polyspecific T cell epitopes in human T cell receptor repertoires. *Sci Transl Med* (2010) 2(6):64ra158. doi:10.1126/scitranslmed.3009759

76. Berer K, Mues M, Koutrolos M, Rasi ZA, Boziki M, Johner C, et al. Commensal microbiota and myelin autointolent cooperates to trigger autoimmune demyelination in mice (2011) 479(7574):538–41. doi:10.1038/nature10554

77. Knights D, Ward TL, McKinlay CE, Miller H, Gonzalez A, McDonald D, et al. Rethinking “enterotype.” *Cell Host Microbe* (2014) 16(4):433–7. doi:10.1016/j.chom.2014.09.013

78. Farrokhdi V, Nemati R, Nichols FC, Yao X, Anstatt E, Fujiwara M, et al. Bacterial lipopolysaccharide, lipid 554, is a microbiome-associated biomarker for multiple sclerosis. *Clin Transl Immunol* (2015) 2(11):e88–8. doi:10.1038/cti.2013.11

79. Choi IY, Lee P, Adany P, Hughes AJ, Belliston S, Denney DR, et al. In vivo evidence of oxidative stress in brains of patients with progressive multiple sclerosis. *Mult Scler Hoennlinds Basingtoke Engl* (2018) 24(8):1029–38. doi:10.1177/1352458517715568

80. Prime M, Mitecich-Turk D, Langerholc T. Analysis of short-chain fatty acids in human feces: A scoping review. *Anal Biochem* (2017) 526:9–21. doi:10.1016/j.ab.2017.03.007

81. Branco V, Al-Ashmaki M, Kowel C, Aamar F, Abbsapor A, Toth M, et al. The gut microbiota influences blood-brain barrier permeability in mice. *Sci Transl Med* (2014) 6(263):263ra158. doi:10.1126/scitranslmed.3009759

82. Koh A, De Vadder F, Kovatcheva-Datchary P, Backhed F. From dietary fiber to host physiology: Short chain fatty acids as key bacterial metabolites. *Cell* (2016) 165(6):1332–45. doi:10.1016/j.cell.2016.05.041

83. Duscha A, Gisevius B, Hirschberg S, Vissarach N, Stangl GL, Eilers E, et al. Propionic acid shapes the multiple sclerosis disease course by an immunomodulatory mechanism. *Cell* (2020) 180(6):807–810.e16. doi:10.1016/j.cell.2020.02.035

84. Wu GD, Chen J, Hoffmann C, Bittinger K, Chen YY, Keilbaugh SA, et al. Linking long-term dietary patterns with gut microbiota enterotypes. *Science* (2011) 334(6052):105–8. doi:10.1126/science.1208344

85. Kaur H, Bose C, Mande SS. Tryptophan metabolism by gut microbe and gut-brain axis: An in vivo analysis. *Front Neurosci* (2019) 13:1365–5. doi:10.3389/fnins.2019.01365

86. Logsdon AD, Erickson MA, Rhea EM, Salameh TS, Banks WA. Gut reactions: How the blood-brain barrier connects the microbiome and the brain. *Exp Biol Med Maywood NJ* (2018) 243(2):159–65. doi:10.1177/1535370717743766

87. Rothhammer V, Mascanfroni ID, Busse L, Takenaka MC, Kenison JE, Mayo L, et al. Type I interferons and microbial metabolites of tryptophan modulate astrocyte activity and central nervous system inflammation via the aryl hydrocarbon receptor. *Nat Med* (2016) 22(6):586–97. doi:10.1038/nm.4106

88. Mayo L, Trauger SA, Blain M, Nadeau M, Patel B, Alvarez J, et al. Regulation of astrocyte activation by glycolipids drives chronic CNS inflammation. *Nat Med* (2014) 20(10):1147–56. doi:10.1038/nm.3881

89. Zheng D, Liwinski T, Elina E. Interaction between microbiota and immunity in health and disease. *Cell Res* (2020) 30(6):492–506. doi:10.1093/celres/caaa020-032-037

90. Lim CK, Brew BJ, Sundaram G, Guillemin GI. Understanding the roles of the kynurenine pathway in multiple sclerosis progression. *Int J Tryptophan Res* (2010) 3:157–67. doi:10.4137/IJTR.S4294

91. Kimm H, Chenn V, Huang S, Heinze M, Tan KL, Kersten S, et al. The role of inflammatory and neurodegenerative diseases. *Int J Tryptophan Res* (2019) 6(1):6604. doi:10.4049/jimmunol.179.10.6604

92. Reay PA, Kantor RM, Davis MM. Use of global amino acid replacements to improve T cell epitope mimicry? *J Immunol* (1994) 152(8):3946.

93. de Paolis ML, Rodrigues DH, Tiesca HR, Barsante MM, Souza MA, Ferreira AP. Genistein down-modulates pro-inflammatory and anti-inflammatory cytokines and reverses clinical signs of experimental autoimmune encephalomyelitis. *Int Immunopharmacol* (2008) 8(9):1291–7. doi:10.1016/j.intimp.2008.05.002

94. Cucca C, Caricchio R, Galfetti S. Targets of autoreactivity: The role of bacterial infections in the extracellular exposure of lupus nuclear autoantigens. *Front Immunol* (2019) 10:267088–8. doi:10.3389/fimmu.2019.02608

95. Garg A, Kumar B, Singhal N, Kumar M. Using molecular-mimicry-inducing pathways of pathogens as novel drug targets. *Drug Discovery Today* (2019) 24(9):1943–52. doi:10.1016/j.drudis.2018.10.010

96. Benoit C, Mathis D. Autoimmunity provoked by infection: how good is the case for T cell epitope mimicry? *Nat Immunol* (2001) 2(9):797–801. doi:10.1038/immunol.2001.79

97. Fardin AM, Miller SD. Molecular mimics can induce novel self peptide-reactive CD4+ T cell clones in autoimmune disease. *J Immunol* (2007) 179(4):6604. doi:10.4049/jimmunol.179.10.6604

98. Reay PA, Kenton RM, Davis MM. Use of global amino acid replacements to define the requirements for MHC binding and T cell recognition of moth cytochrome c (93–103). *J Immunol* (1994) 152(8):3946.

99. Wucherpfennig KW, Allen PM, Celada F, Cohen BR, De Boer R, Garcia KC, et al. Polyspecificity of T cell and B cell receptor recognition. *Semim Immunol* (2009) 19(4):216–24. doi:10.1016/j.smim.2007.02.012

100. Planas R, Santos R, Tomas-Oyar P, Cruciani C, Lutterotti A, Faigle W, et al. L-PFU. L-fucose synthase is a CD4+ T cell-specific autoantigen in DBB2/02 patients with multiple sclerosis. *Sci Transl Med* (2018) 10(462):eaat4301. doi:10.1126/scitranslmed.aat4301

Elsayed et al. 2012.
Dysfunction develops at the onset of experimental autoimmune perfringens. Brain J Neurol

The intestinal barrier in multiple sclerosis: implications for pathophysiology and neuromyelitis optica spectrum disorder: Implication for intervention. Neurosci (2020) 82:193. doi: 10.1177/1352458X16652498

Elsayed et al. 10.3389

A functional and structural basis for TCR cross-reactivity in multiple sclerosis. Hypotheses (2012) 78(6):763. doi: 10.1016/j.hysu.2012.02.026

Lang HLE, Jacobsen H, Izenius S, Andersson C, Madesen L, et al. A functional and structural basis for TCR cross-reactivity in multiple sclerosis. Nat Immunol (2002) 3(10):940–10. doi: 10.1038/nn835

Varrin-Doyer M, Spencer CM, Schulze-Topphoff U, Nelson PA, Stroud RM, et al. Aquaporin 4-specific T cells in neuromyelitis optica exhibit a Th17 bias and recognize clostridium ABC transporter. Ann Neurol (2012) 72(1):53–64. doi: 10.1002/ana.23651

Cree BAC, Spencer CM, Varrin-Doyer M, Baranzini SE, Zamvil SS. Gut microbiome analysis in neuromyelitis optica reveals overabundance of clostridium perfringens. Ann Neurol (2016) 80(3):443–7. doi: 10.1002/ana.24718

Cama-Lemarroy CR, Metz L, Meddings JR, Sharkey KA, Wee Yong V. The intestinal barrier in multiple sclerosis: implications for pathophysiology and therapeutics. Brain J Neurol (2018) 141(7):1900–16. doi: 10.1093/brain/awy131

Nouri M, Bredberg A, Westrom B, Lavasani S. Intestinal barrier dysfunction develops at the onset of experimental autoimmune encephalomyelitis, and can be induced by adoptive transfer of auto-reactive T cells. PLoS One (2014) 9(9):e106335. doi: 10.1371/journal.pone.0106335

Buscarini MC, Cerasoli B, Annibali V, Policano C, Lionetto L, Capi M, et al. Altered intestinal permeability in patients with relapsing-remitting multiple sclerosis: A pilot study. Mult Scler J (2017) 23(3):442–6. doi: 10.1177/1352458X16652498

Secher T, Kaseem S, Benamar M, Bernard I, Roury M, Barreau F, et al. Oral administration of the probiotic strain escherichia coli nissle 1917 reduces susceptibility to neuroinflammation and repairs experimental autoimmune encephalomyelitis-induced intestinal barrier dysfunction. Front Immunol (2017) 8:1096. doi: 10.3389/fimmu.2017.01096

Macpherson AJ, Harris NL. Interactions between commensal intestinal bacteria and the immune system. Nat Rev Immunol (2004) 4(6):478–85. doi: 10.1038/nri1373

Smith PM, Howitt MR, Panikov N, Michaud M, Gallini CA, Bohlooly Y, M, et al. The microbial metabolites, short-chain fatty acids, regulate colonic treg cell homeostasis. Science (2013) 341(6145):569–73. doi: 10.1126/science.1241165

Atarashi K, Tanoue T, Oshima K, Suda W, Nagano Y, Nishikawa H, et al. Patients with relapsing-remitting multiple sclerosis have normal treg function when cells expressing IL-7 receptor α-chain are excluded from the analysis. J Clin Invest (2008) 118(10):3411–9. doi: 10.1172/JCI335365

Smith PM, Howitt MR, Panikov N, Michaud M, Gallini CA, Bohlooly Y, M, et al. The microbial metabolites, short-chain fatty acids, regulate colonic treg cell homeostasis. Science (2013) 341(6145):569–73. doi: 10.1126/ science.1241165

Atarashi K, Tanoue T, Oshima K, Suda W, Nagano Y, Nishikawa H, et al. Treg induction by a rationally selected mixture of clostridia strains from the human microbiota. Nature (2013) 500(7461):232–6. doi: 10.1038/nature12331

Clarke TD, Davis KM, Lysenko ES, Zhou AY, Yu Y, Weiser JN. Recognition of peptidoglycan from the microbiota by Nod1 enhances systemic innate immunity. Nat Med (2010) 16(2):228–31. doi: 10.1038/nm.2087