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**SYSTEMATIC REVIEW** OPEN ACCESS

# Exploring the Role of Microglial Cells in the Gut–Brain Axis Communication: A Systematic Review

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## ABSTRACT

The gut–brain axis (GBA) is a bidirectional communication system between the gastrointestinal tract and the CNS, playing a key role in neurological function, immune response, and metabolism. Microglia, the resident immune cells in the brain, are crucial regulators of neuroinflammation and synaptic plasticity. Recent studies indicate that the gut microbiota modulates microglial activity through metabolic and immune pathways, with implications for neurodegenerative, neurodevelopmental, and psychiatric disorders. However, the mechanisms underlying microbiota–microglia interactions remain unclear. Following a systematic screening of 4481 studies, 20 preclinical studies met the inclusion criteria and were reviewed in depth to assess microbiota–microglia interactions. These studies were found by searching in PubMed, Science Direct, and Google Scholar. The findings synthesize results from 20 carefully selected studies examining the impact of gut microbiota on microglial function. Experimental models, including fecal microbiota transplantation, dietary interventions, and bacterial supplementation, were analyzed. Microglial activity was assessed through immunohistochemistry, gene expression profiling, and functional assays. Most studies suggest that gut dysbiosis promotes microglial overactivation and neuroinflammation through pathways involving microbial-derived short-chain fatty acids (SCFAs), bile acids, and neuroimmune signaling cascades such as TLR4/NF-κB and the NLRP3 inflammasomes, whereas microbiota-targeted interventions reduce inflammation and support cognitive function. Despite these promising findings, inconsistencies in study methodologies and microbiota analyses limit comparability and clinical translation. This review offers a unique synthesis of studies specifically linking gut microbiota alterations to microglial states, neuroinflammatory signatures, and cognitive outcomes across diverse experimental models. It highlights the therapeutic potential of microbiota-based strategies for modulating microglial function and mitigating neuroinflammatory diseases.

**Abbreviations:** APP/PS1, amyloid precursor protein-presenilin 1; AT, Acorus tatarinowii; BBB, blood–brain barrier; CCI, controlled cortical impact; CP, chronic periodontitis; CUMS, chronic unpredictable mild stress; DAMPs, danger-associated molecular patterns; PAMPs, pattern-associated molecular patterns; FD, fiber deficiency; FMT, fecal matter transplantation; FTM-CUMS, chronic unpredictable mild stress; GBA, gut–brain axis; GF, germ-free; ICV, intracerebroventricular; LPS, lipopolysaccharide; MDP, muramyl dipeptide; MCAO, middle cerebral artery occlusion; MWM, Morris water maze; NOR, novel object recognition; OLT, object location test; PVN, paraventricular nucleus; PD, Parkinson's disease; PAMPs, pattern-associated molecular patterns; SCFAs, short-chain fatty acids; SD, sleep deprivation; SCCA, short-chain fatty acids and carboxylic acids; SHR, spontaneously hypertensive rats; SYN, synaptophysin; TBI, traumatic brain injury; T1D, Type 1 diabetes; TβMCA, tauro-β-muricholic acid.

Nadia Suyin Ortiz-Samur and Akshay Kumar Vijaya contributed equally.

## 1 | Introduction

The gut–brain axis (GBA) has emerged as a critical bidirectional communication system between the gastrointestinal tract and the CNS (Burokas et al. 2015; Ma et al. 2017) influencing neurological functions, immune responses, and metabolic pathways (Cryan et al. 2019). This bidirectional signaling is mediated through neural, humoral, and endocrine pathways, with the vagus nerve playing a central integrative role (Burokas et al. 2015). Microglia, the resident immune cells in the brain, are central mediators of neuroinflammatory responses and synaptic plasticity within this complex network (de Deus et al. 2024; Luck et al. 2020; Mela et al. 2023; Pérez-González et al. 2023). Recent research has highlighted the role of gut microbiota in modulating microglial activity, with implications for various neurological disorders, including Alzheimer's disease (AD), Parkinson's disease (PD), schizophrenia, multiple sclerosis, and other neurodevelopmental and psychiatric conditions (Intili et al. 2023; Vijaya et al. 2024; Zhao et al. 2023).

Microglial cells are highly responsive to environmental cues, including microbial metabolites, immune signals, and dietary components. Microglial dysfunction, marked by exaggerated inflammatory responses or impaired synaptic pruning, has been implicated in the pathogenesis of several neurodegenerative and neurodevelopmental conditions, often serving as an early indicator of disease progression. The gut microbiota plays a fundamental role in shaping microglial function through metabolic and immune signaling pathways, such as short-chain fatty acids (SCFAs), other bacterial-derived metabolites, and activating pattern recognition receptors (Churchward et al. 2023; Spielbauer et al. 2024). Dysbiosis, an imbalance in microbial composition, has been linked to altered microglial states that represent context-dependent phenotypes shaped by developmental stage, environment, and pathology, ranging from homeostatic surveillance to proinflammatory and disease-associated profiles (Paolicelli et al. 2022), contributing to chronic neuroinflammation and cognitive impairments (Shi et al. 2021). Studies have shown that specific gut microbiota-derived metabolites, such as butyrate and acetate, exert neuroprotective effects by reducing proinflammatory cytokine production and modulating microglial polarization states (Huang et al. 2024). Furthermore, microbial dysbiosis in conditions like diabetes, obesity, and aging has been associated with increased neuroinflammation, leading to cognitive dysfunction and neurodegeneration (Liu et al. 2024; Vijaya et al. 2024).

Although the gut microbiota is increasingly recognized as a key modulator of brain immune function, major questions remain about the specific molecular signals and cellular pathways involved, particularly those affecting microglial behavior. Despite growing recognition of microbiota–microglia crosstalk, the mechanistic understanding of how microbial dysbiosis drives microglial phenotypic changes remains limited. Particularly, the role of microglia in mediating downstream cognitive or behavioral effects of gut-derived signals remains unclear. While many studies support the connection between gut microbiota and microglial activity, the precise mechanisms underlying this relationship remain incompletely understood. A significant challenge is the heterogeneity of study designs, including variations in animal models, microbiota analysis techniques, and behavioral assessments. Several studies have investigated the impact of gut dysbiosis on microglial function in different pathological contexts, including aging, neurodegeneration, metabolic disorders (Liu et al. 2024; Ma et al. 2024; Zhao et al. 2023), and neurotoxicity (Xue et al. 2020). These studies have employed diverse experimental models, ranging from dietary interventions (Lv et al. 2024; Vijaya et al. 2024) and fecal microbiota transplantation (FMT) (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021) to antibiotic-induced microbiota depletion and bacterial supplementation, to elucidate the effects of gut microbiota on microglial state and neuroinflammatory pathways (Li et al. 2024; Liu et al. 2024). Understanding the specific molecular pathways and microbial taxa involved in neuroinflammation remains one of the main crucial areas for future investigation.

Microglial overactivation is increasingly recognized as a central mechanism in GBA communication, particularly in neuroinflammation and cognitive decline (Shi et al. 2021; Vijaya et al. 2024). Additionally, the modulation of microglial polarization by microbial metabolites has been demonstrated in neurodegenerative disease models, where the administration of beneficial bacterial strains such as *Blautia producta*, some of *Lactobacillus* and *Bifidobacterium* strains, restores homeostatic microglial function and attenuates neuroinflammatory responses (Liu et al. 2024; Megur et al. 2020). However, inconsistencies exist regarding how specific microbial metabolites directly mediate neuroprotection, highlighting the need for standardized methodologies and longitudinal studies (Owusu Kyei-Baffour et al. 2025).

This review aims to critically evaluate preclinical studies that explore the mechanistic pathways connecting gut microbiota alterations to microglial function, with a focus on neuroinflammation and cognitive outcomes. We also aim to identify consistent patterns and knowledge gaps that could inform future research and therapeutic strategies. It synthesizes findings from 20 peer-reviewed studies examining the role of microglial cells in GBA communication. It provides a comprehensive overview of methodologies used to assess microglial overactivation and the gut microbiota interactions, including microbiota sequencing, inflammatory marker analysis, and behavioral assays. By categorizing the studies into key themes, such as microglial overactivation mechanisms, the influence of gut microbiota on neuroinflammation, and the implications for neurodevelopmental and neurodegenerative disorders, we aim to identify common patterns and divergent findings in the literature. Furthermore, we discuss the potential therapeutic implications of targeting microglial–microbiota interactions for neurological diseases.

This review contributes to the expanding body of knowledge on gut–brain communication by thoroughly analyzing microglial involvement in GBA-mediated processes. By integrating data from selected studies following predefined criteria, we aim to improve our understanding of how gut microbiota influences brain immune homeostasis and identify future research directions in this rapidly evolving field. Identifying specific bacterial strains and microbial-derived metabolites that modulate microglial function carries a significant promise for the development of novel therapeutic interventions to mitigate neuroinflammatory diseases and cognitive disorders (Li et al. 2024; Xia et al. 2021; Xu et al. 2023).

## 2 | Materials and Methods

### 2.1 | Research Question and Justification

A comprehensive literature search was conducted across different databases such as "PubMed", "Scopus", "Embase", "Science Direct" and "Google Scholar" using the key words "Gut-brain axis and microglia cells", "Microbiota and neuroinflammation", "Microbiota and microglia cells" and "Gut-brain axis and neurodegeneration". The search results were tabulated in Excel file with the heading like "Keywords", "database", "Journal", "year of publication", "article title," and "DOI number".

Upon collection of the articles by two different independent scholars, they were merged for the selection criteria process. The selection criteria process begins by removal duplicates of the article followed by the exclusion and inclusion criteria.

Inclusion criteria prioritized original studies published in English within the past 10 years (January 2015 to February 2025) that address microglia state and activation in the context of the gut–brain axis, particularly those about microbiota or gut-derived metabolites, and mainly articles that focused on addressing the role of microglia in gut–brain communication. Furthermore, the articles can include studies from animal and human models.

Exclusion criteria eliminated narrative reviews, brief articles, conference abstracts and studies in general that lack any original data. Also, in keeping with the time, any dubious articles especially those from predator journals were omitted as so were the articles not published in English. Furthermore, any articles that does not focus on the theme of this review "microglia role in gut-brain communication" were also omitted. All the applied criteria were recorded in an Excel file for reference and smooth filtration have been included in the review as supplementary sheets (Supporting Information).

The PRISMA diagram guided the study selection process, documenting the number of studies identified, screened for duplication, exclusions, and inclusions. The process of study selection, including filtration and the application of inclusion and exclusion criteria, was systematically designed as depicted using a flow diagram (Figure 1).

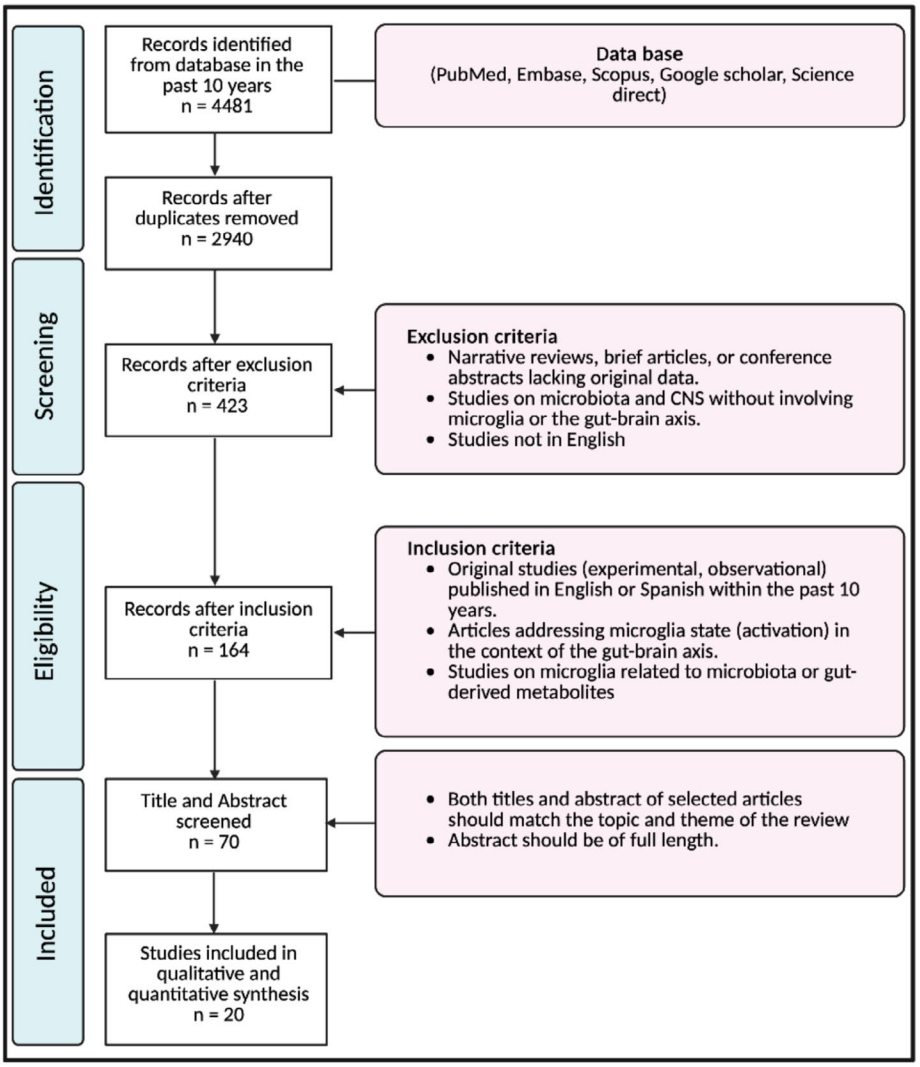


**FIGURE 1** | Flowchart of selection of studies process of systematic review.

### 2.2 | Data Extraction, Quality Assessment, and Analysis

The quality of selected studies was assessed using the Critical Appraisal Skills Program (CASP) checklist, evaluating study design, statistical methods, and potential biases. Two authors independently extracted the data to ensure accuracy and discussed differences. Key findings and limitations are summarized to provide a comprehensive synthesis of the evidence, contributing to a deeper understanding of the role of the gut–brain axis in microglia activation and its implications for neurological health and disease.

## 3 | Results

### 3.1 | Studies

This systematic review examines the role of microglial cells in GBA communication, focusing on microbiota dysbiosis, neuroinflammation, and cognitive function. Following the above-mentioned methodology, we examined a total of 20 peer-reviewed articles about this specific topic. The studies utilized diverse rodent models, including dietary fiber deficiency (FD), aging, hypertension, infection, stress, stroke, neurodegeneration, neurotoxicity, diabetes, and traumatic brain injury (TBI). Methodologies included 16S rRNA sequencing for microbiota profiling, FMT to establish causal links, and bacterial supplementation. Microglial states were assessed through Iba1$^+$, CD68, TREM2 staining, while qPCR and western blot measured neuroinflammatory markers (TNF-α, IL-6, IL-1β, NLRP3). Cognitive functions were evaluated using the Morris Water Maze (MWM), Novel Object Recognition (NOR), Barnes Maze, Nesting Behavior, Y-maze, Object Location (OLT), and Passive Avoidance tests. Key findings reveal that gut dysbiosis triggers microglial overactivation and neuroinflammation, often leading to cognitive decline. Several studies confirmed that FMT and SCFAs modulation can reverse inflammation and restore cognitive function, emphasizing the therapeutic potential of microbiota-targeted interventions in neuroinflammatory conditions.

### 3.2 | General Considerations

Among all the markers used to analyze neuroinflammation, TNF-α, IL-6, and IL-1β seem to be the common ones measured by mRNA expression in specific brain areas such as the hippocampus and cortex or microglial cells under an infectious stimulus (Churchward et al. 2023; He et al. 2024; Huang et al. 2024; Li et al. 2024; Luck et al. 2020; Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021; Shi et al. 2021; Spielbauer et al. 2024; Xia et al. 2021; Xue et al. 2020; Zhao et al. 2023). Other interleukins such as IL-18, IL-17, IFN-γ, and IL-10 mRNA expression were also measured as markers of neuroinflammation, with IL-10 as an indirect marker since it is an anti-inflammatory cytokine (Churchward et al. 2023; Huang et al. 2024). Besides, pTau and Aβ42 protein expression were also increased in a neuroinflammation environment such as neurodegenerative disease linked to microglial overactivation (Lv et al. 2024). Interestingly, some authors also showed an increase in Cox-2, iNOS, and PTP1B protein expression by immunohistochemistry or western immune blotting as an indirect measurement of neuroinflammation (Lv et al. 2024; Shi et al. 2021).

To analyze microglial states, the studies applied different approaches, where Iba-1$^+$ cells in colocalization with other proteins such as CD68, CD16/32, and CD206 measured by immunohistochemistry together with morphological analysis of microglial cells are presented in most of the studies, showing an increase in those markers, or modification in microglial shape (bigger, rounder, shorter branches), associated with neuroinflammation (He et al. 2024; Li et al. 2024; Luck et al. 2020; Ma et al. 2017; Shi et al. 2021; Xia et al. 2021; Zamudio-Flores et al. 2025; Zeng et al. 2024). Another interesting measurement of microglial overactivation was the analysis of PSD95/Iba-1$^+$ cells in the hippocampus of FD mice, showing the phagocytic capacity of those cells (Shi et al. 2021). The CCL3, CCL5, CD16, CD11b, TREM2, and TMEM119 expressions were also analyzed in brain tissue as microglial state markers showing an increase or decrease depending on the neuroinflammatory environment (He et al. 2024; Lv et al. 2024; Spielbauer et al. 2024). NLRP3 inflammasome activation, a clear signal of microglial response against danger-associated molecular patterns and pathogen-associated molecular patterns, was also measured mainly through NLRP3, ASC, and caspase 1 expression by western blot (Ma et al. 2024; Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021).

Most of these articles cited possible mechanisms underlying the neuroinflammation-microglial overactivation events. Some of them describe possible molecules or metabolites involved in the modulation of the neuroinflammatory milieu. SCFAs, closely related to microbiota composition due to their production by bacterial fermentation produced predominantly in the colon, have been related to neuroinflammation and microglial overactivation in a couple of the selected manuscripts (Churchward et al. 2023; Li et al. 2023; Shi et al. 2021). Another article from our selection remarked that some neurotransmitters, such as glutamate, are part of the cause/effect of neuroinflammation and microglial overactivation related to gut dysbiosis (Zhao et al. 2023). Finally, some of the studies mentioned in this section describe NLRP3 or CaMKIId activation as mediator pathways of the neuroinflammation occurring in the different animal models (Shi et al. 2021). However, most of them point out NFkB as the main molecular pathway involved, giving details of the different molecules of the pathway affected by the experimental condition (Liu et al. 2024; Lv et al. 2024; Spielbauer et al. 2024) (see Tables 1, 2 and 3 for further information).

### 3.3 | Microglial State, Neuroinflammation and Cognitive Impairment

Most of the studies of our selection (15 [directly] and 3 [not directly] out of 20) highlighted neuroinflammation as the primary link between microbiota dysbiosis and microglial reactivity. In line with this, nine studies addressed all three key areas: microglial state, neuroinflammation, and cognitive function, where six specifically directly linked microbiota to cognitive outcomes. While 10 of them focused on two of these domains, most frequently microglial state and neuroinflammation, only one explored a single aspect. Microglial overactivation emerged

**TABLE 1** | Microglial state, neuroinflammation, and cognitive performance due to microbiota changes.

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| FD diet (15 weeks) | Male C57BL/6J (11 weeks old, $n = 15$) | ↑ Iba-1, CD68, cell size ↓ microglial branches | ↑ TNF-α, IL-6, IL-1β, PTP1B | OLT (↓DR), TOMT (↓DR), Nesting test (↓score) | ↓ *Bacteroidetes, SCFAs* ↑ *Proteobacteria* | Fiber deficiency disrupts gut–brain axis homeostasis, promoting neuroinflammation, microglial overactivation, synaptic loss, and cognitive impairment | Shi et al. (2021) |
| Aged mice (4 weeks) | C57BL/6J (4–8 weeks vs. 24 months old, $n = 5$–10) | ↑ Iba1, iNOS | ↑ IL-6, TNF-α, IL-1β, IL-18, Saa1, F4/80, *NLRP3, ASC, Caspase-1, ICAM-1* | NOR (↓DR) | ↑ *Lachnospiraceae_bacterium, Muribaculaceae_bacterium, Heminiphilus_faecis, Desulfovibrio_sp., Clostridiaceae_bacterium* ↓ *Akkermansia_muciniphila, Erysipelotrichaceae_bacterium, Mammaliicoccus_lentus, Staphylococcus_nepalensis, Micrococcaceae_bacterium, Yaniella_halotolerans, Bacteroides_acidifaciens* | Natural aging induces gut dysbiosis, increases systemic and brain TβMCA levels, overactivated microglia, triggers neuroinflammation, and leads to cognitive decline | Ma et al. (2024) |
| SD (7 days, 20 h/day) | Male C57BL/6J (5 weeks old, $n = 10$–16) | ↑ Iba1$^+$, CD68$^+$, C1q | Not measured directly | NOR (↓DR) Y-maze (↓SA) | ↑ *Clostridia* ↓ *Bacilli, Betaproteobacteria, Sphingobacteria, Akkermansiamuciniphila* | SD induces gut dysbiosis, reduces SCFAs, overactivated microglia, promotes synaptic loss, and impairs cognitive function without affecting anxiety-like behaviors. | Li et al. (2023) |
| CP model (12 months) | Male C57BL/6 (12 weeks old, $n = 18$) | ↑ Iba1 | ↑ IL-1β, IL-6, TNF-α | Y-maze (↓SA), PA (↓ latency, ↑errors) MWM (↑escape latency, ↓ platform crossings) | Saliva: ↑ *Firmicutes, Actinobacteria, Acidobacteria, Rodentibacter, Dubosiella, Streptococcus* ↓ *Proteobacteria, Delftia, Serratia* Feces: ↑ *Firmicutes, Verrucomicrobia, Dubosiella, Muribaculum, Butyricicoccus,* ↓ *Tenericutes, Lactobacillus, Delftia, Anaeroplasma, Gordonibacter* | CP induces oral and gut dysbiosis, disrupts intestinal and blood–brain barriers, triggers systemic and brain inflammation via TLR4/MyD88/NF-κB pathway activation, leading to microglial overactivation, synaptic deficits, neuronal loss, and cognitive decline. | Xue et al. (2020) |

(Continues)

**TABLE 1** | (Continued)

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| Exercise protocol (16 weeks) in hypertensive rat model | Male SHR/WKY (12-week-old, $n = 8–10$) | ↑ Iba-1$^+$, cell size,% activated morphology ↓ process length | ↑ TNF-α, IL-1β, IL-6 | — | ↑ *Firmicutes, Patescibacteria*, lactate ↓ *Bacteroidetes, Proteobacteria, Actinobacteria*, acetate- and butyrate | Exercise prevented hypertension-induced cognitive deficits, gut dysbiosis, microglial overactivation, and neuroinflammation in SHR rats, while FMT from sedentary hypertensive donors reproduced these impairments, confirming the gut–brain axis role in neurocognitive dysfunction | Xia et al. (2021) |
| CUMS rats (5 weeks) | Male Sprague–Dawley rats ($170 \pm 20$ g, $n = 6$) | ↑ Iba-1 | ↑ IL-1β, TNF-α | — | Not assessed | CUMS induced cognitive impairment, microglial overactivation, neuroinflammation, and inflammasome activation in the HIP and PFC; FMT from healthy donors reversed these effects | Rao, Qiao, et al. (2021), Rao, Xie, et al. (2021) |
| CUMS rats (4 weeks) | Male Sprague–Dawley, ($180 \pm 20$ g, $n = 6$) | ↑ Iba-1 | ↑ IL-1β, TNF-α, NLRP3, ASC, Caspase-1 | — | ↓ *Bacteroidetes, Desulfobacterota, Alloprevotella, Lachnospiraceae, Roseburia, Romboutsia, Ruminococcus* ↑ *Firmicutes, Colidextribacter, Muribaculaceae, Oscillospiraceae* | CUMS induced cognitive impairment, microglial overactivation, neuroinflammation, alterations in 5-HT signaling, intestinal barrier disruption, and dysbiosis of the gut microbiota. These facts were ameliorated by FMT from healthy rats which highlighted the key role of microbiota | Rao, Qiao, et al. (2021), Rao, Xie, et al. (2021) |

(Continues)

**TABLE 1** | (Continued)

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| MCAO/R rats (2 h occlusion +7 days reperfusion) | Male Sprague–Dawley (7 weeks old, $n = 5–9$) | ↑ Iba-1$^+$/CD16/32$^+$ (M1) ↑ Iba-1$^+$/CD206$^+$ (M2) | ↑ TNF-α, IL-6, IL-17, IFN-γ | — | ↑ *Verrucomicrobia, Akkermansia, Phascolarctobacterium, Blautia* ↓ *Tenericutes, Cyanobacteria, Prevotella_copri* | Ischemic stroke caused neurological impairment, increased infarct size, neuronal apoptosis, neuroinflammation, a proinflammatory microglial shift, and gut microbiota dysbiosis with reduced diversity. AT oils ameliorated these effects via gut microbiota; benefits lost with antibiotics, restored by FMT | Huang et al. (2024) |
| MPTP-induced PD mice (4 weeks) | Male C57BL/6 (6 weeks old, 20–22 g) | ↑ Iba-1, COX-2, iNOS, ROS | ↑ IL-1β, TNF-α, RAS, NF-κB (p65/p-p65 ratio) | — | ↓ *Blautia*, Butyrate (SCFA en heces) | MPTP-induced PD mice showed motor deficits, dopaminergic loss, microglial overactivation, and gut dysbiosis marked by reduced Blautia abundance and lower fecal butyrate levels. | Liu et al. (2024) |
| Maternal PSNPs exposure during gestation and lactation (32 days) | C57BL/6 (7 weeks old, $n = 6$) | ↑ Iba-1$^+$ cells; Ameboid-like appearance | ↑ TNF-α | Y-maze (↓SA) | ↑ *Akkermansia, Verrucomicrobiota Family_XIII_AD3011 group, Alistipes,* ↓ *Patescibacteria, Candidatus_Saccharimonas, Lachnoclostridium, Odoribacter, Lachnospiraceae_NK4A136_group, Mucispirillum* | Maternal PSNP exposure induced gut barrier disruption, dysbiosis, microglial overactivation, and TNF-α elevation, impairing neurotransmitter metabolism and promoting systemic and brain inflammation via the gut–brain axis. | Li et al. (2024) |

(Continues)

**TABLE 1** | (Continued)

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| AD model (4 weeks) | Male APP/PS1 mice (6 months old, $n = 12$) | ↑ Iba1, CD11b, iNOS. Hypertrophic ↓branching | ↑ TNF-α, IL-1β | Nesting test (↓score), NOR (↓DR), Barnes Maze (↑latency to escape, ↓target quadrant time) | ↓ *Agathobacter, Faecalibacterium, Ruminococcus, Escherichia-Shigella*. ↑ *Bacillus, Christensenea, norank_f_Erysipelotrichaceae, unclassified_c_Bacilli* | Gut dysbiosis in APP/PS1 mice, marked by reduced SCFA-producing bacteria and butyrate levels, was associated with cognitive decline, microglial overactivation, increased proinflammatory cytokines, and worsened AD pathology. | Lv et al. (2024) |
| T1D-induced mice (2 weeks) | Male C57BL/6 (6 weeks old, $n = 10$) | ↑ Iba-1 | ↑ IL-1β, IL-6, TNF-α | MWM (↓learning & memory), NOR (↓DI), Y-maze (↓SA) | ↑ *Ruminiclostridium, Anaeroplasma, Oscillibacter, Angelakisella, Helicobacter, Harryflintia, Parabacteroides, Candidatus_Saccharimonas, unidentified_Clostridiales, Muribaculum, Intestinimonas, Odoribacter, Alloprevotella, Alistipes* ↓ *Firmicutes, Actinobacteria* | T1D-induced cognitive decline was associated with microglial overactivation, elevated proinflammatory cytokines, and gut dysbiosis characterized by enrichment of pathogenic genera and loss of beneficial bacteria. FMT from healthy control mice reversed these alterations, demonstrating that the gut microbiota mediates these effects. | Zhao et al. (2023) |
| CUMS mice (4 weeks) | Male C57BL/6J (8 weeks old, $n = 8$–12) | ↑ Iba1, CD68, CD11b, microglial number, branch length. Hyper-ramified and ameboid forms. | ↑ IL-6, TNF-α, IL-1β | — | ↓ *Lactobacillus, Bifidobacterium, Akkermansia* ↑ *Helicobacter, Bacteroides, Desulfovibrio* | FMT from CUMS mice induced microglial priming, exaggerated immune responses, impaired hippocampal neurogenesis, and stress sensitivity in recipients. | He et al. (2024) |

(Continues)

**TABLE 1** | (Continued)

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| GF mice (8 weeks) | Kunming; (4–8-week-old, $n = 3$) | ↑ Iba1, CD68, microglial number pro-inflammatory subpopulations (Hip_M1, Hip_M4, PFC_M2) Hyper-ramified and ameboid ↓ homeostatic subpopulations (Hip_M0, PFC_M0) | ↑ IL-1β Inflammasome pathway activation | Y-maze (↓SA) | Complete absence of microbiota | Microbiota absence impaired cognition promoted microglial overactivation, inflammation, oxidative stress and altered transcriptomic profiles, which were reversed by microbial colonization. | Huang et al. (2023) |
| TBI in rats (21 days) | Male Sprague–Dawley, (PND 21–42, $n = 9$–10) | ↓ CEA/cell body area ratio, ramification Amoeboid and bushy | Not measured directly | MWM (↑ escape latency) | ↓ *Firmicutes, Peptostreptococcales-Tissierellales*, ↑ *Prevotellaceae* NK3B31 | TBI caused learning deficits, microglial overactivation with morphological changes, and gut dysbiosis, with microglia and microbiota alterations correlating with cognitive performance, supporting a gut–brain interaction post-TBI | Zamudio-Flores et al. (2025) |
| Pregnancy stress (gestational days 16–20; 5 days) | Female Sprague–Dawley rats (8 weeks old, 250–300 g, $n = 5$) | ↑ $Iba1^+$ area, $Iba1^+$ cell number amoeboid, reactive, rod-like microglia ↓ ramified microglia | No direct measured | — | ↑ α-diversity Altered β-diversity ↑ unclassified_f__*Lachnospiraceae, Treponema, Intestinimonas, Lachnospiraceae_UCG-001, unclassified_k__norank_d__Bacteria* | Pregnancy stress-induced gut dysbiosis impaired fear extinction in offspring, promoting widespread microglial overactivation and reactive morphological changes in the hippocampus and amygdala, leading to synaptic dysfunction. TMF from stressed mothers (ST) replicated these effects. | Zeng et al. (2024) |

(Continues)

**TABLE 1** | (Continued)

| Experimental design | Experimental subject | Microglial state | Neuroinflammation | Cognitive performance | Microbiota changes | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| GF mice, (postnatal day 1–20) | Neonatal gnotobiotic mice, ($n = 5$) | ↓CD11b$^{+}$CD45$^{low}$, Iba-1$^{+}$, CD68, CD36, Macro, Msr1, LOX-1, ameboid morphology ↑ ramified morphology | ↓ TNF-α | — | GF – no microbiota | Neonatal microbiota absence delayed microglial maturation, impaired synaptic pruning, increased synaptic density, and reduced Purkinje cell activity, all of which were reversed by colonization with conventional microbiota or Bifidobacterium, highlighting the critical role of gut microbes in early brain development. | Luck et al. (2020) |
| TBI in mice (28 days) | Male C57BL/6 (8-10 weeks old, 20–25 g) | ↑ CD68$^{+}$cells ameboid and bushy | ↑ IL-1β | No cognitive tests performed. | No major changes in microbiota composition. | Chronic TBI caused long-term gut barrier dysfunction and, upon enteric infection, worsened gut pathology, brain lesion size, and microglial overactivation, highlighting gut–brain axis dysregulation in chronic TBI. | Ma et al. (2017) |

Abbreviations: 5-HT, 5-hydroxytryptamine (serotonin neurotransmitter); APP/PS1, Alzheimer's disease mice model; ASC, apoptosis-associated speck-like protein containing a CARD (inflammasome adaptor protein); AT, Acorus tatarinowii oil; BIF, bifidobacterium-colonized mice; C1q, complement component 1q, involved in synaptic pruning; CCI, controlled cortical impact model for TBI; CD11b, integrin alpha M (microglial activation marker); CD16/32, Fc gamma receptors, markers of proinflammatory M1 microglia; CD206, Mannose receptor C type 1 (marker of anti-inflammatory M2 microglia); CD36, Cluster of Differentiation 36 (scavenger receptor involved in phagocytosis); CD45, Cluster of Differentiation 45 (leukocyte common antigen); CD45^low, Low expression of CD45 (marker of resting surveilling microglia); CD68, Cluster of Differentiation 68 (phagocytic microglia marker); CEA, cell external area; CONV, conventionally colonized microbiota mice; COX-2, cyclooxygenase-2 (enzyme involved in inflammation); CP, chronic periodontitis model; Cr, *Citrobacter rodentium* infection (model of enteric infection); CUMS, Chronic Unpredictable Mild Stress model; DR, discrimination ratio; F4/80, murine macrophage marker; FD, fiber deficiency; FMT, fecal microbiota transplantation; GF, germ-free mice; HC, hippocampus; Hip_M0/M1/M4, hippocampal microglial subpopulations identified via single-cell RNA sequencing; ICAM-1, intercellular adhesion molecule 1; IFN-γ, interferon gamma, proinflammatory cytokine; IL-10, interleukin 10 (anti-inflammatory cytokine); IL-17, interleukin 17 (proinflammatory cytokine); IL-1β, interleukin 1 beta (proinflammatory cytokine); IL-6, interleukin 6 (proinflammatory cytokine); iNOS, inducible nitric oxide synthase (proinflammatory enzyme); MCAO/R, middle cerebral artery occlusion/reperfusion model; mNSS, Modified Neurological Severity Score; MPTP, 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (neurotoxin used to induce PD model); Msr1, macrophage scavenger receptor 1; MWM, Morris Water Maze (spatial learning and memory test); MyD88, myeloid differentiation primary response 88 (TLR signaling adaptor); NF-κB, nuclear factor kappa-light-chain-enhancer of activated B cells; NLRP3, NOD-, LRR- and pyrin domain-containing protein 3 (inflammasome component); NOR, Novel Object Recognition test (recognition memory test), OLT, Object Location Test (spatial memory test); PA, passive avoidance test (learning and memory assessment); PD, Parkinson's disease model; PFC, prefrontal cortex; PFC_M0/M2, prefrontal cortex microglial subpopulations; PSNPs, polystyrene nanoparticles exposure model; PTP1B, protein tyrosine phosphatase 1B (inflammatory regulator); RAS, renin–angiotensin system; ROS, reactive oxygen species; SA, spontaneous alternation in Y-maze (working memory performance); Saa1, serum amyloid A1 (acute-phase protein); SCFAs, short-chain fatty acids; SD, sleep deprivation model; SHR, spontaneously hypertensive rat; SPF, specific pathogen-free mice; ST, step-through (variant of the Passive Avoidance test); T1D, Type 1 diabetes model; TBI, traumatic brain injury; TLR4, Toll-like receptor 4 (innate immune receptor); TNF-α, tumor necrosis factor alpha (proinflammatory cytokine); TOMT, Topographic Orientation Memory Test (spatial orientation assessment); TTC, triphenyltetrazolium chloride staining (brain infarct volume assessment); TβMCA, tauro-β-muricholic acid (bile acid); WKY, Wistar Kyoto Rat (normotensive control for SHR); Y-maze, spontaneous alternation test (working memory measure).

as a central player in the gut–brain axis, with multiple studies emphasizing its role in modulating neuroinflammatory pathways. However, the implication of microglial reactivity in cognitive functions has only been investigated by seven studies from our selection. Different approaches and animal models were used to investigate the interconnection of these facts (see Table 1 for details).

#### 3.3.1 | Disease Animal Models

Across all disease models, neuroinflammation was a consistent feature, often linked to gut dysbiosis and barrier disruption. Depression induced by chronic unpredictable mild stress protocol (CUMS) led to hippocampal and prefrontal cortex neuroinflammation (NLRP3, ASC, Caspase-1, and IL-1β mRNA expression increase), associated with reductions in tight junction proteins (ZO-1, occludin), suggesting that a leaky gut barrier could be in part responsible for those changes (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). Similarly, chronic periodontitis (CP: 12-month ligature around the maxillary second molar) mice showed elevated proinflammatory cytokines (IL-1β, IL-6, TNF-α) in gingival tissue, ileum, and brain, indicating a systemic inflammatory profile. This was coupled with impaired intestinal and blood–brain barrier (BBB) integrity, evidenced by reduced expression of tight junction proteins (claudin-1, occludin, ZO-1) in the gut and brain, as well as increased serum and brain LPS levels. In the brain, CP induced the activation of TLR4/MyD88/NF-κB signaling, further supporting the link between peripheral inflammation and central neuroimmune activation (Xue et al. 2020).

Hypertension (SHR rats) was marked by reduced acetate- and butyrate-producing bacteria linked to an increase in TNF-α, IL-1β, and IL-6 mRNA expression together with microgliosis (Iba-1$^+$ cells) in the paraventricular nucleus (Xia et al. 2021). Microgliosis was also found in the PD model linked to RAS, MAPK, and NF-κB pathway activation, supporting an inflammation-driven pathology (J. Liu et al. 2024). Stroke (middle cerebral artery occlusion- + 7 days reperfusion model) induced both central (CD16/32$^+$ microglia) and systemic inflammation (TNF-α, IL-6, IL-17, IFN-γ serum concentration), alongside broad shifts in microbial taxa, including increases in *Akkermansia* and *Blautia* (Huang et al. 2024). Type 1 Diabetes (T1D) mice also exhibited cortex and hippocampal neuroinflammation (increased IL-1β, IL-6, and TNF-α mRNA expression) with altered microbiota, which negatively correlated with glutamate levels, a key metabolite linked to neuroinflammatory regulation (Zhao et al. 2023).

As mentioned above, microgliosis together or not with significant alterations in microglial morphology emerged as a unifying hallmark between neuroinflammation and the gut microbiota disturbances. CUMS model showed a pronounced microglial overactivation, with increased Iba-1 immunoreactivity detected in both the hippocampus and prefrontal cortex, indicating a sustained neuroimmune response (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). Similarly, in SD mice, hippocampal microgliosis was accompanied by clear signs of excessive synaptic pruning. This was evidenced by enhanced colocalization of synaptic markers such as PSD95 with complement protein C1q and of CD68 and synaptophysin with Iba-1, pointing to overactive phagocytic activity by microglia (Li et al. 2023). CP mice also exhibited significant microgliosis in the hippocampus and cortex, as indicated by increased Iba-1 immunostaining, which occurred alongside synaptic deficits such as reduced synaptophysin expression, linking peripheral inflammation to central immune and synaptic changes (Xue et al. 2020). The microgliosis found in the paraventricular nucleus of SHR rats was related to morphological changes (microglial processes were notably shortened), which indicate a proinflammatory phenotype (Xia et al. 2021). This proinflammatory profile of microglial cells was also evident following ischemic stroke, marked by elevated colocalization of Iba-1 with CD16/32, a marker of classical activation (Huang et al. 2024). Morphological changes in microglial cells were also found in the TBI model, with cellular structure reflecting hypertrophic (reduced area-to-soma ratio) microglia, commonly associated with injury and inflammation (Zamudio-Flores et al. 2025).

Behavioral dysfunction was another feature analyzed in most of the models examined here and was often associated with microglial alterations and gut microbial dysbiosis. Stressed animals exhibited classic behavioral signs of depression and anxiety, such as reduced sucrose preference and increased immobility time, although specific cognitive tests were not performed (He et al. 2024; Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). In contrast, SD mice demonstrated apparent impairments in learning and memory, as evidenced by a reduced discrimination ratio in the NOR and lower performance in the Y-maze, pointing to compromised hippocampal-dependent function (Li et al. 2023). CP mice also showed broad cognitive impairments: reduced short-term memory (Y-maze alternation), long-term memory (passive avoidance test), and spatial learning and memory (longer escape latency and reduced target quadrant time in the MWM). These deficits were tightly associated with neuronal loss in the cortex and hippocampus and underlying neuroinflammatory and synaptic disturbances (Xue et al. 2020). The stroke model found an elevated neurological deficit score (Huang et al. 2024). More direct assessments were seen in TBI and T1D models, where rats and mice, respectively, showed impaired performance in the MWM, highlighting deficits in spatial learning and memory (Zamudio-Flores et al. 2025; Zhao et al. 2023).

Across diverse disease models, gut dysbiosis, often involving SCFA-producing bacteria, is consistently linked to barrier disruption, neuroinflammation, microglial overactivation, and cognitive decline. The shared presence of *Akkermansia* fluctuations, SCFA reduction, and inflammation-prone microbiota (e.g., *Helicobacter, Desulfovibrio*) across the models underscores the central role of the gut–brain axis in driving neuroimmune and cognitive outcomes. Therapeutic strategies targeting microbial composition and their metabolites may offer cross-cutting benefits in managing CNS disorders.

#### 3.3.2 | Dietary Supplementation

FD and Acorus tatarinowii oil treatment represent two contrasting models of how nutritional interventions can shape neuroinflammation, microglial activation states, and cognition via the gut–brain axis. Additionally, environmental exposure to

**TABLE 2** | Mechanistic insights into microbiota–microglia interactions.

| Experimental design | Treatment (dose, duration) | Microglial state | Neuroinflammation | Cognition | Microbiota-derived metabolites | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| T1D mice model<br>BV2 cell line | FMT from healthy donors to T1D mice (100 μL/day, gavage, 14 days)<br>Glutamate (0.1 mM, 12 h) | ↓ Iba-1+ cells (cortex, hippocampus)<br>↓ cell viability | ↓ IL-1β, IL-6, TNF-α | MWM (↑learning & memory), NOR (↑DR), Y-maze (↑SA) | ↑ Glutamate (feces, colon, hippocampus, cortex) | FMT from healthy donors restored gut–brain glutamate levels and improved cognition in T1D mice by reducing microglial overactivation, neuroinflammation, and neuronal loss. Glutamate in vitro treatment reduced high glucose-induced BV2 overactivation, confirming its anti-inflammatory role. | Zhao et al. (2023) |
| Primary microglia (neonatal Sprague–Dawley rat) | SCFA mix (butyrate, valerate, isovalerate, 2-methylbutyrate; 200 or 1000 μM, 1 h pretreatment) + LPS (100 ng/mL, 24 h) | ↓ iNOS, COX-2, ROS | ↓ IL-6, IL-10, NO, NS, IL-1β, TNF-α | Not assessed | ↑ Butyrate, valerate, isovalerate, 2-methylbutyrate<br>↓ 2-hydroxybutyrate | A SCFA mix formulated to mimic the serum profile of mice after FMT was tested in primary microglia. This mix significantly reduced LPS-induced microglial overactivation and increased lipid droplet accumulation, suggesting a combined anti-inflammatory and metabolic regulatory effect. Notably, individual SCFAs (e.g., butyrate alone) did not reproduce these effects, highlighting the importance of SCFA synergy | Churchward et al. (2023) |
| PD mice model<br>BV2 cell line | *B. producta* ($1 \times 10^9$ CFU/day, gavage, 4 weeks)<br>Butyrate (200 μM, 2 h pretreatment +24 h with $MPP^+$) | ↓ $Iba1^+$ cell number, iNOS, ROS, branches number, COX-2<br>↑soma<br>↓ iNOS, COX-2 | ↓ RAS, p-ERK/ERK, NF-κB p65/p-p65<br>↓ ROS, RAS, p-ERK/ERK, p-p65/p65 | Not assessed | ↑ Butyrate (from *B. producta*) | Butyrate-producing *B. producta* reduced microglial overactivation and neuroinflammation by inhibiting the RAS–NF-κB pathway, restoring neuronal markers and improving motor function. | Liu et al. (2024) |

(Continues)

**TABLE 2** | (Continued)

| Experimental design | Treatment (dose, duration) | Microglial state | Neuroinflammation | Cognition | Microbiota-derived metabolites | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| IMG cell line<br>Primary microglia (from adult C57BL/6 mice) | MDP (0.1, 1, 10 μg/mL; 3–72 h; dose- and time-dependent) | ↑ CD11b, Iba-1 (basal); CCL3/5<br>↓ CD16 | ↑ TNF-α, IL-1β, NF-κB2, nuclear p65 | Not assessed | ↑ Peptidoglycan (MDP) | MDP triggered dose- and time-dependent microglial overactivation via NF-κB and MAPK-p38, with low doses boosting CCL chemokines and high doses increasing proinflammatory cytokines. | Spielbauer et al. (2024) |
| WT mice (male C57BL/6J mice;8 weeks old) | TβMCA (10 mM, 2 μL, single ICV) | ↑ $Iba1^+$ cells, soma size, iNOS | ↑ TNF-α, IL-6, IL-1β, IL-1, NLRP3, ICAM1, F4/80, cleaved caspase-1 | NOR (↓ DR), MWM (↓ time in target quadrant, ↓ crossings) | ↑ TβMCA | Central TβMCA administration triggered microglial overactivation, neuroinflammation, and cognitive impairment, mimicking age-related TβMCA buildup from gut dysbiosis. | Ma et al. (2024) |
| SD mice model<br>Neuron/microglia co-culture (primary cell culture; neonatal C57BL/6J mice) | SCFA mix (67.5 mM acetate, 40 mM butyrate; drinking water, 4 weeks)<br>SCFA mix (236 μM acetate, 117 μM butyrate; 15 min pretreatment) + LPS (100 ng/mL, 24 h) | ↓ $Iba1^+$ cell area, $CD68^+$ dots/$Iba\text{-}1^+$ cells, $SYP^+$ puncta/$Iba\text{-}1^+$ cells<br>↓ $Iba1^+$ cell size, $CD68^+$ lysosomes, $SYP^+$ puncta inside microglia | Not measured directly | NOR (↑DR), Y-maze (↑SA) | ↑ SCFA (restored by *A. muciniphila*) | SCFA pretreatment reduced hippocampal microglial overactivation and synaptic engulfment in sleep-deprived mice, preserving synapses and improving cognition. In co-cultures, SCFAs prevented LPS-induced synaptic loss by inhibiting microglial phagocytic activity, highlighting their regulatory role. | Li et al. (2023) |
| FD mice model | SCFA mix (sodium acetate 67.5 mM, butyrate 40 mM, propionate 25.9 mM in drinking water, 15 weeks) | ↓ Iba1 expression, $CD68^+$ signal, $SYP^+$ puncta engulfment | ↓ TNF-α, IL-1β, IL-6 | OLT (↑DR), TOM (↑PI) | ↑ SCFAs (acetate, butyrate, propionate) | SCFA supplementation protected gut integrity, reduced hippocampal inflammation and microglial overactivation, restored synaptic proteins, and improved cognition in FD mice. | Shi et al. (2021) |

(Continues)

**TABLE 2** | (Continued)

| Experimental design | Treatment (dose, duration) | Microglial state | Neuroinflammation | Cognition | Microbiota-derived metabolites | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|---|
| AD mice model BV2 cell line | A. rectalis ($1\times10^9$ CFU/mL, gavage, 4 weeks) NaB (200 μM, 24 h) + LPS (100 ng/mL; 24 h) | ↓ Iba1, CD11b, iNOS ↑ TREM2 ↓ cell size ↑ branch numbers | ↓ IL-1β, COX-2, ROS, p-Akt, p-NF-κB | NOR (↑DR), BM (↑ time in target, ↓ latency) | ↑ butyrate | A. rectalis improved cognition and reduced AD pathology in APP/PS1 mice. Butyrate suppressed microglial overactivation via Akt/NF-κB, while SCFAs promoted a shift toward energy-storing metabolism. | Lv et al. (2024) |

Abbreviations: *A. muciniphila*, *Akkermansia muciniphila*; A. rectalis, *Agathobacter rectalis*; AD, Alzheimer's disease (see Table 1 for details); Akt/NF-κB, Akt/nuclear factor kappa B signaling pathway; BM, Barnes Maze; BODIPY$^+$, boron-dipyrromethene-positive (marker of lipid droplets); BV2, murine microglial cell line; CCL, C–C motif chemokine ligand; CCL3/5, chemokines CCL3 and CCL5; CD11b, microglial/macrophage surface marker; CFU, colony-forming units; COX-2, cyclooxygenase-2; DR, discrimination ratio; FD, fiber-deficient; F4/80, macrophage marker; ICAM1, intercellular adhesion molecule 1; ICV, intracerebroventricular; IFN-γ, interferon gamma; Iba, ionized calcium-binding adapter molecule 1; IL-1β, interleukin-1 beta; IL-6, interleukin-6; IMG cell line, immortalized microglial cell line; iNOS, inducible nitric oxide synthase; LPS, lipopolysaccharide; MPP, 1-methyl-4-phenylpyridinium; MWM, Morris Water Maze; NaB, sodium butyrate; NF-κB p65/p-p65, nuclear factor kappa B p65 subunit and its phosphorylated form; NO, nitric oxide; NOR, novel object recognition; NS, nitric species; p-Akt, phosphorylated Akt; p-ERK/ERK, phosphorylated extracellular signal-regulated kinase/total ERK; p-NF-κB, phosphorylated NF-κB; PD, Parkinson's disease (see Table 1 for details); RAS, renin–angiotensin system; ROS, reactive oxygen species; SA, spontaneous alternation; SCFA, short-chain fatty acids; SD, sleep deprivation; SYP, synaptophysin; T1D, Type 1 diabetes (see Table 1 for details); TβMCA, tauro-β-muricholic acid; TOM, temporal order memory; TREM2, triggering receptor expressed on myeloid cells 2; WT, wild type.

nanoplastics has emerged as another critical factor influencing this triad. FD-induced neuroinflammation in healthy mice and excessive microglial pruning led to synaptic dysfunction and cognitive decline (Shi et al. 2021), whereas oil treatment following ischemic stroke mitigated neuroinflammation, shifted microglial state toward a neuroprotective state, and supported neurological recovery (Huang et al. 2024). Both approaches triggered neuroinflammatory responses but with distinct effects based on the intervention. FD led to a proinflammatory state in the hippocampus, characterized by an increased number of microglial cells (Iba-1$^+$ cell counting) with a reactive morphology (more rounded and less ramified), alongside elevated transcription of inflammatory cytokines (TNF-α, IL-6, IL-1β) (Shi et al. 2021). Conversely, poststroke oil treatment reduced systemic and central inflammation in a dose-dependent manner, lowering levels of TNF-α, IL-6, IL-17, and IFN-γ (Huang et al. 2024). Similarly, prenatal and lactational exposure to polystyrene nanoplastics (50 μg/day by oral gavage suspension for 32 days) also induced neuroinflammation in male offspring, as evidenced by increased brain TNF-α mRNA expression and microgliosis (Iba-1$^+$ cells) (Li et al. 2024).

Microglia reactivity was accordingly changed to the neuroinflammation associated with each approach. FD promoted microglial overactivation in the hippocampus, as indicated by increased colocalization of Iba-1 with postsynaptic density protein 95 (PSD95), suggesting excessive microglial pruning of synapses. This was accompanied by reductions in synaptophysin (SYN) and PSD95, indicative of impaired synaptic integrity (Shi et al. 2021). In contrast, oil treatment after ischemic stroke shifted microglial polarization toward a neuroprotective phenotype, with a decrease in proinflammatory Iba-1+/CD16/32+ microglia and an increase in anti-inflammatory Iba-1+/CD206+ cells in the frontal cortex (Huang et al. 2024).

Regarding cognitive performance, FD adversely affected it, with mice exhibiting deficits in OLT, NOR, and nesting behavior tests. These impairments were linked to hippocampal synaptic dysfunction and neuroinflammation (Shi et al. 2021). In contrast, oil-treated mice after stroke demonstrated improved neurological function in a dose-dependent manner, suggesting a protective effect against ischemic injury (Huang et al. 2024). These outcomes reinforce the idea that neuroinflammatory states and microglial activity critically influence cognitive function and recovery. In the nanoplastic exposure model, while neuroinflammation and microgliosis were confirmed, no cognitive or behavioral outcomes were reported, leaving a gap in the direct interpretation of how gut–brain axis disruption in this context affects behavior (Li et al. 2024).

The link between those events and the gut–brain axis is merely descriptive in these studies, where most authors presented microbiota changes and gut barrier permeability disrupted in their animal models. The three approaches promoted gut barrier disruption (e.g., decreased Muc1/2, occludin, and ZO-1, increased LPS expression and fecal albumin concentration) and altered microbiota composition. Notably, a decrease in Bacteroides and an increase in Proteobacteria phylum in the FD approach, while nanoplastic exposure induced an increase in *Akkermansia* and *Verrucomicrobia* (both decreased with AT oil treatment) and a decrease in *Patescibacteria* and *Candidatus_Saccharimonas*

**TABLE 3** | Gut microbiota modulation of microglial function.

| Experimental design | Treatment | Neuroinflammation | Microglial state | Microglial function | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|
| T1D mice + FMT | FMT from healthy mice (100 μL/day, 14 days) | ↓ IL-1β, IL-6, TNF-α | ↓ Iba-1$^+$ cells | Not specifically evaluated | FMT from healthy mice attenuates neuroinflammation and microglial overactivation in T1D mice, potentially through gut–brain glutamate metabolism regulation, highlighting the role of microbiota in modulating central immune responses | Zhao et al. (2023) |
| Pregnant rats + FMT | FMT from stressed pregnant rats (2 mL, intracolonic, GD16, single dose) Probiotics: *B. longum*, *L. acidophilus*, *E. faecalis* ($\geq 1.4\times10^5$ CFU/mL, drinking water, 5 days) | Not directly measured | ↑ Iba-1$^+$ cell, amoeboid, and rod-shape ↓ branch numbers ↓ Iba-1$^+$ cell, amoeboid and rod-shaped ↑ branch numbers | ↑ PSD95$^+$/ Iba-1$^+$ cells ↓ PSD95$^+$/SYN cell ↓ PSD95$^+$/ Iba-1$^+$ cell ↑ SYN, PSD95, and its colocalization | FMT from stressed pregnant rats induces long-term microglial overactivation and synaptic loss in offspring, contributing to fear extinction impairment. Probiotic treatment during gestation reverses these alterations, restoring microglial morphology and function, supporting a preventive role via the gut–brain axis. | Zeng et al. (2024) |
| SHR + FMT | FMT from exercised rats: (1 mL, oral gavage, every 2 days for 4 weeks) | ↓ TNF-α, IL-1β, IL-6 | ↓ Iba-1$^+$ intensity, microglial number, activation %, cell size, ↑ process length, restored ramified morphology | Not specifically evaluated | FMT from exercised SHR reversed disease-associated microglial and inflammatory changes in hypertensive rats. Findings support microbiota as a mediator of exercise-induced neuroprotection | Xia et al. (2021) |
| CUMS rats + FMT | FMT from healthy rats (1 mL/day, gavage, $2\times10^9$ CFU/mL, 14 days) | ↓ IL-1β, TNF-α | ↓ Iba-1$^+$ | Not specifically evaluated | FMT from healthy donors reversed stress-induced neuroinflammation and glial overactivation, improving depression-like behavior. Findings support the microbiota–gut–brain axis as a therapeutic target in mood disorders. | Rao, Qiao, et al. (2021), Rao, Xie, et al. (2021) |

(Continues)

**TABLE 3** | (Continued)

| Experimental design | Treatment | Neuroinflammation | Microglial state | Microglial function | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|
| CUMS rats + FMT | FMT from healthy rats (1 mL/day, gavage, $2\times10^9$ CFU/mL, 14 days) | ↓ IL-1β, TNF-α, NLRP3, ASC, Caspase-1 | ↓ Iba-1$^+$ | Not specifically evaluated | FMT from healthy rats reversed depressive-like behavior, intestinal damage, and neuroinflammation in stressed rats. Results support the therapeutic role of gut microbiota in stress-related mood disorders via microglial modulation. | Rao, Qiao, et al. (2021), Rao, Xie, et al. (2021) |
| MCAO rats + AT oil ± microbiota depletion | AT oil (100 mg/kg/day, oral gavage, 7 days post-MCAO); ± microbiota depletion; FMT from MCAO rats (1 mL/day, oral, 3 days) | ↓ TNF-α, IL-6, IL-17, IFN-γ (only with FMT) | ↓ Iba-1$^+$/CD16/32$^+$ (M1); ↑ Iba-1$^+$/CD206$^+$ (M2) (only with FMT) | AT oil promotes anti-inflammatory M2 polarization only with microbiota restoration | AT oil reduces neuroinflammation and promotes M2 microglial polarization after stroke, but only when gut microbiota is present. FMT restores AT oil's efficacy after microbiota depletion, confirming microbiota's role in microglial modulation. | Huang et al. (2024) |
| SD mice + *A. muciniphila* + SCFA | *A. muciniphila* ($1\times10^8$ CFU/day, oral gavage, 3 days before +5 days during SD) SCFA mix (67.5 mM acetate +40 mM butyrate, drinking water, 4 weeks) | Not measured directly | ↓ Iba-1, ↓ CD68, ↓ SYP inside microglia, C1q | ↓ SYP$^+$ puncta inside Iba-1$^+$ ↑ PSD95, VGLUT1 | Both *A. muciniphila* and SCFA supplementation prevented synaptic loss and reduced microglial overactivation and synapse engulfment in SD mice, highlighting a gut–brain axis mechanism mediated via microbiota restoration. | Li et al. (2023) |
| PD mice + *B. producta* | *B. producta* ($1\times10^9$ CFU/0.2 mL/day, oral gavage, for 4 weeks) | ↓ IL-1β, TNF-α, RAS, NF-κB (p65/p-p65 ratio) | ↓ Iba-1$^+$, COX-2, iNOS, ROS | ↓ RAS, ↓ p-ERK/ERK, ↓ p-p65/p65 | *B. producta* suppressed microglial overactivation and neuroinflammation via downregulating the RAS–NF-κB pathway, contributing to dopaminergic neuroprotection and improved motor function in a PD mouse model. | Liu et al. (2024) |

(Continues)

**TABLE 3** | (Continued)

| Experimental design | Treatment | Neuroinflammation | Microglial state | Microglial function | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|
| AD mice + A. rectalis | A. rectalis ($1\times10^9$ CFU/mL, gavage, 4 weeks) | ↓ IL-1β, TNF-α, COX-2, ROS, NF-κB (p65/p-p65) | ↓ Iba-1$^+$, CD11b, iNOS, ↑ TREM2 | ↑ p-Akt/Akt, ↓ p-p65/p65, ↑ TREM2 | A. rectalis supplementation alleviated AD-like pathology by suppressing microglial overactivation and neuroinflammation through the Akt/NF-κB signaling pathway. These commensal bacteria exert its effects partly via butyrate production. | Lv et al. (2024) |
| GF neonatal + Bifidobacteria | Bifidobacteria mix (~$1.1\times10^9$ CFU/dose, oral, every other day, P1–P20) | ↑ TNF-α | ↑ CD11b$^+$CD45$^{low}$, Iba-1$^+$, CD68, CD36, Macro, ameboid morphology | ↑ CD68, CD36, Msr1, LOX-1, Purkinje neuron firing ↓ VGLUT2 puncta (normalized pruning) | Early postnatal colonization with Bifidobacterium restored phagocytic gene expression, normalized pruning, and improved cerebellar circuit activity, demonstrating the essential role of gut microbiota in regulating microglial function during development | Luck et al. (2020) |
| Primary microglia + LPS/IFNγ | SCCA mix (butyrate, valerate, isovalerate, 2-methylbutyrate; 200 or 1000 μM, 1 h pretreatment) + LPS or IFNγ (100 ng/mL, 24 h) | ↓ IL-6, IL-10, NO (significant) ↓ IL-1β and TNF-α: (not significant) | ↓ Iba-1$^+$ | ↑ lipid droplet accumulation (BODIPY$^+$) ↓ NO release | SCCA mix reduced proinflammatory signaling and enhanced lipid metabolism in microglia exposed to inflammatory stimuli, indicating modulation of microglial function by microbiota-derived metabolites. | Churchward et al. (2023) |
| MDP activation in microglial cultures | MDP (0.1, 1, 10 μg/mL, 3–72 h); ± MAPK inhibitor (SB202190, 10 μM, 1 h pretreatment) | ↑ TNF-α, IL-1β, NF-κB2 (high MDP) ↓ with MAPK inhibitor | ↑ Iba-1$^+$, ↑ CD11b$^+$; ↓ CD16; ↑ nuclear p65 (NF-κB translocation) Inhibited by MAPK inhibition | Low MDP: ↑ CCL5 (linked to synaptic plasticity) High MDP: ↑ TNF-α, IL-1β MAPK inhibition: ↓ proinflammatory output and NF-κB activation | MDP induced dose- and time-dependent microglial overactivation. Low doses increased CCL5, suggesting a role in synaptic modulation; high doses triggered inflammation via MAPK and NF-κB signaling. MAPK inhibition reversed these effects, highlighting bacterial PGN as a regulator of microglial state and function. | Spielbauer et al. (2024) |

(Continues)

**TABLE 3** | (Continued)

| Experimental design | Treatment | Neuroinflammation | Microglial state | Microglial function | Key findings | Study (Author, year) |
|---|---|---|---|---|---|---|
| TBI mice + *Citrobacter rodentium* | *Citrobacter rodentium* ($10^{10}$ CFU, oral gavage, single dose at 28 days post-TBI) | ↑ IL-1β | ↑ $CD68^+$ cells | Not specifically evaluated | Enteric infection with *C. rodentium* exacerbated microglial overactivation after TBI, indicating gut-derived immune signals worsen chronic neuroinflammation. Highlights gut–brain axis involvement postinjury | Ma et al. (2017) |
| Naïve mice + FMT CUMS stress ± minocycline | FMT from CUMS mice (200 μL/day, oral gavage, 7 days) Minocycline (50 mg/kg/day, intraperitoneal, 14 days) | ↑ IL-1β, IL-6, TNF-α ↓ IL-1β, IL-6, TNF-α | ↑ $Iba\text{-}1^+$, $CD68^+$, CD11b ↓ branch length, ↑ ameboid and hyper-ramified morphology ↓ $Iba\text{-}1^+$, $CD68^+$, CD11b, restoration of branch length and morphology | ↓ $BrdU^+$, $DCX^+$, $BrdU^+DCX^+$ cells (impaired neurogenesis) ↑ $BrdU^+$, $DCX^+$, $BrdU^+DCX^+$ (restored neurogenesis) | FMT from stressed mice induced hippocampal microglial priming, evidenced by enhanced inflammatory marker expression, morphological overactivation, and impaired neurogenesis. Minocycline reversed these effects, confirming microglia as a key mediator in microbiota-driven vulnerability to stress-related pathology | He et al. (2024) |

Abbreviations: A. rectalis, Agathobacter rectalis; AD, Alzheimer's disease; ASC, apoptosis-associated speck-like protein containing a CARD; AT oil, Acorus tatarinowii oil; *B. longum, Bifidobacterium longum*; *B. producta, Blautia producta;* $BODIPY^+$, boron-dipyrromethene positive (lipid droplet marker); $BrdU^+$, bromodeoxyuridine positive (proliferating cells); C1q, complement component 1q; Caspase-1, cysteine-aspartic acid protease 1; CCL5, C-C motif chemokine ligand 5; CD11b, Cluster of differentiation molecule 11b; $CD16/32^+$, Fc gamma receptors (M1 microglial marker); $CD206^+$, Mannose receptor (M2 microglial marker); CD36, Cluster of differentiation 36; CD45, Cluster of differentiation 45; $CD68^+$, Cluster of differentiation 68 positive (microglial/macrophage marker); CFU/mL, colony-forming units per milliliter; COX-2, cyclooxygenase-2; CUMS, chronic unpredictable mild stress; DCX, doublecortin (marker of immature neurons); *E. faecalis, Enterococcus faecalis;* FMT, fecal microbiota transplantation; GD16, Gestational Day 16; GF, Germ-free; $Iba\text{-}1^+$, ionized calcium-binding adapter molecule 1 positive (microglial marker); IFN-γ, interferon gamma;, IL-1β, interleukin-1 beta; IL-10, interleukin-10; IL-17, interleukin-17; IL-6, interleukin-6; iNOS, inducible nitric oxide synthase; LOX-1, lectin-like oxidized LDL receptor-1; LPS, lipopolysaccharide; Macro, macrophage marker; MAPK, mitogen-activated protein kinase; M1, proinflammatory microglial phenotype; M2, anti-inflammatory microglial phenotype; MCAO/R, middle cerebral artery occlusion/reperfusion; MDP, muramyl dipeptide; Msr1, macrophage scavenger receptor 1; NLRP3, NOD-like receptor family pyrin domain containing 3; NF-κB (p65/p-p65 ratio), nuclear factor kappa B activation state; NO, nitric oxide; Nod2, nucleotide-binding oligomerization domain-containing protein 2; P1–P20, Postnatal Day 1 to 20; p-Akt/Akt, phosphorylated Akt to total Akt ratio; PD, Parkinson's disease; PSD95+, postsynaptic density protein 95 positive (synaptic marker); RAS, rnin–angiotensin system; ROS, reactive oxygen species; SCCA, short-chain carboxylic acids; SCFA, short-chain fatty acids; SD, sleep deprivation; SHR, sSpontaneously hypertensive rats; SYN, synaptophysin (synaptic marker); T1D, Type 1 diabetes; TBI, traumatic brain injury; TNF-α, tumor necrosis factor alpha; TREM2, triggering receptor expressed on myeloid cells 2; VGLUT2, vesicular glutamate transporter 2.

(Huang et al. 2024; Li et al. 2024; Shi et al. 2021). In addition, AT oil showed the most significant changes in microbiota abundance, compensating for the decrease in Prevotella due to ischemic stroke (Huang et al. 2024).

These studies implicated gut microbiota alterations as upstream modulators of neuroinflammatory and cognitive outcomes. In the FD model, microbiota changes preceded cognitive impairments, suggesting a causal role in triggering neuroinflammation and microgliosis. Similarly, oil treatment induced microbiota shifts, which may have contributed to its anti-inflammatory effects. These findings underscore the gut–brain axis as a key mechanism linking dietary interventions to neuroimmune modulation.

#### 3.3.3 | Fecal Microbiota Transplantation (FMT)

FMT studies highlight the gut microbiota's pivotal role in neuroinflammation, microglial state, and cognition. While microbiota from healthy or exercise-conditioned donors mitigated neuroinflammation and promoted cognitive resilience in models of stress, hypertension, and diabetes, microbiota from stressed or sleep-deprived animals exacerbated microgliosis, neuroinflammation, and cognitive dysfunction.

In CUMS rats, control microbiota transplantation decreased neuroinflammation in the prefrontal cortex and hippocampus by downregulating inflammasome activation markers (NLRP3, ASC, Caspase-1, IL-1β) and reducing IL-1β and TNF-α expression (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). Similarly, FMT from exercised animals reduced hypertension-associated neuroinflammation by lowering pro-inflammatory cytokines (TNF-α, IL-6, IL-1β) in the paraventricular nucleus (Xia et al. 2021). In T1D mice, FMT from healthy donors led to a significant reduction in IL-1β, IL-6, and TNF-α expression in the cortex and hippocampus, correlating with improved cognitive function through better performance in the MWM (Zhao et al. 2023). Additionally, while AT treatment provided neuroprotection in ischemic stroke models, as mentioned above, its benefits were independent of microbiota ischemic-related changes, as proven by FMT from ischemic rats after the middle cerebral artery occlusion (Huang et al. 2024). Rejuvenized microbiota in aged mice through cohousing them with young mice showed lower hippocampal and cortex neuroinflammation supported by a reduction in NLRP3, IL-6, Icam1, and F4/80 mRNA expression (Ma et al. 2024). Notably, not all microbiota-related neuroinflammatory effects are beneficial; it depends on the source. FMT from stressed dams to pregnant rats led to increased neuroinflammatory responses in the fetal brain, as evidenced by microgliosis (Iba-1$^+$ cell increase) across different developmental stages (P72 and P91) in the hippocampus and amygdala (Zeng et al. 2024).

FMT studies consistently show that microbiota composition influences microglial activation states. In stress models, transplantation of control microbiota reduced microgliosis (Iba-1 intensity) and inflammasome activation markers (NLRP3, ASC, Caspase-1, IL-1β) in both the prefrontal cortex and hippocampus (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). Similarly, T1D mice receiving FMT from healthy donors exhibited decreased microgliosis (Iba-1$^+$ cell increase) in both cortical and hippocampal regions (Zhao et al. 2023). Exercise-induced microbiota changes led to reduced microgliosis (Iba-1$^+$ cell increase) in the paraventricular nucleus of hypertensive rats, accompanied by morphological changes indicative of a nonreactive state (smaller cell size, longer processes) (Xia et al. 2021). In contrast, microgliosis due to FMT from stressed dams (Iba-1$^+$ cell increase) in the fetal brain of healthy litters persisted through development (P72 and P91) in the hippocampus and amygdala, and it was accompanied by a microglial morphology shifted toward reactive phenotypes (increased rod-like, ameboid, and reactive microglia, with decreased ramified microglia), indicating heightened microglial overactivation. The functional consequence of these changes was excessive synaptic pruning, as indicated by increased PSD95/Iba-1$^+$ cell colocalization and decreased SYN/PSD95 puncta colocalization (Zeng et al. 2024). Similarly, FMT from CUMS mice into antibiotic-pretreated healthy mice induced anxiety-like behavior (decreased the time in the open arms of the Elevated Plus Maze), increased hippocampal Iba-1$^+$ and CD68$^+$ cells, and upregulated inflammatory markers such as CD11b and IL-1β. These animals showed greater vulnerability to stress and inflammatory stimuli (e.g., LPS injection or additional CUMS exposure), displaying enhanced microgliosis, ameboid microglial morphology, and depressive-like behavior (He et al. 2024). Additional evidence from germ-free (GF) models further supports the critical role of the microbiota in regulating microglial subtypes and regional distribution. GF mice exhibited significant microglial transcriptomic shifts compared with the rest of the cell populations, reducing the proportion of microglia in the hippocampus while increasing it in the prefrontal cortex. The microglial signature in the GF mice was linked to anti-inflammatory and regulatory T-cell signatures (e.g., *Entpd1, Tgfb1*). Colonization of GF mice (through cohousing with specific pathogen-free mice) reversed many of these changes, restoring microglial heterogeneity and gene signatures associated with AD (*Apoe, Trem2, C1qa*) and major depressive disorder (*FKBP5, AUTS2*). The microbiota also influenced microglia–astrocyte signaling, promoting less inflammatory microglial phenotypes (Huang et al. 2023).

Cognitive function was significantly influenced by microbiota composition across the different models. FMT from healthy donors improved cognitive performance in T1D mice, as evidenced by enhanced spatial memory in the MWM (Zhao et al. 2023). AT treatment improved cognitive function in ischemic stroke models, but its effects were independent of microbiota ischemic-related changes, as proven by no changes between the AT group and AT+FMT from the ischemic rat group (Huang et al. 2024). Conversely, FMT from sleep-deprived mice also negatively impacted cognition, as recipient mice exhibited worse performance in the NOR and Y-maze tests (Li et al. 2023). GF mice displayed impaired short-term memory in the Y-maze test, which was fully restored by microbial colonization through cohousing with specific pathogen-free mice (Huang et al. 2023). Following this cohousing method for microbiota colonization, some authors showed that aged cohoused with young mice reduced their brain tauro-β-muricholic acid (TβMCA) levels, which was associated with age-related symptoms, getting a cognitive performance similar to the young group (Ma et al. 2024). These results reinforce the gut microbiome's crucial role in modulating cognitive resilience and vulnerability to stress-related impairments.

These findings underscore the therapeutic potential of microbiota-based interventions for modulating neuroimmune function and cognition across different neurological and psychiatric conditions.

#### 3.3.4 | Probiotics and Bacterial Supplementation: (Li et al. 2023)

Microbiota-targeted interventions demonstrated pro- and anti-inflammatory effects depending on the bacterial strain, host condition, and life stage. On the one hand, as anti-inflammatory strains, authors highlighted *Akkermansia muciniphila* (*A. muciniphila*), *Blautia producta* (*B. producta*), and *Agathobacter rectalis* (*A. rectalis*). *A. muciniphila* gavage supplementation ($2 \times 10^8$ CFU; 3 days/week for 4 weeks) in sleep-deprived mice restored serum SCFAs (acetate and butanoic acid) and reduced neuroinflammation supported by decreased microglial pruning (CD68_Syn_Iba-1$^+$ cells) (Li et al. 2023). *B. producta* gavage supplementation ($10^9$ CFU/day for 4 weeks) in a PD mice model (protoxin, 1-methyl-4-phenyl-1,2,3,6-tet rahydropyridine -MPTP- treatment: 30 mg/kg/day for 7 days) decreased microgliosis (Iba-1 protein expression by IHC and WB) as a marker of neuroinflammation (Liu et al. 2024). *A. rectalis* oral gavage ($1 \times 10^9$ CFU/day for 4 weeks) in AD (APP/PS1) mice decreased Aβ and pTau accumulation and down-regulated inflammatory microglial markers (Iba-1, CD11b, iNOS) while promoting TREM2 and TMEM119 in CA1 and cortex, supporting the anti-inflammatory effect of the oral supplementation (Lv et al. 2024). In this line, probiotic supplementation in drinking water ($\geq 14 \times 10^5$ CFU/mL) during gestational stress reduced offspring neuroinflammation (Iba-1+ cells) (Zeng et al. 2024). On the other hand, *Bifidobacterium* and *Citrobacter* were highlighted as proinflammatory strains. *Bifidobacterium*-colonized mice (oral gavage from P0-P20:$1.1 \times 10^{10}$ CFU/day) showed neuroinflammation in the cortex, hippocampus, and cerebellum at P4 and P20 (TNF-α mRNA expression increased) with a corresponding increase in microgliosis at P20 (CD11b + CD45$^{high}$ %) (Luck et al. 2020). *Citrobacter rodentium* infection ($10^{10}$ CFU by oral gavage at 28 days after injury) exacerbated neuroinflammation in brain-injured mice, supported by increasing lesion size and reactive microglia (CD68$^+$ cells) despite similar gut inflammation in sham animals (Ma et al. 2017).

All studies assessed microglial state, with consistent findings linking specific microbiota to either increased or suppressed activation, as measured by Iba-1, CD68, CD11b, morphology, and synaptic engulfment. *A. muciniphila* lowered Iba-1$^+$ cell counts, reduced CD68/synaptophysin and C1q/PSD95 colocalization, and protected against over-pruning in SD mice (Li et al. 2023). *B. producta* and *A. rectalis* treatments reduced Iba-1 expression in PD and AD models, respectively (Liu et al. 2024; Lv et al. 2024). In addition, *A. rectalis* enhanced functional microglial phagocytosis of Aβ without hyperactivation supported by decreasing Iba-1, CD11b, and iNOS protein expression (reactive markers), and increasing TREM2 (phagocytic marker) and TMEM119 (homeostatic microglial marker) protein expression in the brain of these supplemented APP/PS1 mice (Lv et al. 2024). Microglial state was also studied by analyzing its morphology and functionality. Probiotic treatment in gestational stress shifted microglial morphology toward a ramified, nonreactive phenotype (less rod-like reactive and ameboid shape), which suggests less overactivated, together with lower microglial pruning in the hippocampus and basolateral amygdala supported by PSD95% engulfment $^+$by Iba-1 cells (Zeng et al. 2024). *Citrobacter* infection worsened microglial state in injured mice, as shown by elevated CD68$^+$ reactive microglia in the lesion area (Ma et al. 2017). *Bifidobacterium* treatment induced a developmental shift toward ameboid, reactive microglia by P20, with an increase in phagocytic (CD68 and CD36) and scavenger (Marco, Msr1, and LOX-1) markers, suggesting premature or overactive immune function in early life (Luck et al. 2020).

Multiples of these studies reported behavioral improvements in cognition and memory following microbial supplementations. *A. muciniphila* and *A. rectalis* supplementation reversed SD/AD-induced memory impairments (NOR test for both, nesting score and Barnes maze for AD model) (Li et al. 2023; Lv et al. 2024).

Together, these studies reinforce the gut microbiota's central role in modulating neuroinflammation, microglial activity, and cognitive outcomes. Specific bacterial species like *A. muciniphila*, *B. producta*, and *A. rectalis* exhibit anti-inflammatory, neuroprotective effects, while others, such as *Citrobacter rodentium* or even early-life *Bifidobacterium*, can worsen microglial overactivation depending on timing and context. Across models of sleep deprivation, neurodegeneration, stress, and injury, modulating the microbiota offers a promising therapeutic route for preserving synaptic function and behavior through the gut–brain axis.

### 3.4 | Mechanistic Insights Into Microbiota–Microglia Interactions

A growing body of evidence reveals how specific microbial-derived metabolites (e.g., SCFAs, glutamate, butyrate, peptidoglycans) regulate microglial state, neuroinflammatory signaling, and potentially cognitive function. All studies presented here provided converging evidence that microbial metabolite pretreatment can modulate inflammatory gene expression and related signaling pathways in microglial cells (Table 2). Glutamate (0.1 mM), identified across feces, colon, hippocampus, and cortex of FMT-treated T1D mice, was shown to reduce IL-1β, IL-6, and TNF-α mRNA expression in BV2 microglia under high glucose stress (35 mM), linking metabolite shifts to a dampened inflammatory profile (Zhao et al. 2023). Sodium butyrate (NaB; 200 μM) derived from *A. rectalis* and SCFAs mix (butyrate, 2-methylbutyrate, valerate, and isovalerate) reversed inflammation in LPS-stimulated microglia (100 ng/mL), decreasing the expression of IL-1β, IL6, iNOS, COX-2, and ROS levels (Churchward et al. 2023; Lv et al. 2024). Butyrate derived from *B. producta* (200 μM) reduced RAS, p-ERK, and p-p65 expression in a PD in vitro approach (25 μM MPP 24 h stimulation), highlighting its anti-inflammatory action through the RAS–ERK–NF-κB axis (J. Liu et al. 2024). Bacterial peptidoglycan (MDP: muramyl dipeptide) from commensal gut microbiota optimal levels reveals its crucial role in healthy host neurodevelopment. MDP (10 μg/mL) treatment led to increased markers of inflammatory activation (e.g., TNFα, IL-1β, CCL3/5 mRNA

expression) while also showing a time-dependent reduction in CD16 mRNA expression, suggesting a proinflammatory dynamically regulated state. It was also shown that this inflammation state was driven by NF-κB and MAPK-p38 activation, which was blocked by pathway-specific inhibitors (Spielbauer et al. 2024). Additionally, a recently highlighted microbial-derived metabolite, TβMCA, was found to accumulate in the hippocampus and hypothalamus of aged mice and was linked to gut microbiota dysbiosis (Ma et al. 2024). Intracerebroventricular administration of TβMCA in young mice resulted in an inflammaging state of the hippocampus and cortex supported by elevated caspase-1, IL-18, IL-1β, and NLRP3 expression. These molecular changes were accompanied by behavioral impairments in the MWM and NOR, indicating that gut-derived bile acids can act directly on the brain to promote neuroinflammation and cognitive deficits (Ma et al. 2024).

Microbial metabolites also influenced both phenotypic and functional activation states of microglia. NaB pretreatment in LPS-stimulated BV2 cells downregulated genes related to neuroinflammation, microglial state, and AD biomarkers, with enrichment analysis revealing suppressed PI3K-Akt and NF-κB signaling. These results were corroborated in an AD mouse model where animals were treated with *A. rectalis* (Lv et al. 2024). BV2 cell line treated with 10 μM TβMCA for 24 h induced an increase of Iba-1 and iNOS immunostaining, suggesting an overactive state of microglia (J. Ma et al. 2024). SCFA-treated (236 μM sodium acetate +117 μM sodium butyrate) stimulated microglia (100 ng/mL LPS) in co-culture with neurons showed reduced microgliosis (Iba-1$^+$ area), fewer CD68$^+$ structures/cell, and decreased synaptic pruning (lower SYN/Iba-1 colocalization), pointing to a restored microglial functional balance. These events were linked to *A. muciniphila* supplementation in SD mice due to its effect in restoring serum SCFA concentrations (N. Li et al. 2023). Moreover, data from SCFA receptor knockout (GPR41/43 KO) mice exposed to FD revealed that the absence of SCFA signaling disrupted gut barrier integrity (downregulation of occludin and ZO-1) and led to cognitive impairment (NOR, OLT, and nesting tests). It also reduced hippocampal pCaMKII and synaptophysin levels. In contrast, SCFA supplementation in FD mice rescued gut permeability, improved cognitive performance, and reduced hippocampal neuroinflammation, confirming a direct mechanistic link between SCFAs, gut integrity, and brain function (Shi et al. 2021). In addition, the SCFA mix also induced lipid droplet accumulation in microglia (BODIPY staining), reflecting a shift in metabolic state and energy storage, potentially tied to less harmful immune activity (Churchward et al. 2023).

These studies collectively demonstrate that microbial-derived metabolites, including SCFAs (e.g., butyrate, acetate, valerate), glutamate, bile acids, and peptidoglycan, have potent effects on neuroinflammation and microglial state, often through modulation of signaling pathways like NF-κB, PI3K-Akt, RAS–ERK, and MAPK-p38. These effects are context-dependent—anti-inflammatory and neuroprotective under stress and disease, or proinflammatory at high doses or in specific immune challenges (e.g., MDP). Some studies also showed a positive impact on cognition, likely via restored synaptic plasticity, reduced microglial overactivation, and enhanced neuroimmune balance, reinforcing the gut microbiota–brain axis as a critical regulator of brain health.

### 3.5 | Gut Microbiota Modulation of Microglial Function

Most of the selected articles use FMT as the primary approach to analyze the direct effect of microbiota on microglial state and function (see Table 3 for details). A total of 14 out of 20 articles studied this topic, of which six used FMT as an approach (Huang et al. 2024; Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021; Xia et al. 2021; Zeng et al. 2024; Zhao et al. 2023). On the one hand, some articles used the FMT to further analyze the direct effects of the microbiota changes due to experimental design. Zheng et al. showed how the FMT from stressed pregnant rats (FMTs) caused a deleterious effect in normal pregnant rats, similar to the ones caused by the stress protocol itself. Regarding microbiota changes, both groups shared an increase in the abundance of *Lachnospiraceae*, *Treponema*, and *unclassified_k_norank_d_Bacteria*, suggesting that those families could be responsible for the above-mentioned effect in microglial cells. The authors showed an increase in the number and percentage of disease-associated microglia (ameboid, reactive, and rod shape) in pups from both stressed and FMTs rats independent of their developmental stage. These changes in microglial morphology were associated with their phagocytic capacity, which was elevated due to the FMT supported by an increase $^+$in PSD95_Iba-1 cells, while SYP/PSD95 puncta colocalization was decreased in the same brain area (Zeng et al. 2024). Interestingly, the authors implied in the title that the communication is bidirectional since it affirmed that microglial is necessary for getting the beneficial effect of probiotics. However, this assumption needs clarification since they did not remove microglial cells from their animal model to analyze that. However, Xia et al. used FMT to analyze whether the beneficial effect of exercise in spontaneously hypertensive rats (SHR) animal model is due to microbiota-related changes. The authors described the increased number, cell size, and length processes of microglial cells due to SHR could be rescued by exercise, which also occurs when healthy animals undergo FMT from the different experimental groups (Xia et al. 2021). Interestingly, the authors investigated the metabolite-producing bacteria associated with the FMT transplant and defined butyrate-producing bacteria as possibly responsible for the beneficial effect of exercise in the SHR model.

On the other hand, other researchers used FMT from healthy animals to rescue the animal phenotype previously affected by the disease procedure (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021; Zhao et al. 2023). Regarding the microglial state, these studies showed how the gut microbiota from healthy donors decreased inflammasome activation (NLRP3, ASC, Caspase-1 and IL-1B) and Iba-1 intensity in brain tissue. Comparing the three studies, we found similar changes in microbiota abundance that could be responsible for those beneficial effects in microglial cells. Particularly, these studies shared changes in *Alloprevotella* and *Muribaculaceae (Muribaculum* genus in Xie studies). Finally, one specific study shows the importance of the gut microbiota for rescuing the animal's phenotype through dietary supplementation such as Acorus tatarinowii oil (Huang et al. 2024). The authors showed that microbiota depletion prevents the positive AT

oil effects in microglial cells. To analyze that fact, the authors show the effect of AT oil in stroke rats with microbiota depletion compared with nondepleted stroke animals and depleted stroke animals but FMT with their own microbiota after the stroke.

Another common approach used by some articles from our collection was bacteria supplementation. Li et al. specifically supplemented sleep deprivation (SD) animals with *A. muciniphila*, a bacteria species drastically reduced by the SD model, to analyze whether it could be responsible for the changes observed in the animal phenotype. Regarding microglial changes, *A. muciniphila* counteracted the Iba-1 and iNOS increased by the SD protocol, and the synaptic pruning increased (Li et al. 2023). Similar results were obtained in a neurodegenerative mice model, where *A. rectalis* and *B. producta* supplementation counteracted most of the deleterious effects of AD and PD, respectively, including the microglial state (Liu et al. 2024; Lv et al. 2024). Aligned with this approach, Luck et al. investigated the importance of gut microbiota for microglial function during brain development. They showed how GF pups present less active microglial cells, rescued by bifidobacteria colonization, which was the most abundant during the critical neurodevelopment window (P4-P10) (Luck et al. 2020). Although these supplementations could have beneficial effects in specific environments, *A. muciniphila* and *B. producta* were increased in rats after suffering from a stroke (Huang et al. 2024) which potentially could indicate a deleterious effect of the exacerbated increase in their abundance. Further studies exploring the presence of bacteria in the brain region are needed to understand the limitation or risk of the use of these supplementations.

A couple of articles from our selection used different microglial primary cell culture treatments to elucidate the direct effect of microbiota products in microglial cells. One study, based on the increase of SCFAs and carboxylic acids (SCCA) after FMT in patients with recurrent *Clostridioides difficile* infection, used an SCCAs mix to treat microglial cells stimulated by LPS or IFN (Churchward et al. 2023). The authors find a reduction in inflammatory cytokine release by microglial cells such as IL-β1, IL-6, and TNF-α. They also showed an increase in the capacity of lipid droplet accumulation by microglial cells pretreated with the SCCAs mix. These beneficial effects were only observed with the SCCAs mix and not by any individual compounds. The other article explored the effect of bacterial peptidoglycan bioactive fragment (MDP- treatment), continuously released by commensal gut bacteria, at physiological and pathological conditions on microglial cells (primary cell culture and immortalized cell line). The authors showed that physiological doses kept microglial cells active through increased CCL5 which regulates synaptic plasticity and memory formation, and a partial activation of Nod receptor. However, higher doses of MDP caused microglial cells to increase their release of inflammatory cytokines (IL-1β, TNF-α) and fully activate Nod and NFkB signaling, triggering sustained inflammation (Spielbauer et al. 2024). To understand whether MDP acts over a specific signaling pathway, the authors used a MAPK inhibitor and obtained a partial rescue of the microglial phenotype. Although in vitro studies provide a clear connection between the direct effects of microbiota-derived metabolites on microglial states and functions, these studies cannot conclude that their outcomes could be extrapolated to in vivo situations due to the limitations regarding the difficulty of mimicking the brain environment. Further analysis with microbiota-derived metabolites specifically targeting microglial cells (e.g., microglial-targeting nanoparticles) will be needed to clarify their actual effects on microglial function.

Two interesting studies of this collection explored the bidirectional communication of the GBA (He et al. 2024; Ma et al. 2017). On the one hand, Ma et al. induced a traumatic brain injury in mice (CCI, controlled cortical impact) and showed gut dysbiosis due to the procedure, highlighting the reduction in *Bacteroidetes* and *Firmicutes*, two major beneficial bacterial phyla. To understand the bidirectional communication of the GBA, they induced a *Citrobacter rodentium* infection in sham and CCI to create a gut barrier dysfunction supported by a decrease in Claudin protein levels, which magnified the deleterious effects of CCI in microglial cells (increased number by CD68 staining). This GBA communication was further analyzed by studying GFAP staining (as an enteric glial marker) and GFAP and Sox10 mRNA expression in the colon of those animals as an indirect vagal stimulation (Ma et al. 2017). On the other hand, He et al. explored the role of microglial cells in microbiota stress-related effects in the CUMS protocol. These authors removed microglial cells using a minocycline treatment for 2 weeks (50 mg/kg) during the 3-week CUMS protocol. Microbiota abundance (increased in *Oscillospiraceae*, *Lachnospiraceae*, and *Muribaculaceae;* decreased in *Akkermansiaceae*, *Enterobacteriaceae*, *Bifidobacteriaceae* and *Lactobacillaceae*), microgliosis (Iba-1$^+$ cell) together with microglial morphology (an ameboid shape with shorter processes) changes in FMT-CUMS animals was ameliorated by the minocycline treatment, suggesting that microglial is essential for those microbiota stress-related changes (He et al. 2024).

## 4 | Discussion

This systematic review aimed to explore the role of the gut microbiota in modulating neuroinflammation through microglial activation states and its impact on brain function. The studies reviewed consistently demonstrate the existing link between gut dysbiosis and neuroinflammation via microglial functional states, contributing to brain dysfunction, which is cognitive decline, the most studied event. Fecal microbiota transplantation, bacterial supplementation, and microbiota-derived metabolites (e.g., SCFAs) seem to be potential therapeutic interventions, although most studies are needed to clarify their effects in clinical trials, considering that the analyzed studies are mostly preclinical. Indeed, most of them report only associations, with few directly establishing causality between gut dysbiosis and microglial dysfunction, highlighting the need for interventional studies using gnotobiotic models, receptor antagonists, or microglial partial ablation.

While these associations are compelling, it is critical to emphasize that correlation does not imply causation. Most reviewed studies are preclinical and descriptive, showing associations between gut microbiota composition and neuroinflammatory responses. Only a subset employed interventional designs, such as FMT or bacterial supplementation, to demonstrate partial causality. These interventions often lack longitudinal follow-up or mechanistic dissection (e.g., through microglial depletion

models or targeted microbial knock-in/out). For instance, leaky gut and BBB disruption are often inferred rather than directly measured across the studies. Therefore, longitudinal studies, causal modeling, and functional knockdown approaches are needed to move from association with actual causation, which is an essential step if diagnostic protocols or early interventions are to be developed.

Talking about gut microbiota dysbiosis, most of the studies reviewed found a decrease in Bacteroidetes, *Lactobacillus*, and *Bifidobacterium* and an increase in Proteobacteria, which have been linked to neuroinflammation by different authors (Ma et al. 2024; Shi et al. 2021; Zamudio-Flores et al. 2025). Notably, the last two have been associated with increased gut barrier permeability, which could worsen the neuroinflammatory situation during disease development. Considering that most bacterial products or segments are considered inflammatory stimuli, it is not surprising that provoking a leaky gut through gut dysbiosis could facilitate the transport of those stimuli toward the brain. In fact, some authors also found an increase in the BBB permeability in their animal models, which will lead to neuroinflammation (Huang et al. 2024; Xue et al. 2020). It cannot be assumed from the reviewed manuscripts that gut dysbiosis leads to leaky gut and BBB disruption since most of them did not explore those facts, but it could be the perfect link between both events. The FMT experiments performed by many of the articles in our selection corroborate the effect of specific microbiota changes in neuroinflammation. Although further studies will be needed to understand the direct effects of some bacteria in neuroinflammation, these FMT studies showed how an increase of *Lactobacillus, Alloprevotela, Romboustsia*, and butyrate-producing bacteria, and a decrease in *Lachnospiraceae, Treponema, Oscillibacter, Ruminisclotridium, Anaeroplasma*, and *Angelakisella* reduced neuroinflammation (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021; Xia et al. 2021; Zeng et al. 2024; Zhao et al. 2023).

It is well-known that neuroinflammation situations lead to microglial overactivation, which is also explored by this systematic review. Most of the selected manuscripts found an increase in microglial cells in mice brain tissue that occurs with inflammation. However, in vivo studies remain solely descriptive since the authors only explore a few markers of microglial overactivation (Huang et al. 2024; Liu et al. 2024; Lv et al. 2024; Ma et al. 2017; Ma et al. 2024; Spielbauer et al. 2024), and only a few articles analyzed other signs of microglial overactivation, such as morphological changes or phagocytic capacity (Li et al. 2024; Luck et al. 2020; Xia et al. 2021; Zeng et al. 2024). To better understand the activation state of microglial cells due to the neuroinflammation caused by gut dysbiosis, further experiments based on microglial isolation and RNAseq or transcriptomics together with functional analysis are needed to shed light on the mechanisms involved. In this regard, Huang et al. (2023) showed the importance of having a balanced microbiota composition for microglial state, although the study lacks functional analysis. Again, the relationship between the gut microbiota changes and microglial states remains descriptive in these studies. This need for mechanistic clarity has also been highlighted in recent literature emphasizing the importance of epigenetic and translational perspectives in the gut–microglia axis (Ben-Azu et al. 2023). Microglial depletion in those studies would shed light on the role of microglial cells in GBA communication, especially in those articles where FMT or bacteria supplementation was performed. In this regard, He et al. showed that removing reactive microglial cells with minocycline treatment compensates for the deleterious effect of FTM-CUMS, suggesting that microglial cells are essential for those microbiota stress-related changes (He et al. 2024). In addition, the in vitro studies (Churchward et al. 2023; Liu et al. 2024; Spielbauer et al. 2024) gather information about the inflammatory state and molecular pathways involved in GBA communication, with NFkB being the common signaling pathway among the studies, mainly modulated by microbiota products (Liu et al. 2024; Lv et al. 2024; Spielbauer et al. 2024). Interestingly, different studies also showed the activation of the NFkB signaling pathway in brain tissue during inflammation and microglial overactivation, which corroborates the in vitro results obtained by the other authors mentioned above-mentioned (Jia et al. 2024; Lv et al. 2024; Xue et al. 2020). The most studied microbiota products were SCFAs, which have been shown to bind different G-protein-coupled receptors on immune and glial cells (GPR41, GPR43, GPR109A) (Shi et al. 2021). A couple of studies showed the different steps of this signaling pathway, highlighting the fact that TLR4 and MyD88 are activated (Jia et al. 2024; Xue et al. 2020). Being TLR4/MyD88 key receptors activated by pathogen-associated molecular patterns in glial cells, such as peptidoglycans and LPS (Lv et al. 2024; Spielbauer et al. 2024), it is plausible that neuroinflammatory events caused by microbiota dysbiosis involve this activation pathway. However, the reviewed studies are merely descriptive without a clear direct relationship between microglial overactivation and the triggered TLR4/MyD88 signaling pathway. LV et al. also showed PI3K signaling as an alternative pathway in neuroinflammation induced by microbiota-related changes, which could induce changes in butyrate production. Another fact that corroborates the close relationship between neuroinflammation caused by microbiota changes and microglial overactivation is the NLRP3 inflammasome activation in the brain shown by different studies (Ma et al. 2024; Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021). NLRP3 is mainly expressed by microglial cells in the brain, so its activation in the brain due to microbiota changes indicates a specific role of microglial cells in GBA communication. Indeed, Ma et al. showed how TβMCA accumulation in the aged brain, a bile acid metabolized by the gut microbiota, suggests that gut dysbiosis in aged animals leads to this accumulation in the brain that triggers neuroinflammation and microglial overactivation, which was further corroborated in the in vitro study (Ma et al. 2024). While signaling components like NF-κB and TLR4/MyD88, among others, are highlighted, their functional involvement remains mostly inferential, with limited experimental validation in the reviewed studies.

The potential therapeutic implications of these articles are based on three different approaches. On the one hand, the FMT from healthy individuals could be a neuroprotective strategy based on neuroinflammation and cognitive decline rescue in the different animal models (Rao, Qiao, et al. 2021; Rao, Xie, et al. 2021; Zhao et al. 2023). Similar to this approach, using specific bacteria as a probiotic or food supplement (e.g., *Akkermansia muciniphila*, *Agathobacter rectalis*) could ameliorate the microglial activation state, reducing neuroinflammation (Li et al. 2023; Liu et al. 2024; Luck et al. 2020; Lv et al. 2024). However, this

approach needs to analyze the specific beneficial bacteria depleted or reduced in the individual for precise and successful outcomes. On the other hand, pharmacological targeting of microglial inflammatory pathways could be an interesting approach to counteract or slow down some of the deleterious effects caused by the different diseases collected in this review. NLRP3, NF-κB, and MAPK inhibitors could be developed as therapeutic strategies in that sense. However, they might be delivered directly to the brain to avoid unwanted side effects due to the importance of those signaling pathways for many biological functions. Further investigations about the microglial delivered systems (e.g., nanoparticle by nasal administration) are needed for an accurate pharmacological approach. In general, translating these findings into clinical practice presents several key challenges. First, most existing data are preclinical and correlative, lacking longitudinal or mechanistic studies to demonstrate causality between gut dysbiosis and neuroinflammatory brain outcomes. Second, the gut microbiota's interindividual variability complicates universal therapies' design, suggesting that personalized interventions will be necessary. Third, current probiotic and FMT strategies lack regulatory standardization regarding microbial composition, delivery method, and safety evaluation. Fourth, while promising results have been shown for microbial metabolites (e.g., SCFAs, bile acids), our understanding of their mechanistic effects on microglia and brain circuits remains incomplete. Fifth, even if we identify effective microbial therapies, delivering their effects across the BBB remains a technical challenge, as mentioned above. Finally, there is a need for validated biomarkers to track therapeutic response and disease progression to assess unintended consequences of microbiota manipulation.

## 5 | Future Directions

As mentioned above, most of the articles analyzed here are barely descriptive, lacking specific and detailed mechanistic studies which could shed light on the particular bacterial species and metabolites that modulate microglial function. Multiomics approaches integrating microbiota composition, its transcriptomic, and metabolomic data would improve our understanding of GBA communication and the role of microglial cells in it. On top of that, human clinical trials studying FMT, probiotics, and SCFA-based treatments in patients with neurodegenerative or psychiatric conditions would also corroborate the results of the animal models. Importantly, one critical gap in the current literature, and not captured in this systematic review, is the role of sex differences in shaping GBA communication. Sexually dimorphic responses in microglia are well documented, with evidence showing that males and females exhibit distinct microglial developmental trajectories, immune responses, and susceptibilities to neuroinflammation (O'Neill et al. 2022; Guillot-Sestier et al. 2021; Mela et al. 2022, 2023). Similarly, sex-related differences in gut microbiota composition and metabolite production have been observed (Kim et al. 2019), which may influence the efficacy of microbiota-based interventions. Unfortunately, only one of the studies included in this systematic review analyzed the different effects of aging in both males and females (Ma et al. 2024). However, the study lacks appropriate statistical analysis that clarifies possible sex dimorphism. Future studies should explicitly address sex as a biological variable, both in experimental design and analysis, to uncover potentially sex-specific therapeutic targets within the GBA framework.

## 6 | Limitations of the Reviewed Studies

The variability in animal models, microbiota analysis techniques, and behavioral assessments across studies creates heterogeneity, making it difficult to compare the results and ensure reproducibility. The included studies used a wide range of animal models, treatments, and experimental techniques, which makes it difficult to directly compare their results or combine them in a meta-analysis. Translating findings from rodent studies to human conditions remains challenging due to differences in the gut microbiota composition, immune responses, and neuroinflammatory mechanisms. Furthermore, most studies focus on short-term microbiota changes, leaving the long-term effects on neuroinflammation and cognition largely unexplored, highlighting the need for longitudinal research.

Notably, few human studies have been conducted in this area, particularly those involving human brain tissue, cerebrospinal fluid, or direct measures of microglial activity in relation to gut microbiota. Although a limited number of clinical studies have begun to explore associations between gut dysbiosis and neurodegenerative conditions (e.g., PD, AD), these are often correlative (Denman et al. 2023; Liu et al. 2023) and rarely include functional readouts or mechanistic insight. The lack of robust human datasets limits the validation of preclinical findings and the design of microbiota-targeted interventions with clinical relevance.

Moreover, the translational value of mouse model findings to human neurobiology remains unclear. While rodent studies offer valuable mechanistic insights, their immune and nervous systems do not fully mimic human complexity, particularly with respect to microglial heterogeneity, BBB function, and microbiota–host interactions. These gaps represent a major challenge in the GBA field. Future efforts must prioritize the integration of human-based research, including studies using noninvasive biomarkers, neuroimaging, patient-derived organoids, and multiomics approaches, to enhance the clinical applicability of these preclinical findings.

## 7 | Conclusion

This review highlights the critical role of gut microbiota in regulating neuroinflammation and cognitive function through microglial overactivation. Therapeutic interventions targeting gut microbiota, such as FMT, probiotics, and SCFA modulation, can potentially treat neuroinflammatory conditions. However, translating these findings into clinical applications will require further research, including: (1) Personalized medicine approaches, where interventions are tailored to an individual's unique microbiota profile and inflammatory status; (2) longitudinal and mechanistic studies, essential to establish causality and identify reliable microbial biomarkers and therapeutic targets; (3) interdisciplinary collaboration among microbiologists, immunologists, neuroscientists, and clinicians to fully unravel the gut–brain axis and develop effective, scalable microbiota-based therapies for neurodegenerative and neuropsychiatric conditions.

### Author Contributions

**Nadia Suyin Ortiz-Samur:** investigation, methodology, writing – original draft. **Akshay Kumar Vijaya:** methodology, investigation, writing – original draft. **Aurelijus Burokas:** conceptualization, writing – review and editing, validation, writing – original draft. **Virginia Mela:** conceptualization, writing – original draft, writing – review and editing, validation, funding acquisition, supervision.


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### Conflicts of Interest

The authors declare no conflicts of interest.

### Data Availability Statement

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

### Peer Review

The peer review history for this article is available at https://www.webofscience.com/api/gateway/wos/peer-review/10.1111/jnc.70154.

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