Powering up Microbiome-Microglia Interactions

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ABSTRACT

Despite extensive evidence implicating the microbiota in regulating the immune system, the precise mechanisms underlying microbial control of microglial maturation remains unclear. In a methodological *tour de force*, Erny et al. (2021) identify acetate as an essential microbiota-derived molecule driving microglial metabolic pathways and functions during healthy and diseased states.

MAIN TEXT

The gut microbiota has come to the forefront of biomedical research and emerged as a key regulator of brain neurocircuitry, physiology and behaviour (Cryan et al., 2019). The mechanisms of how gut microbes influence the brain are slowly being unraveled and involve neuronal, hormonal and immune pathways (Boehme et al., 2021; Fung et al., 2017; Nagpal and Cryan, 2021). Microbiota-derived metabolites such as short-chain fatty acids (SCFAs; acetate, butyrate, and propionate) and aryl hydrocarbon receptor ligands are the most commonly investigated signaling molecules affecting microglia, the brain's resident innate immune cells (Mossad and Erny, 2020). Microglia are instrumental in the development, function, and maintenance of brain homeostasis. Dynamic in their response to environmental signals, microglia can regulate a variety of processes across the lifespan such as synaptic pruning, neural precursor migration, neuronal survival and death, oligodendrocyte differentiation, and myelination. Consequently, it is not surprising that microglia have been implicated in a myriad of CNS disorders associated with neuroinflammation, neuropsychiatric, neurodegenerative, and neurodevelopmental disorders (Mossad and Erny, 2020).

Previously, Prinz and colleagues demonstrated that the gut microbiota modulates the maturation and steady-state function of microglia (Erny et al., 2015). They found that mice raised devoid of host microbiota (germ-free; GF), displayed an immature microglial phenotype along with altered cell numbers, morphology, and impaired immune responses. Further, supplementation with SCFAs ameliorated the defective microglial phenotype observed in GF and antibiotic-treated mice. It has also been shown that the maternal microbiota shapes microglial phenotypes by affecting chromatin landscapes and transcriptomic signatures in a sex-specific manner during prenatal development (Thion et al., 2018). Indeed, such studies propose the gut microbiota as an epigenetic regulator of microglia due to differential transcriptomic signatures identified between conventionally raised mice and those with a perturbed microbiota (Erny et al., 2015; Matcovitch-Natan et al., 2016; Thion *et al.*, 2018). However, the precise control mechanisms underpinning microbiota-microglia interactions remained previously unresolved.

In this issue of *Cell Metabolism*, Prinz and colleagues now provide an exciting mechanistic update on the role of the gut microbiota in governing microglial maturation and function (Erny *et al.*, 2021) (Figure 1). The authors studied the transcriptional and epigenomic regulation of immature microglia in GF mice and found that the altered gene expression is epigenetically imprinted by the enrichment of acetylation and methylation marks (H3K4me3, HK3K9ac) on metabolic genes. Subsequently, the authors profiled the metabolic state of *ex vivo* (FACS)-isolated microglia from GF and control mice, identifying seven metabolites associated with mitochondrial mass, and a dysfunctional respiratory chain that differed significantly. To address this further, a comprehensive functional analysis of microglial mitochondria was performed. They demonstrate that the host microbiota selectively controls Complex II (CII) of the electron transport chain, displayed by diminished CII function in GF mice compared to controls. Interestingly, metabolite profiling identified oxaloacetate, a competitive inhibitor of CII, as a significantly increased metabolite in GF-isolated microglia, supporting the concept that oxaloacetate could be responsible for CII inhibition and increased reactive oxygen species (ROS) production. Indeed, this is an interesting finding considering the long proposed predominant endosymbiotic theory; a theory in which mitochondria are the hosts bacterial ancestors.

Advancing previous work, the authors identify that the SCFA acetate is the primary bacterial-derived molecule responsible for modulating microglial maturation and function in GF mice. The authors found acetate concentrations to be significantly reduced in the brain and serum of GF mice and demonstrated that supplementation of acetate restored microglial fitness by fueling intermediates of the tricarboxylic acid cycle, and therefore restored GF-associated deficits in CII function. Elegantly the authors tracked gut-derived ^{13}C-acetate and found that it can traffic to the brain where it is metabolized by microglia and other CNS cells. Acetate, present in mitochondria in the form of acetyl-coenzyme A, serves as a carbon donor essential for cellular homeostasis. The authors speculate that GF mice in demand for acetate convert citrate into acetyl-coenzyme A resulting in the overproduction of oxaloacetate and subsequent inhibition of CII function.

Earlier reports had implicated the gut microbiota as a modulator of amyloid beta (Aβ) deposition in mouse models of Alzheimer's disease (AD) (Harach et al., 2017; Mezo et al., 2020). To identify if microbial-derived SCFAs mediate these effects via microglia, Prinz et al., orally administered acetate to male GF and conventional 5xFAD mice through drinking water for 8 weeks. Interestingly, the authors found that supplementation of acetate to GF 5xFAD animals affected microglial metabolic state, enhanced microglial maturation, and suppressed phagocytosis of Aβ depositions. Erny and colleagues also demonstrated that acetate supplementation induced proinflammatory cytokines,

which has been previously shown to attenuate microglial phagocytosis in the presence of Aβ. Interestingly, the authors did not observe CII dysfunction in acetate treated GF 5xFAD mice as seen in healthy GF housed mice. However, the authors speculated that this may be because of oxaloacetate being used for the generation of glucose and other essential metabolites required for ATP generation in this specific mouse model of AD.

While these encouraging findings offer novel insights into the mechanistic relationship between the bacterial-derived metabolite acetate and host microglial maturation, function and metabolic properties under steady-state and pathological conditions, their discoveries raise questions for future investigations. Firstly, are the mitochondrial changes observed in GF mice responsible for the microglial morphology and functional deficits? Secondly, mice deficient in free fatty acid receptor 2 (FFAR2) also display an immature microglia phenotype as observed in GF mice (Erny et al., 2015). Could FFAR2 signaling be indirectly involved in microbiota-microglia interactions? The activity of CII of the respiratory chain plays a role in initiating proinflammatory responses. Consequently, diminished CII activity may account for dampened immune responses observed in GF mice. Further investigations should evaluate the impact of immune insults on host commensals and subsequent effects on microglia metabolic states. Indeed, microglia are essential cells involved in a variety of processes, and deficits in microglia induced by a perturbed microbiota could also elicit dysfunction in neighboring cellular communities. It remains to be seen if the metabolic changes induced by a perturbed microbiota alter microglial function and subsequently affect disease onset and pathophysiology. Further insights into the long-term effects and identification of bacteria responsible for aberrant acetate production will enable the identification of potential therapeutic targets. Future studies will need to evaluate the potential impact of sexual dimorphic effects and the translatability of their findings to human disease. Although many questions remain to be answered, this powerhouse study advances our mechanistic understanding of how the gut microbiota regulates metabolic fitness of microglia and offers an exciting avenue for developing non-invasive microbiota-specific therapeutic targets that modulate microglia in CNS associated diseases.

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DECLARATION OF INTERESTS

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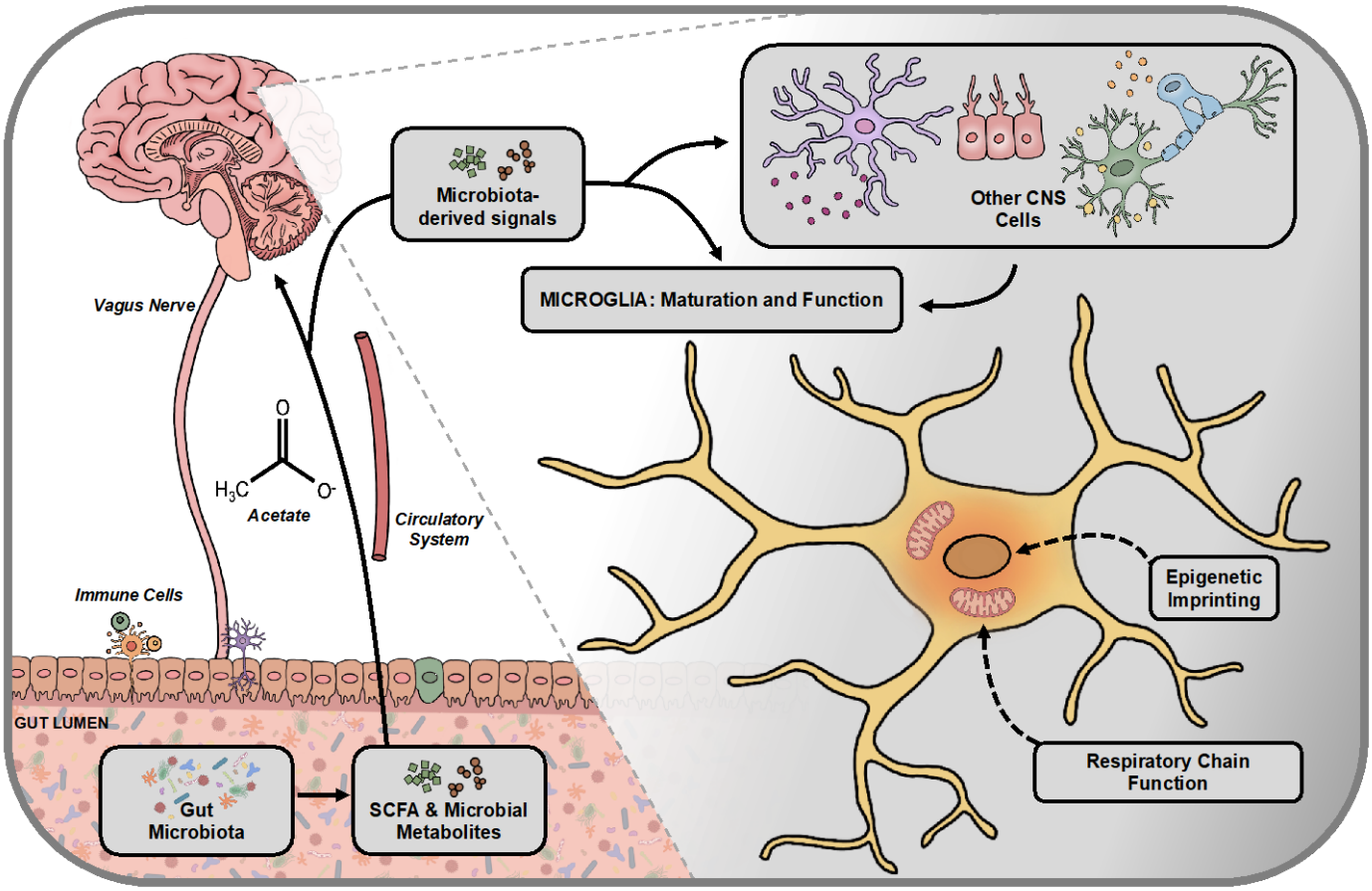

Figure 1: Overview of microbiota-derived signaling molecules and their role in regulating microglial maturation and function during health and disease.

New evidence suggests that the microbial-derived SCFA acetate is a key regulator of microglial metabolic pathways and function. It reaches the brain, perhaps via the circulation, where it is metabolized by microglia and other CNS cells. Subsequently, acetate influences histone methylation marks on genes associated with microglial proliferation, morphology, activation, and metabolic functions. Additionally, microbiota-derived acetate influences microglial fitness by fueling intermediates of the tricarboxylic acid cycle. Further understanding of the mechanistic role of acetate and other microbiota-derived signaling molecules will enable the development of novel therapeutic interventions targeting the gut microbiota for the treatment of microglial-associated disorders.