Title	More than just a number: the gut microbiota and brain function across the extremes of life
Authors	Nuzum, Nathan D.;Deady, Clara;Kittel-Schneider, Sarah;Cryan, John F.;O'Mahony, Siobhain M.;Clarke, Gerard
Publication date	2024
Original Citation	Nuzum, N.D., Deady, C., Kittel-Schneider, S., Cryan, J.F., O'Mahony, S.M. and Clarke, G. (2024) 'More than just a number: the gut microbiota and brain function across the extremes of life', Gut Microbes, 16(1), 2418988 (42pp). https://doi.org/10.1080/19490976.2024.2418988
Type of publication	Article (peer-reviewed)
Link to publisher's version	https://www.tandfonline.com/doi/full/10.1080/19490976.2024.2418988 - 10.1080/19490976.2024.2418988
Rights	© 2024, the Author(s). Published with license by Taylor & Francis Group, LLC.This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial License (http://creativecommons.org/licenses/by-nc/4.0/), whichpermits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited. The terms on which this article has beenpublished allow the posting of the Accepted Manuscript in a repository by the author(s) or with their consent. - http://creativecommons.org/licenses/by-nc/4.0/)
Download date	2026-08-30 07:33:43
Item downloaded from	https://hdl.handle.net/10468/16838

University College Cork, Ireland
Coláiste na hOllscoile Corcaigh

Taylor & Francis
Taylor & Francis Group

Gut Microbes

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/kgmi20

More than just a number: the gut microbiota and brain function across the extremes of life

Nathan D. Nuzum, Clara Deady, Sarah Kittel-Schneider, John F. Cryan, Siobhain M. O'Mahony & Gerard Clarke

To cite this article: Nathan D. Nuzum, Clara Deady, Sarah Kittel-Schneider, John F. Cryan, Siobhain M. O'Mahony & Gerard Clarke (2024) More than just a number: the gut microbiota and brain function across the extremes of life, Gut Microbes, 16:1, 2418988, DOI: 10.1080/19490976.2024.2418988

To link to this article: https://doi.org/10.1080/19490976.2024.2418988

Published online: 20 Nov 2024.

GUT MICROBES
2024, VOL. 16, NO. 1, 2418988
https://doi.org/10.1080/19490976.2024.2418988

Taylor & Francis
Taylor & Francis Group

REVIEW

OPEN ACCESS Check for updates

More than just a number: the gut microbiota and brain function across the extremes of life

Nathan D. Nuzum [a], Clara Deady [a,b], Sarah Kittel-Schneider [a,c], John F. Cryan [a,b], Siobhain M. O'Mahony [a,b], and Gerard Clarke [a,c]

[a]APC Microbiome Ireland, University College Cork, Cork, Ireland; [b]Department of Anatomy and Neuroscience, University College Cork, Cork, Ireland; [c]Department of Psychiatry and Neurobehavioural Science, University College Cork, Cork, Ireland

ABSTRACT

Understanding the interrelationship between the gut microbiota and host physiology, although still in its relative infancy, has taken important steps forward over the past decade. In the context of brain disorders including those characterized by neurodevelopmental and neurodegenerative changes there have been important advances. However, initially research involved correlational analyses, had limited translational scope, and lacked functional assessments. Thus, largescale longitudinal clinical investigations that assess causation and underlying mechanisms via in depth analysis methods are needed. In neurodegeneration research, strong causal evidence now links the gut microbiome to Alzheimer's (AD), and Parkinson's Disease (PD), as supported by human-to-animal transplantation studies. Longitudinal interventions are being conducted in AD, PD, amyotrophic lateral sclerosis, Huntington's disease, and multiple sclerosis. Neurodevelopmental research has also seen a boon in microbiome-related clinical research including in autism, Attention-deficit/hyperactivity disorder, and schizophrenia, which is confirming prior animal model work regarding the key time-windows in the gut microbiome important for infant cognition. While recent research advances represent important progress, fundamental knowledge gaps and obstacles remain. Knowing how and why the gut microbiome changes at the extremes of life will develop our mechanistic understanding and help build the evidence base as we strive toward counteracting microbial missteps with precision therapeutic interventions.

ARTICLE HISTORY
Received 28 May 2024
Revised 24 September 2024
Accepted 14 October 2024

KEYWORDS
Neurodevelopment; neurodegeneration; gut bacteria; gut-brain-axis; early-life; older adult

1 Introduction

Over the course of the past decade, gut microbiome research has continued to provide a greater understanding of the important connections between the gut and the brain. This bidirectional communication network, now conceptually reframed as the microbiota-gut-brain axis,[1] has further benefited from continuing advances in next-generation sequencing technologies,[2] and the associated development of superior analysis via more advanced bioinformatic pipelines.[3,4] The initial focus in the field was based on characterizing the gut bacteria that were present, be it enriched or depleted (assessing diversity and composition),[5] how they differed between different populations/regions,[6,7] across aging,[8] or differences in gut bacterial composition between healthy individuals and in various disease states.[9–12] While this research broadly implicated the relevance of gut bacteria for different disease states or across the lifespan, from early life to older adulthood, this overreliance on compositional and correlational observations has been a common critique of the field.[13–15] These compositional assessments have now been complemented with causal and mechanistic research. For example, shotgun metagenomics is now favored over 16S rRNA sequencing, sampling of metabolic data over solely microbial composition is more common, as are larger sample sizes, and mechanisms of host microbe dialogue have been interrogated via interventional, longitudinal, and translational research to address the question of causality of the gut microbiome in neurodevelopment, healthy aging, and neurodegeneration (Figure 1).

These advances have been facilitated by guidelines for consistent and transparent reporting of gut microbiome results and preclinical studies (e.g. STORMS checklist and GRAFT guidelines),[18,28] open access platforms and methods of analyzing metagenomic

CONTACT Gerard Clarke g.clarke@ucc.ie APC Microbiome Ireland, Biosciences Institute, Biosciences Research Institute, College Rd, University College , Cork, T12 YT20 Ireland

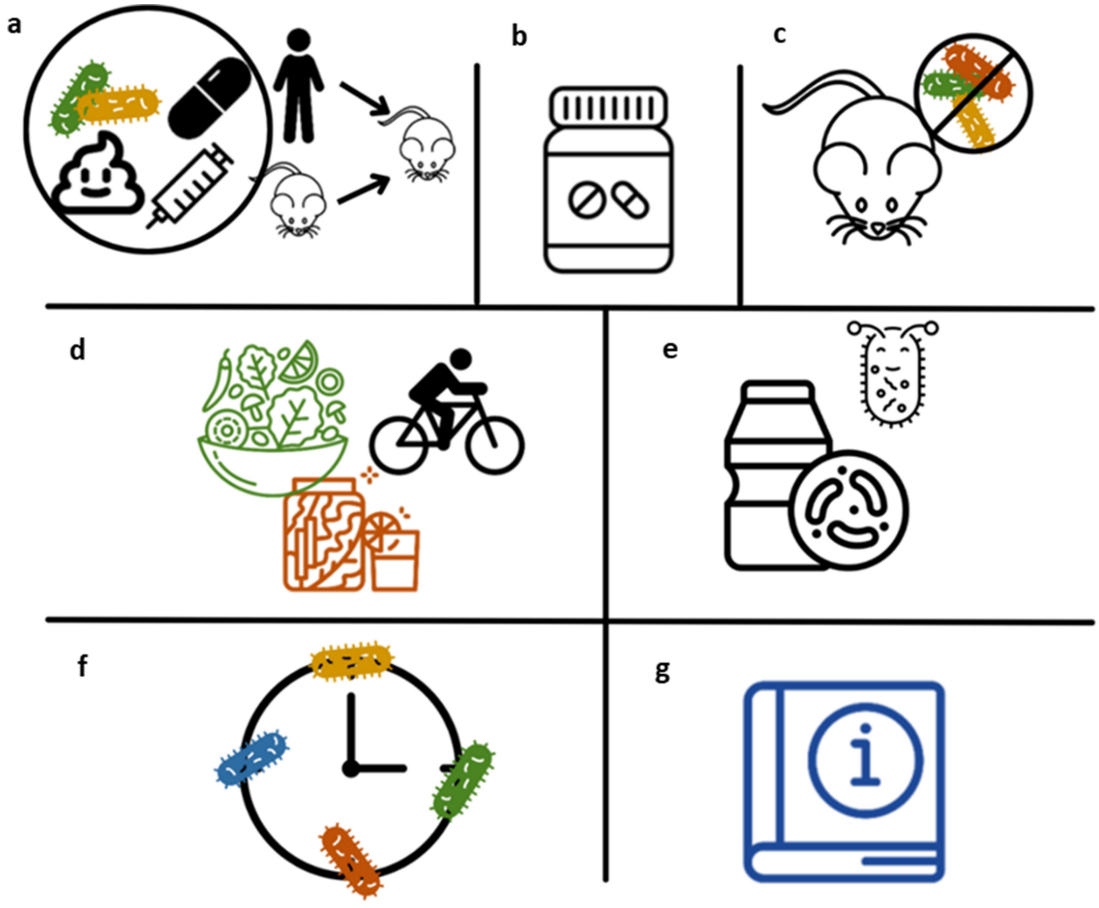

Figure 1. Descriptions of various modalities to assess causality in gut microbiome research, including (a) FMT in humans and animal research,[16–19] (b) antibiotics to 'knock-out' the gut microbiome and (c) using specific pathogen/germ free animals.[20] Various interventions modulate the gut microbiome, including (d) diet and/or fermented foods,[21] and exercise,[22] or (e) probiotic, prebiotic ("a substrate that is selectively utilized by host microorganisms conferring a health benefit"[23]) or synbiotics ("a mixture comprising live microorganisms and substrate(s) selectively utilized by host microorganisms that confers a health benefit on the host".[24]) New and emerging analysis techniques include (f) assessing circadian rhythms of the gut microbiome[25,26] (g) along with field guides for gut microbiome analysis [3,4] and methods for assessing functionality of the microbiota-gut-brain axis,[27] and reporting guidelines[28] to create consistency across the gut microbiota field. NB: current evidence supports the use of FMTs for treating *C.diff* infections, while for any other disease/disorder it is considered an investigational procedure.[45] Strict guidelines should be adhered to when conducting FMT in clinical contexts as there are some safety concerns and considerations, with some severe adverse events having occurred previously[46,47]

sequences,[29] and accessible guides providing a framework for how to statistically assess the gut microbiome.[3,4] Altogether, these enable researchers to better understand, better plan, and better implement informative research about the gut microbiome. Furthermore, updates to statistical and bioinformatic approaches that focus on functionality,[27,30] have allowed the field to move past 'who is there' (composition), and facilitate answering the questions of 'what are they doing?' or 'why are they important?'. Regarding experimental studies, the use of interventions in the form of pre- or probiotics,[31] fecal microbiota transplants (FMT),[19] or lifestyle factors which are known to modulate the gut microbiome such as diet,[32] or exercise,[33] which are also implicated in healthy aging and neurodegeneration,[34] have enabled the inference of causal connections. Finally, longitudinal research measuring how the gut microbiota fluctuates over time at an individual level have begun to help answer the 'chicken-and-egg' problem of whether the gut microbiome is influencing disease/development, or if disease/development affects the gut microbiome. While the genetic risk(s) for various neurodevelopmental disorders and neurodegenerative diseases will not be focused on presently, it is important to contextualize the potential role of the microbiota-gut-brain axis in relation to genetic risk, and to consider how genetic and environmental risks,

in concert with the microbiota-gut-brain axis, may modulate disease states and severity.

In relation to early life, the microbiome has seen much research scrutiny in many neurodevelopmental disorders such as autism spectrum disorder (ASD), attention-deficit hyperactivity disorder (ADHD), and schizophrenia (as a neurodevelopmental mental disorder in the broader definition). Altered maternal gut microbiomes, or disruptions to the infant gut microbiome, have both been linked to an increased risk of developmental disorders.[35,36] While this does not explain why those affected from these disorders have a different microbial profile, some interventional research is beginning to close this research gap. Understanding the gut microbiome changes across neurodevelopment, and how they may be causally related to the etiology of these disorders is yet to be fully elucidated.

A greater understanding of causality and the role of the gut microbiome has also begun to be elucidated in terms of healthy aging and neurodegenerative research. Primarily, this has related to healthy/unhealthy aging involving the investigations of chronological and biological aging, and a large focus on Alzheimer's,[37,38] and Parkinson's disease research.[10,39,40] Less research has been conducted on additional neurodegenerative conditions, e.g., Multiple Sclerosis (MS), Huntington's disease (HD), and Amyotrophic lateral Sclerosis (ALS), however these fields have been expanding in recent years.

In this review, we highlight how the neurodevelopment and neurodegeneration fields have progressed in the last decade focusing both on advances in basic research and outline the clinical evidence linking the gut microbiome to healthy/typical development and aging, along with links to neurodevelopmental disorders and neurodegenerative diseases. The environmental risks, and shared mechanisms of action across neurodevelopmental and neurodegenerative conditions (Figure 3), will be addressed.

Info box 1: Technological, statistical and experimental techniques used in the basic science, pre-clinical and clinical studies of the microbiome. Rather than only looking at the gut microbiome and 'who is there', more recent papers have begun to implement 'multi-omics' where multiple methods are employed to better understand the mechanisms, such as by Morton et al. who investigated metagenomic and metabolomic data across various studies.[41] Multi-omics involves the integration of multiple dataset types including:

- Metagenomics: Assessment of the structure and functions of the bacterial genes that are present. May include inferred functionality of 16S rRNA sequencing via PICRUSt2,[42] or direct assessment via whole genome shotgun sequencing [29], and the use of gut metabolic or gut brain modules (GMMs and GBMs).[27,30,43]
- Metatranscriptomics: Focuses on what genes are expressed by the entire microbial community and allows for investigation of not just the functional potential, but the active functional profile of a microbial community. As an example, the bioBakery 3 platform allows investigation of not just the metagenome, but also the metatranscriptome.[29]
- Metaproteomics: An umbrella term covering various approaches of studying all the proteins in microbial communities and microbiomes from environmental sources. Metaproteomics goes past Metatranscriptomics by giving information about whether transcribed genes result in protein translation. See Li et al. for more details and examples.[44]
- Metabolomics: The study of small molecules, i.e., metabolites, within cells, biofluids, tissues or organisms, that are produced via bacteria. The metabolites and their interactions within a biological system/host is termed the metabolome. Common methods for assessing the metabolome include mass-spectrometry or nuclear magnetic resonance (NMR) spectroscopy.[4]

2 Early-life and the gut microbiota

2.1 Neurodevelopment

Across the duration of pregnancy, the fetal brain is continuously developing to soon face the challenges of postnatal living and beyond. The very early folding of the neural tube, the precursor of the central nervous system, aids in aligning correct neuronal patterning and differentiation and is essential for life.[48] If this is not fully sealed neural tube defects will form, leading to neurodevelopmental consequences for the fetus,[48] including spina bifida occulta, meningocele, or anencephaly.[49] Disruption to development in later pregnancy also leads to complications. While most growth and drastic morphological change occurs within the first 20-weeks, specific fine tuning, including neuronal migration, myelination, and pruning, occurs later in pregnancy.[50–52] Complications during these processes may be associated with cognitive and behavioral neurodevelopmental complications, including ASD and schizophrenia.[53,54] These neuronal and behavioral changes occur in parallel with significant remodeling of the maternal gut microbiome across each trimester.[55–57] Over the last decade the involvement of the microbiome has been investigated in relation to neurodevelopment, particularly regarding ASD, schizophrenia, and ADHD, and a newer body of research now strongly supports earlier indications that the microbiome and neurodevelopmental trajectories are intricately linked.[35]

During this period, important evidence has emerged linking the microbiome and neurodevelopment (See Table 1 for a summary of several key studies and the characteristics of each). It is important to note however that there are numerous approaches to microbiome testing, with techniques changing and improving overtime. The time of sample collection or the behavioral and milestone assessments may also differ between studies, and this should be considered when comparing various research outputs. Alterations seen in the gut microbiome during specific time windows have been linked with contemporaneous and later time point assessments of infant temperament, with correlations noted between increased relative abundances of *Bifidobacteria and Streptococcus*, with an overall more positive emotionality and an increased abundance of *Akkermansia* linked to more social behavior.[88,89] Conversely, microbial changes have also been associated with more negative behavioral outcomes, including impaired stress coping. Increased abundances of *Bifidobacteria and Parabacteroides* has been linked to increased negative reactivity, along with reduced abundance of *Prevotella*.[90] Interestingly, decreased abundances of *Prevotella* has also been implicated in adverse behavioral outcomes, notably internalizing problems.[91] Separately, decreased relative abundances of *Bifidobacteria, Lactobacillus, and Haemophilus* have been linked with increased fear bias, which can be representative of emotionally directed attention.[92] Cognition has also been linked to differential microbial profiles in infancy.[93]

Specific microbiome windows under consideration have expanded to include the maternal microbiome in preconception, pregnancy, at birth, in the early postnatal period and the infant's microbiome in the context of how these windows shape the long-term health and well-being of the infant (See Figure 2). Some major factors influencing the microbiome will be discussed in the following sections, including maternal diet, antibiotic use, and breastfeeding and birth mode are also known to shape neurodevelopment, mediated by the gut microbiome. Subsequently, the gut microbiome and specific neurodevelopmental conditions will then be discussed.

2.2 The maternal microbiome: From preconception to birth

The presence of microbes in the womb has long been disputed, however current research indicates that the placenta and the amniotic fluid is most likely sterile.[94,95] Since there is no direct transfer from the maternal microbiome in utero, all initial colonization occurs during the birth process and postnatal life. Despite this, it is widely accepted that the maternal microbiome plays a key role in neurodevelopment which will be discussed in detail. It has been suggested that the fetus is influenced indirectly by microbial metabolites produced by the mother.[96] These metabolites may cross the blood-placenta-barrier,[64] and then influence the immune response of the fetus.[97] The role that maternal metabolites have in neurodevelopment has been demonstrated in both animal,[64] and human studies.[98] The emergence of the paternal gut microbiome helps to further our understanding of potential mechanisms. A study examining the disrupted gut microbiome of fathers revealed that the offspring were at a greater risk for growth restriction and mortality,[99] and recent in mice suggests depletion of the paternal microbiome can induce intergenerational effects on offspring behavior and physiology.[100] Authors hypothesize that the gut microbiome may interact with male germ cells, whether directly or indirectly, and disruptions to the gut-germline axis may negatively affect the placenta. It is likely these changes are occurring due to epigenetic alterations during spermatogenesis.[101] Between this, and the constant presence of the maternal microbiome, the gut health of both parents is key for shaping the long-term health of the fetus. Additional factors shaping the maternal and infant microbiome are discussed below.

2.2.1 Diet during preconception and pregnancy

Research links elevated homocysteine levels with poor psychomotor development,[102] and folic acid, which aids in closure of the neural tube, to beneficial verbal and cognitive development.[103,104] Additionally, overconsumption of highly processed foods is associated with lower infant developmental

Table 1. Summary of research studies and key characteristics of each research study in relation to early-life/neurodevelopment and the gut microbiome.

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention/ Exposure	Outcomes
Bhagavata Srinivasan et al.[58]	Sprague-Dawley rats	Female (mothers) + offspring (male/ female)	Maternal obesity/western diet and exercise	6-weeks of diet then 10-days of exercise with mating occurring after 10 days of exercise - Control diet + sedentary (C_S) - Control diet + Exercise (C_E) - Obesogenic diet + sedentary (O_S) - Obesogenic diet + exercise (O_E)	- Maternal exercise (C_E) decreased bacterial alpha-diversity of offspring - Maternal obesogenic diet decreased alpha-diversity irrespective of exercise - Clostridium sensu stricto, Clostridium_XlVa, Bacteroides, Blautia and Lactobacillus were enriched in obesogenic sedentary group compared to control sedentary group
Leyrolle et al.[59]	CD1 mice	Female (mothers) + offspring (males only)	Maternal immune activation (MIA) and maternal omega-3 deficiency	Upon males introduced to mating cage, Omega-3 deficient (DEF), and omega-3 sufficient (SUFF) diet was given for 48 hours. At embryonic day 17 pregnant females given saline (control), or LPS injection (to induce MIA)	Low maternal Omega-3 intake worsens maternal immune activation-induced gut dysfunction. MIA DEF mice at adulthood displayed learning deficits At phyla, family, and genus levels diet affected gut bacterial relative abundance
Sanguinetti et al.[60]	B6129SF2/J mice	Female (mothers) + offspring (male/ female)	Maternal high-fat diet	Normal diet-fed mothers (NDm) and high-fat diet-fed mothers (HFDm). Mating occurred after 3-months of diet. Diet continued through gestation and lactation.	In offspring at 1- and 6-months gut microbiota compositional differences were present between diet groups
Leclercq et al.[61]	BALB/c mice	Female (mothers) + offspring (male/ female)	Prenatal and early postnatal antibiotic exposure	One week before delivery pregnant females were treated with either drinking water (control), penicillin V (AB) or penicillin V and *Lactobacillus rhamnosus* JB-1 (AB/JB1), until pup weaning (postnatal day 21)	Penicillin had lasting effects on gut microbiota, increased cytokine expression in frontal cortex, modified blood–brain barrier integrity and altered behavior, irrespective of sex
Champagne-Jorgensen et al.,[62]	BALB/c mice	Female (mothers) + offspring (male/ female)	Prenatal antibiotic exposure	Low-dose penicillin given in water from gestational day 15 to birth (approx. 6 days), control group received water no penicillin in water	Prenatal penicillin exposure induced anxiety-like behaviors for female mice, and abnormal social behavior for males Altered microbiota composition present for both sexes
O'Connor et al.[63]	C57BL/6 mice	Female (mothers) + offspring (male/ female)	Postnatal antibiotic exposure	One day following birth, weaning mothers received either: Vehicle, Penicillin V, or antibiotic cocktail via drinking water for 7-days	Early life exposure to maternal antibiotics altered anxiety, social, and cognitive behaviors. Antibiotic cocktail broadly caused greater effects than single antibiotic
Vuong et al.[64]	C57Bl/6 J mice	Female (mothers) + offspring (male/ female) reared as SPF or GF	Maternal microbiome influence on prenatal neurodevelopment	Four groups: animals reared without microbes (GF), animals with depleted microbiome (Antibiotic [ABx] treated from before conception to E14.5), conventionally colonized (SPF) animals (via SPF bedding at E14.5 till gestation and offspring had SPF bedding until behavioral testing), and colonization of ABX dams with Clostridia-dominant spore-forming (Sp) bacteria.	Embryos from ABX and GF dams had reduced expression of genes related to axonogenesis, impaired outgrowth of thalamic axons Colonization of microbiome depleted dams (Sp) prevented abnormal fetal brain gene expression and axonogenesis.
Lynch et al.[65]	NIH Swiss mice	Female (mothers) + offspring (male/ female)	Antibiotic exposure during either; postnatal (P2-9), pre-weaning (P12-18) or post-weaning (P21-27)	Litters randomized to receive antibiotic cocktail, or 0.9% saline, for either the early postnatal (P2-9), pre-weaning (P12-18), or post-weaning (P21-27) periods.	The early-life antibiotic exposure elicited sex and time-dependent physiological effects. Antibiotic exposure, particularly in weaning period, elicited cecal microbiota disruption into adolescence. Microglial morphology was also altered by early-life antibiotic exposure

(*Continued*)

Table 1. (Continued).

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention/ Exposure	Outcomes
Kayyal et alo.[66]	BALB/c mice	Female (mothers) + offspring (male/ female)	Postnatal antibiotic exposure and concurrent probiotic administration	Intervention started at postnatal day 14. Three groups: antibiotic group, antibiotic + *L. rhamnosus* JB1 and control (fed PBS only)	Male mice exposed to antibiobics developed long-term social behavior changes. All changes seen in brain, behavior and immune cells that were associated with antibiotic exposure, were absent in the antibiotic + *L. rhamnosus* JB1 group.
Gur et al.[67]	C57/Bl6 mice	Female (mothers) + offspring (female only [males reported in separate study])	Maternal prenatal stress	Pregnant females randomly assigned to stressed or non-stressed control group. Stressed group had restraint stress between embryonic day 10–16 for 2 h between 09:00 and 12:00. Offspring behavioral testing occurred between P60-P70.	Prenatal stress associated with fecal microbiota changes in pregnant females. BDNF was decreased from prenatal stress in utero and in adulthood. Anxiety and cognitive changes accompany microbial, IL-1β, and BDNF alterations
Gur et al.[68]	C57/Bl6 mice	Female (mothers) + offspring (male only [females reported in separate study])	Maternal prenatal stress	Pregnant females randomly assigned to stressed or non-stressed control group. Stressed group had restraint stress between embryonic day 10–16 for 2 h between 09:00 and 12:00. Offspring behavioral testing occurred between P60-P70.	Males exposed to prenatal stress had reduced social behavior in adulthood, neuroinflammation was present, along with decreased serotonin metabolism in adulthood. Males exposed to prenatal stress had reduced commensal microbes (i.e., Bacteroides and Parabacteroides) compared to non-prenatal stressed group
Nicolas et al.[69]	Sprague-Dawley rats	Offspring (female only)	Postnatal stress (Maternal separation)	Two groups: maternal separation from postnatal day 2–12 for 3 h/day, or non-separated group	Maternal separation altered LPS-induced inflammatory response in plasma of juvenile rats, enhanced LPS-induced increase in IL-1β in the ventral hippocampus and maternal separation attenuated an LPS-induced increase
Collins et al.[70]	Sprague Dawley rats	Offspring (male only)	Maternal separation/ postnatal stress + milk fat globule membrane administration	At postnatal day 0, litters randomly assigned to maternally separated (separated at postnatal day 2 to postnatal day 12 for 3 h/day) or non-separated groups. Non-separated and separated groups split into control diets or diets + milk fat globule membrane	Early life stress alters changes in bejhaviour and visceral sensation. Early life stress-induced visceral pain was reversed by milk fat globule membrane
O'Mahony et al.[71]	Sprague Dawley rats	Offspring (male only)	Maternal separation/ postnatal stress + milk fat globule membrane administration and prebiotic (GOS and PDX)	At postnatal day 0, litters randomly assigned to maternally separated (separated at postnatal day 2 to postnatal day 12 for 3 h/day) or non-separated groups. Non-separated and separated groups split into control diets, diets + milk fat globule membrane, diet + prebiotic, or diet + milk fat globule membrane and prebiotic at postnatal day 21.	MS-induced visceral hypersensitivity was ameliorated by MFGM and to greater extent with the combination of MFGM and prebiotic blend. Spatial learning and memory were improved by prebiotics, MFGM and with them combined. The combination of milk fat globule membrane and prebiotic reduced the long-term impact of maternal separation on a marker of myelination in the prefrontal cortex.
McVey Neufeld et al.[72]	Sprague-Dawley rats	Offspring (male only)	Maternal separation/ postnatal stress + prebiotics and Lactobacillus rhamnosus GG (probiotic)	At postnatal day 0, litters randomly assigned to maternally separated (separated at postnatal day 2 to postnatal day 12 for 3 h/day) or non-separated groups. At postnatal day 21 animals randomized into control diet + Lactobacillus rhamnosus GG, control + water (no probiotic) or diet with prebiotics + Lactobacillus rhamnosus GG, or diet with prebiotic and water (no probiotic)	Diet containing prebiotic and probiotic attenuated early-life maternal separation effects on anxiety-like behavior and hippocampal-dependent learning.

(*Continued*)

Table 1. (Continued).

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention/ Exposure	Outcomes
Morais et al.[73]	NIH Swiss mice	Offspring (male only)	C-section, co-housing and probiotic/prebiotic administration.	Two groups: pups born via vaginal birth and a group born via C-section. A third control group to control for the effects of fostering was included (i.e., cross-fostered vaginal delivery). C-section born mice were either exposed to probiotic (*Bifodobacterium breve M16V*), prebiotic (shot chain GOS and long chain FOS) starting from birth and throughout experiment. Control vaginally born mice were given water.	C-section altered *Bifidobacterium spp*. Abundance in early-life C-section born mice had behavioral deficits through lifespan, and co-housing these mice with vaginally born mice corrected social deficits. The probiotic or prebiotic mixture also improved behvaiour in C-section born mice.
Sharon et al.[74]	C57BL/6 J mice	Male and female mice undergoing FMT	Gut microbiome transplant from ASD (or neurotypical) individuals to mice	Mice received FMT from either neurotypical donors, or ASD donors. FMT delivered via single oral gavage	FMT from ASD donors elicited impaired social communication and interaction behaviors in mice. Microbiome profiles of mice who had FMT, showed that specific bacterial taxa and metabolites may modulate ASD-like behaviors.
Sgritta et al.[75]	C57BL/6 J (Germ free), Shank3B+/– mice, DAT-cre mice, and BTBR mice	Male mice used for mouse models only	ASD mouse model, and treatment with	PBS (vehicle) or *L.reuteri* were added to drinking water of mouse models and treated water was consumed ad libitum for treatment period	L. reuteri treatment rescues social deficits in several ASD mouse models (including in germ free mice) L. reuteri also reverses social deficits via the vagus nerve
Dervola et al.[76]	Spontaneously hypertensive rats and Wistar Kyoto rats	Male and female rats used	Rat model of ADHD +	During pregnancy rats given n-3 PUFA enriched feed and offspring continued this diet. Control groups received control feed (n-6/n-3 of 7:1).	In males, PUFA supplemented feed enhanced reinforcement-controlled attention, while reducing hyperactivity and impulsiveness. Opposite or no effects seen in females. Enhanced dopamine and serotonin turnover rates observed in the male mice, whereas females exhibited no/limited change.
Zhu et al.[77]	C57BL/6 J mice. Fecal samples collected from 11 patients with schizophrenia, and 10 healthy controls	Male only mice use	FMT from drug-free schizophrenia patients to mice	Mice underwent oral gavage of antibiotics, and at 48-hours after last gavage, were randomized to receive FMT via oral gavage from either patients or controls, 9 times over 3 weeks	FMT from schizophrenia patients into mice caused psychomotor hyperactivity, and impaired learning and memory. 60 bacterial species were different between control FMT mice and schizophrenia FMT mice.
Clinical Studies					
Slykerman et al.[78]	342 of the initially eligible 474 children completed cognitive testing at 11-years of age	Sex of children/ participants not reported	Antibiotic exposure in early life	Maternal probiotic treatment during pregnancy: either *Bifidobacterium animalis* HN019, *Lactobacillus rhamnosus* HN001 or placebo	Children who received antibiotics within first 6-months of life had significantly lower overall cognitive and verbal comprehension and increased risk of problems with executive function, anxiety and attention-deficit hyperactivity disorder (after adjusting for mode of delivery, probiotic treatment, breastfeeding and income).
Aatsinki et al.[79]	Participants were mothers and 2.5-month-old infants with fecal microbiota composition data from the FinnBrain Birth Cohort study (N = 446)	Infant participants (male = 204, female n = 195)	Maternal prenatal psychological distress	Maternal prenatal psychological distress measured 3 times during pregnancy + hair cortisol measured at gestational week 24.	Positive associations were present between maternal chronic prenatal psychological distress and bacteria from the Proteobacteria phylum. Chronic prenatal psychological distress also negatively associated with *Akkermansia*. Prenatal psychological distress and hair cortisol did not correlate with infant fecal microbiota diversity.

(*Continued*)

Table 1. (Continued).

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention/ Exposure	Outcomes
Korpela et al.[80]	Mothers who had scheduled/ planned C-Section deliveries were enrolled in study (	Infant participants (Female = 5, Male = 2)	Maternal FMT for C-section delivered infants	FMT group had maternal FMT given alongside mothers breastmilk (5 ml total) at first feeding. Two observational cohorts were used for control samples (collected at same time points as for the FMT-treated infants [HELMi and Jorvi cohorts]	Mode of delivery determines fecal microbiota development of infants The microbiota development of the FMT-treated C-Section born infants differed to the untreated C-section infants and showed significant similarity to vaginally born infants.
Zhou et al.[81]	Mothers who had scheduled/ planned C-Section deliveries were enrolled in study (N = 68)	Infant participants (Female = 26, Male = 42)	Maternal vaginal microbiota transfer (via skin) for C-section delivered infants	Infants randomized to control group (exposed to sterile gauze) or vaginal microbiota transfer group (exposed to gauze with vaginal fluids)	Vaginal microbiota transfer associated with improved neurodevelopment in C-section born infants. Vaginal microbiota transfer treated infants and altered gut microbiota compared to controls, potentially this transfer could restore C-section born infants microbiome to resemble vaginally born infants.
Wilson et al.[82]	Mothers who had scheduled/ planned C-Section deliveries (N = 25), or vaginal birth (control group, N = 22)	Infant participants (Female = 25, Male = 22)	Maternal vaginal microbiota transfer (via oral administration) for C-section delivered infants	At birth, infants randomized to receive 3 ml solution of maternal vaginal microbes (CS-seeded, n = 12) or sterile water (CS-placebo, n = 13). Vaginally born were the controls (VB, n = 22)	No observed differences in gut microbiome composition or functional potential between CS-seeded and CS-placebo infants at 1 month or 3 months of age
Shaaban et al.[83]	ASD children (N = 30) + Controls (N = 30)	Male = 19, Female = 11	Children with ASD administered probiotics	Probiotic contained *Lactobacillus acidophilus, Lactobacillus rhamnosus* and *Bifidobacteria longum*. 5 g of probiotic powder dissolved in water, given once daily for 3-months	Compared to baseline, following probiotic supplementation severity of autism symptoms was improved as were gastrointestinal symptoms.
Kang et al.[84]	ASD children (N = 18), neurotypical children (N = 20)	ASD children (Male = 16, Female = 2), neurotypical (Male = 18, Female = 2)	FMT given to ASD children	2-week antibiotic treatment, then FMT given at a 'high dose', plus daily, lower maintenance FMT doses for 7–8 weeks.	GI symptoms were reduced by 80% following treatment and persisted for 8-weeks after treatment. Behavioral ASD symptoms also improved after 8-weeks treatment ended. Overall bacterial diversity and abundance of *Bifidobacterium, Prevotella, and Desulfovibrio* increased following FMT
Kumperscak et al.[85]	ADHD Children & adolescents (N = 32)	Placebo group (Female = 3, Male = 11), probiotic group (Female = 6, Male = 12)	ADHD children & adolescents given probiotic	Probiotic group (N = 18) received *Lactobacillus rhamnosus* GG ATCC53103, and control group (N = 14) received placebo capsule. Capsules were taken once daily for 3-months	Self-report quality of life scores significantly improved in probiotic group, but not control. Results from psychometric tests and levels of inflammatory cytokines were ambiguous.
Skott et al.[86]	Children (N = 68), and Adults (N = 114) with ADHD diagnosis but no ASD diagnosis.	Children (Male = 50, Female = 18), Adults (Male = 33, Female = 81)	Effects of a synbiotic on patients with ADHD.	Synbiotic group (Children = 42, Adults = 57), placebo (Children = 26, Adults = 57). Treatment given once daily for 9-weeks. Synbiotic contained Pediococcus pentosaceus, Lactobacillus casei ssp paracasei, Lactobacillus plantarum 2362, and 2.5 g each of fermentable fibers betaglucan, inulin, pectin and resistant starch	Synbiotic and placebo group had equal improvements in ADHD symptoms, and neither group had significant changes to functioning or sub-diagnostic autism symptoms. However, Synbiotic treatment reduced sub-diagnostic autism symptoms in the domain restricted, repetitive and stereotyped behaviors in children, and improved emotion regulation in the domain of goal-directed behavior in adults.
Wang et al.[87]	ADHD children (N = 30)	Children (Male = 24, female = 6)	Effect of *Bifidobacterium bifidum* on ADHD	Probiotic group only (no control group), consumed 2-probiotic sachets daily for 8 weeks.	Across8-week intervention, patients ADHD symptoms improved. LEfSe analysis revealed that *Firmicutes* significantly decreased while *Proteobacteria* significantly increased during the 8-week treatment period.

SPF = specific pathogen free, GF = germ free, PBS = Phosphate Buffered saline, GOS = galacto oligosaccharide, FOS = fructo oligosaccharide, PDX = Polydextrose, FMT = Fecal microbiota transplant, ASD = Autism Spectrum Disorder, ADHD = Attention-deficit/Hyperactivity Disorder, FMT = Fecal Microbiota Transfer, GI = Gastrointestinal, PUFA = Polyunsaturated fatty acids

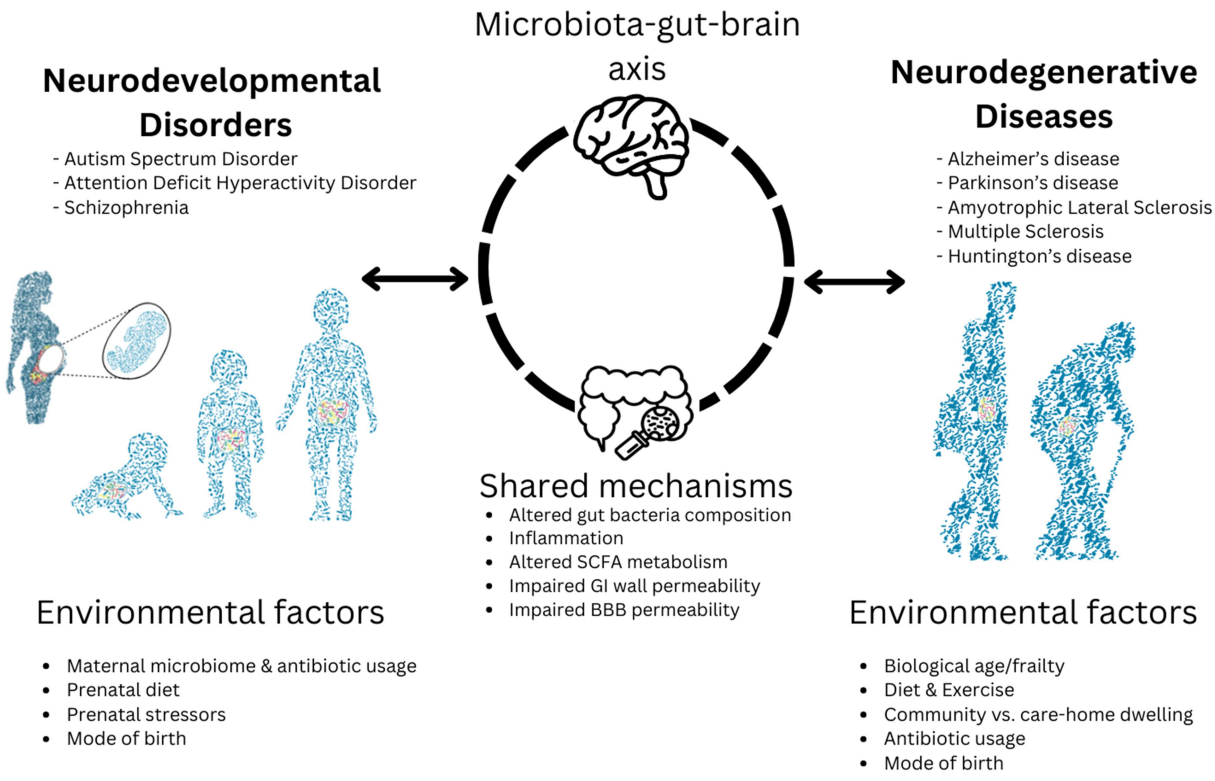

Figure 2. Opportunities for the microbiome to affect development of an infant include both the maternal and potentially paternal microbiome in pre-conception, during pregnancy via indirect effects of maternal microbiome metabolites, birth mode which determines initial microbial colonization, along with the postnatal and infant periods that may be affected by environmental factors including breastfeeding vs formula feeding, antibiotic usage, diet and introduction of solid foods.

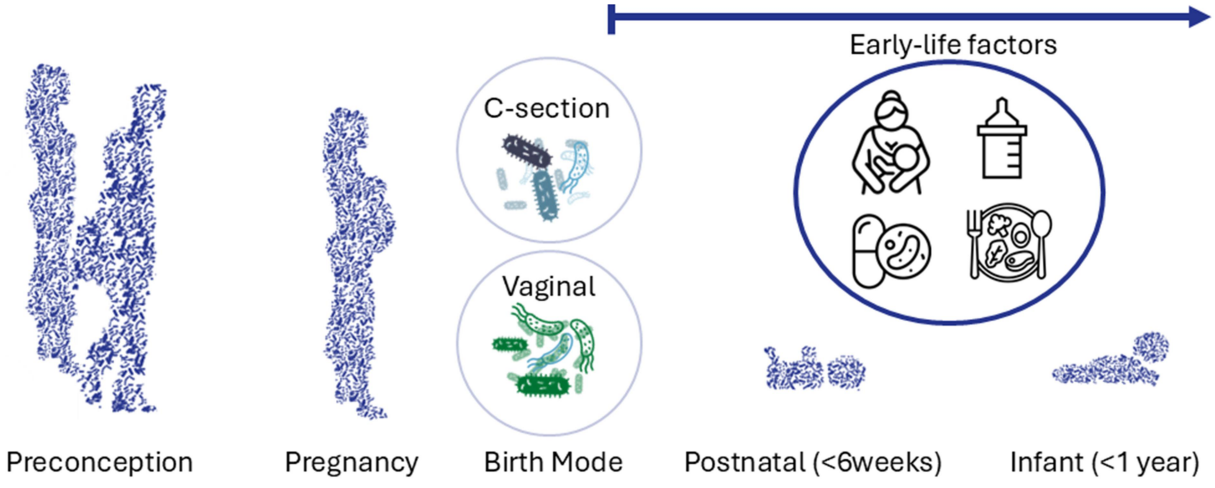

Figure 3. Recent research has underscored the relevance of the microbiota-gut-brain axis across the extremes of life, from neurodevelopmental disorders through to neurodegenerative disorders. Various environmental factors are involved in these relationships, with shared mechanisms of action of the gut microbiome to both neurodevelopment and neurodegeneration. SCFA = Short chain fatty acid, GI = gastrointestinal, BBB = blood brain barrier.

scores.[105] Animal models have also demonstrated the potential for nutrient deficiencies, such as polyunsaturated fatty acids, to amplify the effects of stressors such as maternal immune activation on locomotion and memory in affected offspring.[59] However, when studies examine the effects of

consumption of dietary micro- and macronutrients during pregnancy on neurodevelopment, the results are mixed with a systematic review finding limited associations.[106]

One of the key factors shaping gut microbiome composition is diet.[107,108] Recent evidence evaluates in greater detail the links between the overall state of the maternal microbiome and the long-term health of the offspring. An overall anti-inflammatory diet in the preconception period seems to lower the risk of impairments in numerous neurodevelopmental parameters, such as motor skills, communication, and problem-solving skills.[109] Diets rich in fat and sugar have also been linked to poor cognitive development, while disrupting the gut microbiome of the offspring of rodent obesity models.[58,60] In humans, pre-pregnancy obesity was associated with alterations in the relative abundance of *Lactobacillus* and *Bifidobacteria* genera, both strongly associated with inflammation and has subsequently been shown to increase their risk of developing neurodevelopmental disorders.[110,111] Despite growing interest, the role of the microbiome in preconception diet has yet to be fully elucidated as most observational dietary studies do not yet routinely examine the maternal microbiome. However, future evidence from studies using animal models may give insight and direction toward a better understanding of the underlying mechanisms.

2.2.2 The impact of antibiotic use during pregnancy and early life

Antibiotics are prescribed to 20.8% of pregnant women, as they are vital for healthcare and their use cannot be avoided in severe cases of bacterial infections.[112] Initial studies using animal models in this field often used high doses of antibiotics, sometimes as a cocktail, over longer durations that spanned critical time windows.[35] Recently, there has been a shift in how these studies are performed, with more targeted microbial depletions at specific time points during gestation taking preference. Rodent studies have indicated that lower doses of a single antibiotic, in this case penicillin, during late pregnancy and early postnatal life, produces lasting effects in both sexes on gut microbiota, including increased cytokine expression in frontal cortex, modification of blood–brain barrier integrity and altered behavior in the offspring.[61] Prenatal exposure to low-dose penicillin only during the last week of gestation has a sex-dependent effect on neurodevelopmental outcomes, with female mice less anxious while males displayed abnormal social behaviors.[62] Comparisons between early life exposure to maternal antibiotics led to persistent alterations in anxiety, sociability, and cognitive behaviors. Animal models also note that the profile of behavioral effects in terms of anxiety, sociability, and cognitive behaviors due to a broad-spectrum antibiotic cocktail were greater than in animals treated with just penicillin, with the exception of greater deficits in social recognition.[63] Mechanistic insights have also accrued and following exposure to an antibiotic cocktail mixture, offspring displayed altered tactile sensitivity which was rescued by concurrent administration of specific beneficial microbial metabolites.[64] Little work has been done however to examine any potential differences between absorbable and nonabsorbable antibiotics. Most animal models now use various combinations of nonabsorbable antibiotics whose antimicrobial effects are confined to the gut.[113–115] The direct and indirect effects that absorbable antibiotics may have on neurodevelopment is yet to be fully elucidated, and it may prove that the different antibiotic drug classes along with different mechanisms of action (e.g. neurotoxic effects of metranidozole)[116] may produce differential effects on both the offspring microbiome as well as many neurodevelopmental parameters, depending on the doses used.

In relation to human observational studies, prenatal antibiotic exposure significantly increases the risk of developing ADHD and ASD.[117–119] Regarding the increased odds of developing ADHD, studies report varying levels of antibiotic exposure are required for neurodevelopmental consequences to occur, with one study finding an association after one course and another study requiring three courses.[117,118] Maternal antifungal exposure has also been associated with an increased the risk of developing ADHD, however this has only been noted in males, hinting at a strong sex difference of prenatal exposures, which aligns with the current literature on sex differences in the prevalence of ADHD.[118] While currently only evaluated in animal models, the supplementation of

microbial metabolites or certain bacterial strains may be a beneficial future intervention option.

Antibiotics are often given prophylactically to infants in the neonatal intensive care unit [120] and overuse is common,[121] with earlier exposure during the first year of life a greater risk factor for subsequent difficulties.[122] Antibiotic exposure in early life is known to negatively impact the infant's gut microbiome, which is of lower diversity and more unstable, and thus more susceptible to being influenced by external factors compared to that of an adult.[123] Early antibiotic use can delay the maturation of the infants microbiome and reduces the diversity of species present,[124] negatively impacting the microbiota-gut-brain axis, and influencing brain function and behavior.[1,125,126]

Infants who received a course of antibiotics within their first 6 months have been found to have a higher risk of developing behavioral and emotional problems, as well as having deficits in language development.[78] The result from this study strongly supports the accumulating evidence that the microbiome is involved in neurodevelopment. Interestingly, the developmental outcomes are not as severe in infants that received antibiotic treatment aged 6–12 months which have improved cognitive outcomes.[78] Infants receiving their first course of antibiotics aged 12–24 months were shown to have enhanced cognition in comparison to both previous timepoints.[78] This suggests that a critical microbiome-gut-brain axis window may potentially exist. However, it is important to note that this does not mean that antibiotic exposure in later life will have no neurodevelopmental consequences. Research previously performed by the same group has noted that following antibiotic exposure within the first 12-months of life negatively impacts overall intelligence and reading ability.[127] Additionally, higher levels of ADHD-like behavior and depressive-like symptoms were noted in the peri-adolescent period, with an increased risk of ADHD being found if the exposure to antibiotics was within the first two years.[117,127] However, the risk of children developing ASD was found to be quite mixed, with some studies finding an increase in ASD risk following antibiotic usage in early life,[117,128] whereas others found no association.[129] In contrast to other findings, it has also been reported that oral vancomycin treatment can transiently alleviate ASD symptoms.[130] Despite this, it is important to note the small sample size of this study, containing only 10 participants, and the improvement significantly waned at the follow-up visit after the initial treatment.

Animal studies also confirmed the results found from observational human studies, demonstrating brain and behavioral changes following early life antibiotic administration.[65,66,131] Recent work has aimed to identify a specific temporal window in which microbial depletion is most critical.[65] For example, a more targeted depletion of the gut microbiome following broad-spectrum antibiotic treatment has unveiled varying microglia morphologies and gene expression in rodents, dependent upon time of treatment. This work highlights differential effects on cellular morphology and genetic expression dependent upon timing of the exposure, and this may be the key in relation to future neurodevelopmental outcomes. While this study only notes very subtle behavioral alterations, it provides insight into potential mechanisms.

2.2.3 The impact of prenatal and postnatal stressors on the microbiome and offspring brain

Maternal prenatal stress, whether it be psychological or experiencing a life-changing event, can lead to neurodevelopmental consequences for the fetus. It is possible that an altered gut microbiome may promote these outcomes, as stress has been shown to negatively impact the microbiome, although more work is required to establish definitive links.[67] In animal models, prenatal stress was shown to lead to increased anxiety and alterations to levels of brain derived neurotrophic factor in the placenta and the amygdala in adulthood.[67] The authors reported that the affected offspring's microbiome was disrupted compared to controls, with an altered relative abundance of Firmicutes, Bacteroidetes, and Rikenellaceae.[67] Disrupted social behavior is often reported, accompanied by a profile of gut-brain axis alterations including altered microbial composition, neuroinflammation, and tryptophan processing/metabolism, [68,132 p. 2]. Indeed, the interface between the gut microbiome and the social brain is seen across species.[133]

Shifts in infant microbial composition following prenatal stress are also seen in humans. This includes

an increased abundance in many possibly inflammatory genera from the Proteobacteria group, such as *Haemophillus and Campylobacter*, following chronic maternal prenatal stress, as well as *Finegoldia and Veillonella* from the Firmicutes group.[79] While there is no doubt that prenatal stress impacts cognition and motor development, the association with the maternal microbiome is currently in need for further elaboration.[134]

The relationship between postnatal stressors, such as maternal separation, and neurodevelopment has also been explored, albeit mostly performed in animal studies. Maternal separation has long been used as a model of early life stress, adverse exposures, and importantly, GBA dysfunction.[135] The isolation of the pups from postnatal day 2 until 12 has not only been shown to disrupt the serotonergic system, alter microglia, and lead to a chronic state of inflammation via polyunsaturated fatty acids,[69,136,137] but has also been shown to alter behavior in rodents,[70,138] such as increased anxiety levels and reduced learning and memory. Since the initial observation that this is also associated with microbiome disruption,[139] it has subsequently been demonstrated that different aspects of the emergent phenotype are microbiota dependent (anxiety-, depression-like behavior) and independent (HPA axis dysfunction).[138] Interventions targeting the microbiota that rescue or ameliorate the phenotype, with positive results are emerging. Prebiotic supplementation following early life stress has been shown to regulate the HPA axis, along with milk fat globule membrane (MFGM) which has been shown to reduce visceral sensitivity, with the compounds also improving cognition.[70,71] Anxiety phenotypes, stemming from early life stress has also been rescued following supplementation with prebiotic and *Lactobacillus rhamnosus GG*.[72]

In relation to human studies, little work has been done to examine the effect that this specific stressor may have on neurodevelopmental outcomes. What is known however, is that separation of mother and child does negatively impact breastfeeding, which has been shown to impact long-term neurodevelopmental outcomes.[323,140]

2.3 Early shaping of the infant microbiome

2.3.1 How preterm birth and birth mode impacts long-term neurodevelopment

Preterm birth, defined as delivery before 37 weeks' gestation, affects just under 15 million infants annually and this premature delivery can lead to severe neurological consequences.[141] Major brain development occurs in the final trimester, and delivery during this critical window may lead to brain injury and other neurodevelopmental consequences.[142,143] The gut microbiome of these infants has previously been shown to have unique microbial profiles.[144] Using 16S sequencing, recent work has shown overgrowth of *Klebsiella* in the gastrointestinal tract of these premature infants, an increased abundance of *Bifidobacteria and Finegoldia* species were also noted. Interestingly, these changes were seen in those with severe brain injuries and those without were found to have elevated levels of *Streptococcus and Enterococcus*.[143] Despite the many microbial changes, *Klebsiella pneumoniae* was found to be significantly association with brain injury, with the authors noting that *Klebsiella* overgrowth is a key for the dysregulation of the infant's microbiome-immune-brain axis, perhaps aggravating the brain injury.[143,145]

The newborn's microbiome is heavily influenced by mode of delivery. If born vaginally, the infant is colonized with bacteria present in the mother's vaginal canal, including *Lactobacillus* which is beneficial and predominant in healthy women, and certain fecal bacteria, such as *Bacteroides and Escherichia*.[81] In contrast, a cesarean section (C-section) delivery results in the infant colonized with bacteria including *Staphylococcus and Klebsiella*.[81] Despite C-sections being a vital and often lifesaving healthcare innovation, they may be overutilized in high-to-middle income countries,[146] and they may have neurodevelopmental risks. Research has examined the long-term effect of C-section on cognition and C-sections are linked to an increased risk of intellectual disability, ADHD, and ASD.[147,322] Additionally, evidence shows, at least for ASD, that this association may be due to familial confounding by genetic and/

or environmental factors.[149] Deficits in gross and fine motor development, along with language skills have also been noted by 5 months old.[150] Problem-solving has also been found to be delayed at 3 years following C-section delivery.[151] Interestingly, the risk of certain disorders, such as ASD, seems to be greater if the C-section is elective.[148] It is possible that the underlying reasons for the C-section, along with the altered microbial colonization, may play a role in the neurodevelopmental deficits, and pre-clinical research highlights a role for the gut microbiome in the enduring behavioral effects induced by a c-section model.[152] Given the gut microbiome alterations following C-section do not endure, there remains an urgent need to demarcate the time windows when interventions should be applied. The behavioral alterations that are seen in the human population has also been demonstrated in animal models. C-section has been shown to negatively affect behavior across the lifespan of mice, noting higher levels of anxiety and altered sociability.[73] Interestingly, these neurobehavioural alterations were partially rescued following co-housing with mice born conventionally, allowing a natural transfer of the microbiome. Similar positive changes were also noted following administration of *Bifidobacterium breve*, highlighting the potential impact that a probiotic intervention may have against these known C-section deficits. It has also been noted that this model results in increased body weight.[153] Interestingly, prenatal and peripartum exposure to antibiotics interface with C-section delivery to modify microbiome profiles in human infants,[154] a feature which needs to be built into more translationally faithful versions of this animal model.

Research has attempted to ameliorate the effects of a C-section birth on microbial colonization, by examining the impact of 'seeding' the neonate with maternal vaginal secretions. Seeding aims to recolonize the neonate's gut to match a vaginally born infant, which will have the microbes of the maternal gut and vagina, as opposed to the microbes of the maternal skin.[155] Recolonization may alleviate the negative developmental outcomes that can be associated with C-section.[81] While not widely used, preliminary results have found that vaginal seeding of C-section exposed infants leads to neonate intestinal recolonization which partially matches a vaginally born infant.[81,156] Recolonization also helped to regulate levels of important metabolites and metabolic functions, including an increase in carbohydrate, amino acid, and energy metabolism and increased levels of L-lactic acid, homovanillic acid, and N-acetyl-D-glucosamine, which may potentially lead to better outcomes in later life.[81,156] Individuals receiving the vaginal microbial transplant scored higher in neurodevelopmental questionnaires than those who received a saline control.[81] However, additional research indicates no positive effect of seeding.[82] Separate research has shown that maternal FMT displays promising results in normalizing gut bacterial composition in C-section compared to vaginal births in mice.[80] While findings suggest the possibility of intervention at this early stage, to prevent development of certain disorders, there is currently limited evidence supporting this technique given that most studies are too small and exploratory in nature to form an actionable evidence base at present. Others have highlighted the potential dangers of seeding the infant with a suboptimal vaginal microbiome and/or one containing harmful pathogens, including group B Streptopcoccus, and various sexually transmitted infections, i.e., *Neisseria gonorrhoeae* or *Chlamydia trachomatis*, with screening of donor samples an important consideration in future guidelines.[157]

2.3.2 The neurodevelopmental benefits of breastmilk

Breastfeeding has been recommended by the World Health Organization for a minimum of 6 months, but should ideally remain part of the diet until the infant reaches 2-years.[158–160] Through breastfeeding, the infant receives various nutrients including necessary proteins (lactoferrins and caseins), prebiotics (human milk oligosaccharides), polyunsaturated fats, and immunoglobulins.[161] These nutritional compounds positively influence the developing microbiome of the infant and can increase the abundance of beneficial microbes, including *Bifidobacteria*, and can decrease abundance of other bacteria including *Bacteroides*.[162] Interestingly, infants fed breast milk exclusively have a less diverse and more immature microbial composition than those receiving infant formula as their diet.[163] However, in this context, low diversity is perhaps more beneficial for the infant and recent evidence

supports the idea that breastfeeding supports an appropriately paced assembly of the gut microbiota to control the acquisition of microbial species and functions.[164] Feeding type is known to alter gene expression and this ultimately enhances the transcription of genes that are associated with various immune and metabolic processes.[165] In comparison, the microbiome of formula fed infants is more adult-like, and this may not be ideal in early-life.[165] Breastfeeding for a prolonged period is strongly associated with optimal neurodevelopment, including a reduced risk of ADHD, with a similar protective effect found with ASD and schizophrenia.[166–170] Breastfeeding is also associated with greater IQ scores and enhanced cognition.[171,172] Magnetic resonance imaging studies show white matter microstructural development is increased in areas associated with language, vision, and higher order thinking and cognition in longer durations of breastfeeding, or when comparing breastfeeding to formula fed infants.[173] The role that the gut microbiome has in the neurodevelopmental benefits of breastmilk is still to be fully elucidated, but one potential component, MFGM, has shown great potential in improving certain neurodevelopmental outcomes following maternal separation in rats.[70,71] This includes spatial learning and memory, along with reducing visceral sensitivity stemming from the early life stress.[70,71] In the same study, prebiotics also displayed potential in improving learning and memory along with helping the HPA axis return to baseline levels following restraint stress, with the combination of prebiotics and MFGM proving to have some greater effects.[71]

2.4 Neurodevelopmental disorders and the gut microbiome

While the subsequent sections will focus on the role of the microbiota-gut-brain axis in neurodevelopment disorders, it is important to highlight the genetic component to neurodevelopmental disorders. Autism, ADHD and schizophrenia all have a genetic component increasing risk of developing the disorder,[174–176] with ASD genetic risk not only inherited but also involving de novo copy number variations,[177] while ADHD and schizophrenia may mostly involve heritable genetic risk.[178–180] Despite the literature highlighting the genetic component to these disorders, how this relates to environmental factors, such as breast feeding, and to the gut microbiome is not yet understood. Additionally, the genetic risk factors only explain a smaller part of the disease risk (despite high heritability in twin and family studies) and the gut microbiome could be one of the further contributing factors in developing a disorder. To develop a more whole understanding of the risk factors contributing to these disorders, researchers need to investigate gene-environment-development interaction.

2.4.1 Autism Spectrum Disorder (ASD)

As described above numerous factors influence the microbiome in early life and may increase the risk of neurodevelopmental disorders. People with ASD often suffer from gastrointestinal symptoms, which may be due to a disruption of the microbiota-gut-brain axis.[181,182] A gut bacterial disruption could relate to the significant relative bacterial abundance differences in 27 genera, including *Akkermansia, Bifidobacterium, Clostridium, and Ruminoccocus*, when comparing ASD to controls.[181,183] Gut microbiome differences are also observed in animal models of ASD, including greater relative abundance of Firmicutes and Clostridia, along with lower Bacteroidetes abundance.[181,184] Recently, 16S sequencing has identified a signal unique to ASD patients that has been shown to be driven by 591 microbes.[41] This, coupled with 138 microbial encoding genes linked to ASD through shotgun sequencing, may prove to be a beneficial aid in diagnoses practices.[41]

Of note, common ASD symptoms, i.e., repetitive behaviors and sensory preferences, are linked to certain food choices, and typically a restrictive diet leads to a less diverse microbial composition.[185] While this suggests that diet drives the microbiota alterations that have been reported, we should not discount the ongoing role of the gut microbiome in ASD symptom expression, or that it might represent a tractable target in this context.

In experimental research, FMTs from donors with ASD to mice transferred both the patient's microbiome, and an ASD-like phenotype to the mice (i.e., decreased communication and locomotion, and increased repetitive behaviors).[74] The shift in microbiome composition correlated with reduced social behaviors and repetitive behaviors,

and *Eisenbergiella tayi* (02b40_Lachnospiraceae) correlated with increased repetition and social defects.[74] Overall, this study shows the potential causal role that the gut microbiome has in the pathophysiology of ASD, or at least of specific symptom profiles, and indicates that targeting the gut microbiome may be an effective therapeutic option for ASD. Yet, other work has deduced other potential mechanisms, such as the vagus nerve, to modulate certain ASD symptoms.[75] This has been seen following *Lactobacillus reuteri* administration in the *Shank3B*-/- mouse model, noting a negative impact on social interaction following vagotomy, showing an indirect mechanism for beneficial microbes to alter the symptom severity.[75] Probiotic interventions, using species such as *Bifidobacterium longum, Lactobacillus rhamnosus*, and *Lactobacillus acidophilus*, helped restore the microbiome and even reduced the severity of gastrointestinal and autistic symptoms in children with ASD.[83,183] FMT interventions have also been tested, which involve antibiotic consumption, fasting and bowel cleansing, and a particular refinement of FMT from neurotypical donors along with stomach acid suppressants, which resulted in children with ASD reporting a reductions in gastrointestinal symptoms, enhanced microbial richness and plasma metabolites, and alleviation of autistic symptoms.[84,186,187] These positive changes occurred after the initial microbiota transfer therapy and persisted for 2-years following cessation of the transfer, with some symptoms even improving further after the end of the study.[186] Across the timeframe of the study, an increase in Prevotella was noted, along with significant decreases in the abundance of certain microbes that had low uncertainty.[41] Abundances of *Anaerobutyricum and Butyricimonas*, SCFA producers, remained stable across the study which may be potentially crucial in modulating symptoms.[41] The beneficial effects of probiotics and microbial transplants highlight the intricate relationship between the gut microbiome and ASD, as well as the potential for it to act as a therapeutic target.

2.4.2 Attention-deficit/hyperactivity disorder (ADHD)

The gut microbiome has also been implemented as a factor that may be influencing ADHD. A significant amount of our understanding comes from human studies. When compared to those without ADHD, studies have found significant alterations in the composition of the gut, yet findings are not consistent.[188–191] Differences between patients and controls include an elevation of *Odoribacter splanchnicus*, *Bifidobacterium* and Bacteroidaceae abundances, and lower abundance of *Faecalibacterium prausnitzii*.[191–193] Greater Actinobacteria abundance is also associated with reduced ADHD symptom severity. [191] Given that the prior studies indicate relationships between gut bacteria and ADHD, studies have investigated the use of probiotics in ADHD. *Lactobacillus rhamnosus GG*, when given during pregnancy and early life, can significantly reduce the overall risk of ADHD and improve outcomes such as overall social and emotional functioning in children.[85,189,194] Beneficial impacts on behavior in children with ADHD were also observed with *Bifidobacterium bifidum* consumption, and a multi-strain probiotic, which alleviated symptoms of inattention and impulsivity.[189] However, no effect was seen with a synbiotic consisting of a Lactobacillus based probiotic and 3 different fermentable fibers on overall ADHD symptoms.[86] The synbiotic seemed to have a greater effect on those with elevated levels of soluble vascular cell adhesion molecule-1 before the beginning of the trial, with these adults displaying improved emotional regulation. Apart from probiotic interventions, some research has examined the effect of FMT on ADHD, where material from a neurotypical donor led to increased abundance of, the anti-inflammatory and beneficial, *Faecalibacterium prausnitzii*, reduced abundance in *Bifidobacterium longum*, and a reduction in ADHD symptoms.[87,195] While this result is promising, the research is a case-study of one participant. To generate more conclusive results extensive work needs be carried out to fully understand if targeting the microbiome via probiotics or FMT is a useful intervention in ADHD. Furthermore, the use of animal models has been greatly underutilized in studying

this topic. Enriching rats diet with omega 3 polyunsaturated fatty acids was found to ameliorate symptoms of ADHD in males,[76] particularly through decreasing impulsive behaviors and improving attention, but not much more has come from animal studies.

2.4.3 Schizophrenia

As with ASD and ADHD, schizophrenia is a mental health disorder counted among the neurodevelopmental disorders, although occurring later in life compared to ASD and ADHD, and is becoming more convincingly associated with the gut microbiome.[196–198] Patients with schizophrenia display a different gut microbial composition to those without schizophrenia, including lower abundance of *Clostridium, Faecalibacterium and Ruminococcus*, with greater abundances of *Lactobacillus, Collinsella, and Anaerococcus*.[199,200] At the genus level, those with schizophrenia had greater α and β diversity, and had greater abundance of anaerobic bacteria, along with various species that typically reside in the oral microbiome.[201] While limited animal work has been performed tying the microbiome and schizophrenia together, one study demonstrated severe behavioral changes in mice that had received FMT from patients with schizophrenia.[77] These changes include impaired learning and memory and psychomotor activity and highlights the severe disruption of the patient's microbiome and provides insight into how the microbial alterations seen in the gut may be affecting the gut-brain axis and behavior. While probiotic treatments have improved certain GI symptoms, potentially due to the immunomodulatory effects of the probiotic bacteria, no definitive change in schizophrenia symptom profiles were shown.[202,203] Of note, distinct microbial profiles are associated with different symptoms of schizophrenia, i.e., positive symptoms including hallucinations and delusions, or negative symptoms such as anhedonia, avolition, and issues with intrapersonal relationships.[191,204] Positive symptom phenotypes are positively correlated with Lactobacillus, and negative symptoms are positively correlated with Lachnospiraceae and Ruminococcaceae.[205] The severity of symptoms can also be correlated with numerous bacterial species in the gut, and furthermore, the microbial profile is known to shift and change following a first episode of psychosis,[199] p. 202.[206] Given the literature highlights differential microbial profiles between patients based on their symptomology, future research should consider different phenotypes and trajectories of schizophrenia when assessing and implementing gut-based therapeutics. The potential for modulating the gut microbiome is not as clear with schizophrenia as it is for ASD and ADHD, yet the hope remains that careful microbial targeting, and regular psychotropic medication, may improve the outcomes and lives of those with schizophrenia.

3 Aging and the gut microbiota

Several hallmarks of aging have recently been proposed that relate to the microbiota-gut-brain axis; chronic inflammation and compositional/functional gut microbiome alterations.[207] Aging can be both 'healthy', where age-related declines occur at a normal pace or are delayed, or can be 'unhealthy', when declines are accelerated.[208] Gut bacterial differences have been observed across the age range from newborns to centenarians.[8] In long living individuals (i.e., nonagenarians and centenarians), gut bacterial composition differs to younger age ranges,[209,210] and compositional and functional differences are also present in these populations depending on health status.[321] Additional research assessing 21,000 fecal samples determined that a loss of the 'core' microbiome and an increase in disease associated taxa were most correlated to unhealthy aging.[211] In animal models, it has been shown that germ free mice can live longer than specific pathogen free mice, despite the fact that germ free animals have several altered body systems and behaviors, indicating the need for a gut microbiome for 'normal' aging and immune/nervous system function.[20] However, there is little current research that has thoroughly investigated the differences in physiology of aging between germ free mice and those with a gut

microbiome, and the absence of pathological infections also needs to be considered as a factor.[20]

Longitudinal assessments of cohorts across aging would help assess the link between gut bacteria and aging. However, existing research in humans is limited. A published protocol paper aimed to track older adults over a 4-year period,[212] and investigations of centenarians' gut bacteria over 1.5 years has occurred.[213] Compared to older adults, centenarians had a 'youth-associated' gut microbiota profile, including less potentially pathogenic bacteria and a greater abundance of Bacteroidetes. Centenarians also had greater within-individual similarity over time, compared to between-individuals, and individuals with the most within-individual similarity had greater Shannon's diversity, and Pielou's evenness index, indicating that greater evenness and diversity contribute to gut microbial stability in centenarians.

An important consideration when investigating aging, is chronological age (time since birth) vs. biological age [refers to biological and physiological (I.e., DNA), or lifestyle (I.e., nutrition and physical activity) related factors affecting longevity and functional capacity].[214,215] For research on determining biological vs chronological age see Rutledge et al.[216] Biological age relates to frailty, a decrease in function and physiological reserves that coincide with an increased risk of morbidity and mortality.[217] Assessment of biological age is important for gut microbiome research as frailty, or unhealthy aging may not relate to chronological age, and the associations between the hallmarks of biological age and gut microbiota may be more informative. Assessment of frailty and chronologic age in relation to the gut, oral and skin microbiota showed associations between the skin microbiota and frailty were present for several taxa at several skin microbiota sites.[218] The skin microbiota was not associated to chronological age, indicating the importance of assessing frailty, i.e., biological age. However, authors did not report associations between frailty and the oral or gut microbiota, and it is not explicitly stated if assessment of gut bacteria and frailty were conducted for both older adult groups, or separately for their community and nursing facility dwelling groups. Of note, nursing home dwellers were significantly frailer than community dwellers, which is to be expected, however location, not just frailty, would affect the observed skin microbiota associations and the reported associations between skin microbiota and frailty, cannot be disentangled from location. Other research shows associations between gut bacteria diversity and frailty of nursing home residents, with trends for greater composition of butyrate producers in individuals with lower frailty, and greater LPS biosynthesis in frailer individuals.[219]

Several FMT trials in mice have helped elucidate causality of the gut microbiome in relation to aging. When FMT was conducted from young donor (3–4 months) mice to older recipients (19–20 months) the aging-associated differences in brain and peripheral immunity, and the hippocampal metabolome and transcriptome, were reversed.[220] The reversal of age-related brain differences in mice from FMT has been replicated, with declining cognition rescued from FMT of young-to-older mice.[221] Additionally, intestinal microbiota were exchanged between young (3-months) and old (18-months) mice (i.e., young to old, and old to young FMTs).[222] When the aged microbiota was received by young mice, there was increased intestinal barrier permeability, greater inflammation (measured via serum LPS-binding protein and IL-6), and more markers of systemic and tissue markers of 'inflammageing'. In the opposite direction, older mice in receipt of a young donor microbiota, the age-associated elevations of inflammation were reversed (i.e., reduction in serum TNF and LPS-binding protein). Understanding the role of aging on the gut microbiome is important in the context of neurodegenerative diseases as advancing age is the biggest risk factor for Alzheimer's and Parkinson's disease,[223–225] is a major risk factor for ALS,[226,227] and while disease onset occurs much earlier in terms of age (years) in HD and MS, it is still important to understand how aging impacts disease onset and progression.[228,229]

3.1 Cognitive decline, MCI, and Alzheimer's disease

Some cognitive decline normally occurs with advancing age. When the rate of cognitive decline increases it may be considered mild-cognitive impairment (MCI), a common precursor to dementia.[230,231] Several observation and

intervention studies have investigated cognition and the gut microbiome in humans, as covered previously.[232] Much of the literature focuses on the gut microbiome and cognitive impairment or dementia, predominately AD. Additionally, the potential role of the oral microbiome in AD is emerging.[233] Purported mechanisms whereby the gut microbiota may relate to AD and cognitive decline is covered previously,[11,234] but to summarize, an altered gut microbiota may contribute to AD via inflammatory mediated pathways, and via microbial metabolites affecting cognition too. A systematic review of observational research comparing the gut microbiota of healthy controls to AD and/or MCI groups found the AD/MCI groups had lower diversity than controls, and differences in bacterial abundance at the phyla level, family level, and genus level (greater Proteobacteria abundance; lower Clostridiaceae abundance; greater *Bifidobacterium* abundance in AD/MCI, respectively).[37] Additionally, a meta-analysis found that probiotics were effective in improving cognition in people with AD or MCI.[235] Another meta-analysis, solely focusing on AD, included three studies, covered in the prior review,[235] but showed no beneficial effect of probiotic supplementation on cognition.[236] The disparate findings may be due to the small number of trials, but may reflect that probiotic consumption is most effective earlier on in cognitive decline, prior to substantial cognitive decline occurring, such as in AD.

By comparing healthy individuals to preclinical AD (determined via investigating tau and/or Aβ plaques through PET imaging), but lacking AD symptoms, Ferreiro et al., found distinct gut microbiome abundance profiles associated with either preclinical AD or healthy controls.[237] The differences in the gut microbiome composition correlated with Beta amyloid and tau pathological biomarkers, but not with biomarkers of neurodegeneration. Additionally, microbial pathways of L-arginine, L-ornithine, and 4-aminobutanoate degradation were associated with preclinical AD status via regression models, with all three pathways sharing succinate as a product. Overall, this shows an altered gut microbiome may contribute to initial AD pathophysiology, but might be less relevant to ongoing AD progression, and highlights the potential for an altered SCFA metabolism state to be present in preclinical AD. Prior research shows lower SCFAs levels in MCI compared to controls, with AD patients compared to MCI having lower levels of SCFAs.[238] Supporting this, research shows reduced levels of putatively butyrogenic bacterial genera in AD compared to healthy controls.[239] Apart from butyrate, succinate is likely immunomodulatory,[240,241] and inflammation via the gut is associated with AD pathophysiology.[242] Secondly, succinate is a precursor to propionate, and excess propionate has been associated with AD in humans.[243] Despite research indicating the likely involvement of gut microbiota in AD pathophysiology via multiple routes,[242] particularly early in the disease progression, research is currently not at the point where a gut microbiota biomarker is available to be used as an early-detection biomarker.

FMT research has investigated cognition and the gut microbiome, and shows that a 'young' microbiota helps improve cognition when transplanted into older mice.[220] Mechanistically, FMT from aged to young recipient mice decreased adult neurogenesis and novelty-induced neuronal activation of the hippocampus.[244] By interrogating vagus-nerve related neuronal activity the authors showed that mice receiving the aged microbiota FMT had reduced neuronal activity in the ascending vagus-nerve output brain structure. In an AD animal model, near daily FMTs across 4-months (wild-type donors to AD model) rescued cognitive deficits, and reduced formation of amyloid β plaques and neurofibrillary tangles.[245] Increasing the translatability of FMT research, Grabrucker et al.,[246] transplanted human fecal samples from healthy donors and people with AD into animals. Authors displayed that behavioral AD symptoms, specifically cognitive/memory deficits (which correlated with donors Clinical Dementia Rating scale), were transferred to previously healthy animals via gut microbiota, confirming a causal and ongoing role of gut microbiota in AD symptoms. Therefore, while human research showed the gut microbiome to be related to early AD pathophysiology,[237] human-to-animal FMT studies indicate the prolonged role of the gut microbiota throughout AD. Grabrucker et al.,[246] also substantiated prior research, with FMT from AD patients to animals impairing behaviors dependent on adult

hippocampal neurogenesis, and the severity of impairments in mice were correlated with cognitive scores of the AD donors.

A case study of FMT to treat clostridium difficile infection, in a person with Alzheimer's, showed improvements in cognition (measured via mini-mental state examination[MMSE] and Montreal Cognitive Assessment[MoCA]) one-month following FMT.[247] The same group assessed 5 people with AD and clostridium difficile infection over 3-months, reporting no serious adverse events from the FMT and all patients having improved cognition (measured via MMSE, MoCA and Clinical Dementia Rating Scale Sum of Boxes).[248] Whether cognitive improvements are related to the FMT affecting AD pathophysiology, or simply treating the infection, is not clear. Apart from FMT, probiotics have been investigated as a therapeutic option across several animal models for AD, which has been summarized previously.[249] Future research assessing FMT in AD specifically, should use cognitive tests that address specific cognitive domains, and that are more sensitive to changes to cognition (I.e., N-back, spatial working memory etc.), rather than cognitive screening forms including the MoCA or MMSE.

3.2 Additional neurodegenerative diseases

Alzheimer's, discussed above, and PD, ALS, MS, and HD described below (with key gut microbiome research studies and study characteristics described in Table 2), all have a genetic/heritable component. In AD the presence of the apolipoprotein E gene-e4 allele is a contributing risk factor [260] with additional gene mutations implicated in AD onset.[223,261] A genetic component to PD onset and severity/symptoms is also present,[262,263] which similar to the genetic risk in AD, mostly relates to early-onset cases with reduced relevance to late onset cases. For ALS,[264] and MS,[265] genetic/familial risk factors exist, although genetics alone explain only a small proportion of the risk of disease development.[266] Comparatively HD is considered a solely genetic disease, with children of those who carry the increased number of 'CAG' triplets on chromosome 4 having a 50% chance of inheriting the gene mutation and developing HD,[267] with the number of 'CAG' repeats associating with age of HD onset (along with additional genetic factors explaining variance).[268] Despite the genetic component to HD, environmental factors may exist that alter disease progression or age of onset.[269] Given the genetic and environmental factors underpinning these neurodegenerative diseases, future neurodegenerative disease research should assess the genetic, environmental and microbiome factors. Doing so will be important to understand the interaction between genetics and environmental factors and to uncover where the microbiota-gut-brain axis fits into the web of risk factors.

3.2.1 Parkinson's disease (PD)

Early indications that the gut may be involved in PD including constipation and gastrointestinal issues (GI) preceding motor impairments,[270] and the dual-hit hypothesis,[271] led to researchers investigating the role of the microbiota-gut-brain axis in PD. Since then, it has been shown that gut microbiota can metabolize PD medication,[272] and several reviews and meta-analyses have surmised that 1) the gut microbiome differs between healthy controls and people with PD (at a compositional and functional level) and 2) have proposed potential mechanisms underpinning the involvement of the gut microbiota in PD pathophysiology.[10,40]

Observational and mechanistic studies have enabled some insight into the mechanisms of the microbiota-gut-brain axis in PD pathophysiology. Broadly, SCFA (particularly butyrate) producing bacteria appear to be reduced in PD compared to healthy controls, and a greater abundance of pro-inflammatory bacteria and an increase in bacteria that may degrade the gastrointestinal wall is observed in PD compared to controls.[10,40] However, most prior research uses 16S sequencing, limiting the functional inferences that can be drawn, reducing taxonomic resolution compared to shotgun sequencing, and meaning species or strain level analysis is not typically possible [strain-level analysis is also dependent on shotgun sequencing depth].[273,274] Wallen et al., [275] conducted shotgun sequencing on 490 PD, and 234 controls and showed depletion of butyrate producing bacteria in PD compared to controls, but did not find statistical significance for elevation of *Akkermansia* in PD compared to controls, as prior research had.[276,277] Lower levels of fecal

Table 2. Summary of research studies and key characteristics of each research study in relation to later-life, neurodegenerative diseases and the gut microbiome.

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention	Outcomes
Armstrong,[220]	C57BL/6 J mice	Male only used	Aging related behavioral deficits in mice countered with 'young' FMT	Aged mice randomized to receive FMT from young or aged mice. FMT delivered via oral gavage once a day for 3-days, then twice weekly for 8-weeks	FMT from young donor (3–4 months) mice to older recipients (19–20 months) reversed the aging-associated differences in brain and peripheral immunity, and the hippocampal metabolome and transcriptome.
Hou et al.[221]	C57BL/6 J mice	Male only used	Age-related cognitive decline model	Antibiotic treatment preceded FMT, with FMT performed twice, 72 h apart. Mice (15–16 months old) received FMT from young (8-week-old) donor mice,	Cognitive deficits from aging-related gut microbiome remodeling were rescued by FMT from young donors.
Reeve et al.[222]	C57BL/6 J	Male only used	Effects of aging in mice and FMT from 'young' donors	Young (3 months), old (18 months), or aged (24 months) male mice were randomized to experimental cages to receive either FMT or PBS (control). 3-day delivery of broad-spectrum antibiotic given via oral gavage with palatable antibiotics via drinking water. FMT delivered twice 72 h apart	Older/aged and young microbiome profile was successfully transferred via FMT to young and older mice respectively. Aged donor FMT to young mice accelerated CNS related inflammation. Such effects were reversed in older mice by young donor FMT.
Kim et al.[244]	RjOrl: SWISS mice	Male only	Effects of 'young' and 'aged' gut microbiome (from humans and mice) FMT on young mice	Following antibiotics (1-week) young mice received FMT from either young mice donors, or aged mice donors. Separately, young mice received FMT from old or young humans.	FMT from aged, but not young, human/animal donors into young mice triggers profound hippocampal alterations, including astrogliosis, decreased adult neurogenesis, decreased novelty-induced neuronal activation, and impairment in hippocampus-dependent memory
Ağagündüz[245]	WT mice and ADLP^{APT} transgenic mice	Female only mice	AD mouse model (ADLP^{APT} transgenic mouse model) and FMT	FMT from WT mice given orally to ADLP^{APT} mice for 16-weeks, and given to ABX-pretreated ADLPAPT mice for 4 weeks	Gut microbiota composition differed between the WT and ADLPAPT mice. ADLP^{APT} mice also displayed loss of epithelial barrier integrity and intestinal and systemic chronic inflammation. FMT from WT mice ameliorated formation of amyloid β plaques and neurofibrillary tangles and cognitive impairment.
Fratiglioni et al.[246]	41 healthy controls, 54 AD patients. Adult Sprague-Dawley rats	25 female controls, 31 female AD patients. Male rats used only.	FMT from AD patients and age-matched healthy controls into young adult rats	Rats administered ABX-cocktail for 7-days via drinking water. 72 h later FMT given via oral gavage of donor microbiota daily for 3-days. N = 16 rats received control FMT, N = 16 rats received AD FMT	AD symptoms were transferred to rats via FMT from human AD patient donors. Behavioral Impairments in rats receiving FMT from AD patients related to adult hippocampal neurogenesis, and severity of these impairments correlated with clinical cognitive scores of AD donors.
Ingre et al.[250]	C57BL/6 J mice	Male mice	Rotenone induced PD mice model	FMT from control mice given to rotenone induced PD-like mice for 2-weeks, once daily. 15 rotenone/PD mice received FMT while 15 mice did not.	Rotenone-induced mice developed gut microbiota dysbiosis, GI impairment and poor behavior performances. *Akkermansia* and *Desufvibrio* bacteria were increased also. In FMT treated mice, FMT restored the gut microbiome community, ameliorating GI dysfunction and motor deficits, and reduced systemtic inflammation levels.
Waubant et al.[251]	C57BL/6 mice	Male mice	MPTP induced PD mice model + FMT treatment	N = 15 control mice, N = 15 MPTP + PBS group, and N = 15 MPTP then FMT treated mice. MPTP injection for 5-days, then FMT treatment (from control mice) for 7-days via gavage.	Murine model of PD exhibited gut microbial dysbiosis. FMT from control mice to PD mice improves motor function, and ncreased striatal DA and 5-HT content of PD mice.

(*Continued*)

Table 2. (Continued).

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention	Outcomes
Olsson et al.[252]	C57BL/6 J mice Human PD patients and human controls as stool donors for FMT	Male mice Sex of human donors not reported	MPTP-induced PD mouse model and FMT treatment	MPTP given once daily for 5-days + PBS (N = 12), control mice received saline + PBS in place of any FMT treatment (N = 12). FMT given via gavage daily for 10-days for the PD FMT mouse group (N = 12), and the control FMT mouse group (N = 12)	MPTP-mice had elevated Desulfvibrio, mice receiving FMT from PD donors had elevated Akkermansia. FMT from PD donors to MPTP treated mice significantly aggravated motor impairments, dopaminergic neurodegeneration and colonic inflammation. FMT from healthy controls improved these factors in MPTP treated mice.
Curran et al.[148]	$SOD1^{G93A}$ ALS model mice and age-matched wild-type mice	Male and female mice used	ALS mice model, treated with butyrate or antibiotics	$SOD1^{G93A}$/ALS model mice randomized to either non-treatment group (N = 6), or sodium butyrate treatment (N = 10). Sodium butyrate given in 2% concentration in filtered drinking water. $SOD1^{G93A}$/ALS model mice randomized to either non-treatment group (N = 8), or ABX treatment (N = 11). ABX given via filtered drinking water.	Butyrate or ABX treated ALS mice had longer latency to fall (i.e., improved performance) in rotarod test, reduced $SOD1^{G93A}$ aggregation and enhanced enteric neuromuscular function.
Myers et al.[253]	G93A transgenic mice and age matched wild type mice	Not disclosed	G93A transgenic ALS mouse model, treated with butyrate	G93A mice randomized into two groups. Butyrate-treated group received sodium butyrate 2% concentration in filtered drinking water. Control group received filtered drinking water (no sodium butyrate)	Butyrate treatment of ALS model mice restored intestinal microbiota homeostasis, improved GI integrity. Abnormal Paneth cells were significantly decreased in ALS model mice following butyrate treatment
Lee et al.[254]	C57BL/6 mice	Female mice	Experimental autoimmune encephalomyelitis (mouse model of MS] + FMT treatment	Immunized mice randomized into a FMT or no-FMT group. FMT group received FMT via oral gavage daily for 42 days.	FMT may rectify altered gut microbiota in the MS-model mice. FMT also reduced microglia and astrocyte activation and conferred protection on the blood-brain-barrier
Mo et al.[255]	Hemizygous R6/1 transgenic mice crossed with F1 (CBA x C57Bl/6) mice to have male wild-type (WT) and R6/1 (HD) littermates	Male mice only	Transgenic mouse model of HD	No intervention, observation of HD mouse model across time and compared to WT mice	HD mouse model had gut dysbiosis at 12-weeks, not earlier, Gut microbiome volatility was increased in HD mouse model, pre-symptomatic stage. Increase in butanoate metabolism pathway in HD mice model
Fasano et al.[256]	R6/1 transgenic male mice (CBA x C57Bl/6 strain) crossed with females (CBA x C57Bl/6) to generate wild-type (WT) and R6/1 (HD) littermates	Male mice only	Transgenic mouse model of HD and fiber diet	WT and R6/1 mice randomized to control diet (5% fiber), zero-fiber diet or high-fiber (10% resistant starch fiber) from 6–20 weeks.	High-fiber reduced pro-inflammatory bacteria in gut microbiome, and treated affective/cognitive deficits in HD mice. Functional analysis revealed high-fiber diet decreased potentially pathogenic functional bacterial pathways in HD mice.
Hawkes et al.[257]	Male R6/1 hemizygous mice were crossed with female CBAxC57Bl/6 F1 mice to generate male and female WT and R6/1 (HD) littermates	Male and female mice	Transgenic mouse model of HD, and FMT	Mice randomized to control (vehicle/vehicle), ATB-only (ATB/vehicle) or ATB/FMT group. FMT group received oral administration of WT donor ceca content for 3-days	FMT from WT donor to HD mice positively alters cognitive outcomes (particularly in females). In HD male mice, FMT engraftment was inefficient.

Clinical Studies					
Study	Participants	Sex	Disease/Exposure	Intervention	Outcomes
Billingsley et al.[248]	5 AD patients, + stool donors (healthy controls)	3 females, 2 males	AD + *Clostridioides difficile* infection then undergoing FMT	AD patients receiving FMT first underwent bowel preparations as indicated by their condition and doctor's instructions. FMT delivered to intestinal tract during colonoscopy by gastroenterologist, following FMT guidelines	Improvements in MMSE and MoCA were observed pre-post FMT. Gut microbiota composition changes post-FMT, with increase in Bacteroidaceae and a decrease in Enterococcaceae.

(*Continued*)

Table 2. (Continued).

Animal model studies					
Study	Animal + Strain	Sex	Disease/Model	Intervention	Outcomes
O'Neill et al.[258]	PD patients and healthy donors	N = 3 females, N = 9 males	PD + FMT	PD patients in FMT group (N = 8) received 60 g of donor fecal material lyophilized into powder and in capsule, administered twice a week for 12-weeks. Placebo group (N = 4) received placebo capsules at same rate.	Some non-severe upper GI symptoms occurred in FMT group. Baseline Beta-diversity similar between groups, but FMT group beta-diversity significantly increased at 6 and 13-weeks. Objective motor behaviors/symptoms showed transient improvements, with subjective symptom improvements reported compared to baseline in FMT group.
La Reau et al.[259]	PD patients and healthy donors	N = 11 male, N = 4 female	PD + FMT	FMT delivery occurred within 1-hour of fecal sample collection and was done via colonic route (N = 10) or nasointestinal route (N = 5). FMT occurred once.	Scores for PSQI, HAMD, HAMA, PDQ-39, NMSQ and UPDRS-III significantly decreased (i.e., improved) after FMT treatment. 5 cases had adverse events, including diarrhea (N = 2), abdominal pain (N = 2) and flatulence (N = 1)

FMT = Faaecal microbiota transplant, CNS = central nervous system, AD = Alzheimer's Disease, WT = Wild type, ABX = antibiotics, MMSE = Mini Mental State examination, MoCA = Montreal Cognitive Assessment, PD = Parkinson's disease, MPTP = 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP), PBS = Phosphate Buffered Saline, ALS = Amyotrophic Lateral Sclerosis, MS = Multiple Sclerosis, HD = Huntington's disease, PSQI = Pittsburgh Sleep Quality Index, HAMD = Hamilton Depression rating scale, HAMA = Hamilton Anxiety rating scale, PDQ-39 = Parkinson's disease Questionnaire, NMSQ = Non-motor symptom questionnaire and UPDRS-III = Unified Parkinson's Disease Rating Scale – III

SCFAs have also been observed in PD compared to controls.[278,279] However, in the plasma, PD patients had elevated levels of SCFAs.[278,280] Of note, it is difficult to interpret SCFA concentrations (fecal or plasma) as true reflections of microbial production, given the utilization of SCFAs by colonocytes, alterations in gastrointestinal absorption and processing by the liver.[281] Such research may indicate a breakdown in the typical use of SCFAs by colonocytes, subsequently altering SCFA levels, rather than SCFA concentrations being influenced via production alone. While Wallen et al. do not report PD symptoms or severity,[275] other research indicates participants with more PD non-motor symptoms correspond with lower levels of the butyrogenic *Butyricimonas synergistica*.[282] However, Nuzum et al. [282] did not observe a statistical difference of butyrogenic bacteria abundance between PD and control groups, as observed by Wallen et al.,[275] The authors postulate this may be due to PD severity level differences, as the cohort in Nuzum et al., [10] had on average fewer years since PD diagnosis and less severe symptoms when compared to the studies included in a prior systematic review.[10]

Longitudinal research in PD and the gut microbiome has also now been conducted,[283–286] and research covering PD severity,[287] has begun to shed light on causality. Cilia et al., [284] conducted a 3-year prospective study and found that lower abundance of *Roseburia* at baseline was associated with worsening motor (via Hoehn and Yahr staging) and non-motor symptoms (via non-motor symptom questionnaire) at the 3-year follow-up. Similarly, Lubomski et al., showed a underrepresentation of butyrate producing genera across 0, 6 and 12 month timepoints when comparing PD and control groups,[285] however, these findings were not replicated in prior longitudinal research. Minato et al., showed lower counts of *Bifidobacterium* were associated with worsening PD symptoms,[286] and Aho et al., did not find an association between bacterial genera and progressing disease severity.[283] As, these papers spanned only a few years and all used 16S sequencing, longer timeframes, and the use of shotgun sequencing to assess functional differences with time or between stable vs. progressed PD phenotypes, is warranted. Huang et al. assessed early PD, REM sleep behavior disorder (RBD), first-degree relatives of RBD, and control participants.[287] Investigating across these different groups aligns with the brain/body-first hypothesis of Parkinson's disease,[288,289] as isolated RBD was aligned with a body-first progression subtype,[289] and allows for insights to be drawn on the early stages of Parkinson's disease development, potentially prior to onset of motor symptoms. Huang et al., showed that abundance of two different genera, *Faecalibacterium* and *Butyricicoccus*, were reduced with increasing α-synucleinopathy with the RBD group then the early PD group having the second-highest and highest amounts of α-synucleinopathy.[287] While other research did not show iRBD groups had decreased SCFA producing bacteria, they did contain increased *Akkermansia*, which may relate to a potential route of PD pathophysiology through impaired intestinal barrier function.[290] Indeed, both a lower abundance of SCFA producing bacteria, and a greater abundance of mucin degrading bacteria predicted increases in disease severity/progression among people with PD.[291] This led the authors to postulate that individuals with an altered gut microbiome, may progress through PD faster than those with no disturbances.

In addition to the involvement of SCFAs in PD pathophysiology, animal experiments indicate curli producing *E.coli* bacteria results in increased alpha-synuclein deposition in the brain and gut of animals, with the same animals having increased expression of markers of inflammation (i.e., TLR2, IL-6 and TNF).[292] Similar research with *Desulfvibrio* bacteria, harvested from individuals with PD and transplanted into *C. elegans*, showed increased amounts of, and larger, alpha-synuclein aggregates compared to the same bacteria taken from healthy individuals.[293] FMT in animal models has also been utilized in PD research with daily FMTs (from control mice) administered over 2-weeks to a rotenone induced model of PD in mice, subsequently restoring gut microbiota, reducing colon LPS levels, and improving GI dysfunction and motor impairments observed prior to FMT.[250] Similar findings were observed in a 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) mouse model, with FMT attenuating

the MPTP induced gut microbiota changes.[251] Authors also completed FMT from PD to control mice, eliciting motor dysfunction and neurotransmitter loss.[251] Fecal samples taken from healthy people and those with PD, were used in FMT to control and PD model mice (MPTP model of PD).[252] Daily FMT (10-days) from people with PD to MPTP-mice worsened motor impairments, dopaminergic neurodegeneration, and colonic inflammation, while FMT from healthy people improved motor behavior, slowed dopaminergic neurodegeneration, and relieved colonic inflammation. These results indicate the causal role the gut microbiota has in PD pathophysiology. Preliminary FMT studies in people with PD have been conducted,[258,259] with research indicating that FMT is well-tolerated, with some mild-to-moderate adverse events reported (E.g., GI disturbances). Dupont et al., [258] administered the FMT orally (N = 12; placebo group = 12) showing temporary improvements to objectively assessed motor function. Xue et al., [259] administered the FMT via nasal-jejunal tube (N = 5) or via colonoscopy (N = 10), finding that the colonic route offered greater improvements to PD symptoms which persisted for longer than in the nasointestinal route. Lastly, animal model research on α-synuclein and gut microbiota, indicated that α-synuclein pathology alters the ENS and gut microbiome, but those gut microbiota alterations can be modulated by exercise.[294] In addition to FMT, exercise in PD should be investigated for its therapeutic benefits, and capacity to modulate the gut microbiota.

3.2.2 Amyotrophic Lateral Sclerosis (ALS)

Like PD, ALS patients often suffer from constipation. Whether constipation precedes ALS diagnosis, as it can in PD, is not currently known.[295] Non-motor functions in ALS are not well understood, however, prior hypotheses have implicated the microbiota-gut-brain axis in ALS progression/onset.[296] Animal models have shown correlation(s) between ALS and gut microbiota composition, with ALS mice having significant gut microbiota composition differences compared to controls.[297] Researchers also took bacteria associated with ALS and mono-inoculated ALS model mice, showing that *Parabacteroides distasonis* and *Ruminococcus torques* were associated to increased disease severity.[297] Authors also observed reductions in *Akkermansia muciniphila* aligning with ALS progression.[297] Research using $SOD1^{G93A}$ mice conducted longitudinally with and without butyrate treatment showed that without butyrate treatment gut bacterial differences in the $SOD1^{G93A}$ mice compared to wild-type were observed, whereas less differences were observed in the butyrate-treated group.[148] Importantly, butyrate treatment lead to greater motor task performance, and prior research indicates butyrate treatment may delay ALS progression in mice,[253] potentially highlighting that reduced butyrate production is involved with ALS progression.

Research in patients with ALS is sparse, with inconsistent findings. Several bacteria at the genus level were reportedly more abundant in ALS patients compared to controls [298] (including pro-inflammatory bacterial genera *Escherichia and Shigella*, and *Akkermansia*, which opposes animal-model work).[297] Other research has found some differences at a genus level, including the genera *Subdoligranulum* [which contains the butyrate producer *Subdoligranulum variabile* [299]] but no fecal SCFA differences were observed between cohabiting controls and people with ALS.[300] Authors also reported people with ALS had lower cytokine levels (IL-15, IL-8, MCP-1 and VEGF-A) compared to controls. Separate research reveals no difference in diversity, composition or function (via PICRUSt1) between ALS and controls.[301] Additional research that has observed differences between control and people with ALS had small sample sizes (N < 6 per ALS group), limiting interpretability and comparability to larger studies.[302,303] Overall, a lower abundance of butyrate producers, and increased proinflammatory bacteria may be involved in ALS progression. Impairment of the intestinal, blood-spinal cord,[304] and blood-brain barrier, have also been reported in ALS patients, and may relate to microbiota-gut-brain axis involvement in ALS pathophysiology.

3.2.3 Multiple sclerosis (MS)

A systematic review of case-control observational studies of MS and the microbiota-gut-brain axis identified ten studies (all using 16S sequencing), with none of the eight that investigated alpha diversity finding a difference between MS and control groups.[305] At a genera and species level, ≥2 studies observed lower

abundance of *Prevotella, Faecalibacterium prausnitzii* (with one study showing increased *Faecalibacterium* comparing MS to controls), *Bacteroides coprophilus, Bacteroides fragilis*, and a higher abundance of *Methanobrevibacter* and *Akkermansia muciniphila* in MS cases versus controls. While there are concordant differences observed, the evidence is not universally consistent but nevertheless indicates the microbiota-gut-brain axis involvement in MS. A different review also highlights the influence the gut microbiome has on immune function and the relevance of this in relation to MS pathophysiology.[306]

FMT studies in MS mice models indicate trends of FMTs to ameliorate some of the observed gut microbiome differences between MS mice and controls.[254] FMT also led to reduced activation of microglia and astrocytes while conferring some protection on the BBB, myelin and axons in the MS mice models.[254] These findings showcase the potential mechanisms of the microbiota-gut-brain axis influencing MS, and highlight the usefulness of FMT for MS. Several case studies of FMT in people with MS have indicated its potential safety and efficacy in addressing MS symptoms.[307,308] A pilot FMT trial in MS patients (N = 9) were given monthly FMTs for 6-months.[309] Authors indicated that FMT was safe for this population, however this study was underpowered and stopped early (see paper for details), therefore potential benefits of FMT cannot be determined currently.

Several papers have assessed MS and the microbiota-gut-brain axis longitudinally.[310–312] Different findings were observed across all papers, with some broader community differences observed dependent on disease severity or whether it worsened or not,[311] but no distinct bacterial communities were identified between MS and control groups, with some bacterial composition differences observed (lower *Bifidobacterium longum, Clostridium leptum, and Faecalibacterium prausnitzii* in MS compared to controls).[310] In terms of composition, findings also appear to be specific to disease severity or worsening status, as lower levels of *Akkermansia* were observed in disease progressors compared to non-progressors, and lower *Akkermansia* abundance correlated to worsening MS progression.[312]

3.2.4 Huntington's disease (HD)

Evidence of the microbiota-gut-brain axis' potential to affect HD pathophysiology included plasma metabolome differences of gut-derived metabolites between controls and premanifest HD individuals or early symptomatic HD individuals.[313] Research comparing HD mice models to wild-type controls showed gut bacteria compositional differences at the phylum level, and increased diversity in only male HD mice compared to controls.[314] In a follow-up study authors longitudinally assessed fecal microbiome data for wild-type and HD mice models via shotgun sequencing,[255] and at phylum and family levels no gut bacterial differences were observed between groups, across any time point. However, between group differences were observed for gut microbiome function at week 12 (prior to motor symptom onset) with a greater relative abundance of sulfur metabolism, lysine degradation, glutathione metabolism and butanoate metabolism in the HD compared to wild-type mice. Greater butanoate metabolism did not correspond to greater plasma butyrate levels in the HD group (in fact authors note a non-significant [p = 0.11] decrease in the HD mice), nor other SCFAs. Authors discussed that pathogenic butyrate producers, such as *Fusobacterium*, may preferentially use lysine over pyruvate for butyrate production, which releases toxic byproducts, i.e., ammonia,[315] with this perhaps relating to the increased lysine degradation pathways they observed. The same lab also conducted a fiber intervention in HD mice,[256] reporting several benefits of the high-fiber diet to HD mice, including improvements to affective and cognitive deficits and improved GI function. Within the HD mice groups exposed to the different diets (no fiber, control, and high fiber), there were no specific changes to the gut microbiome (study used 16S sequencing) observed at 14 or 20 weeks of age. Comparisons between wild-type and HD mice exposed to the high-fiber diet revealed gut microbiome abundance differences observed at 14-weeks only at the phyla and family level. Examples of these included Actinobacteriota and Proteobacteria being decreased in the HD mice in the high fiber group compared to the high fiber wild-type mice. At the family level Bacteroidaceae,

Butyricicoccaceae, and Ruminococcaceae had increased abundance in the high fiber HD mice when compared to wild-type high fiber mice. Additionally, FMT from wild type to R6/1 HD model mice positively altered cognitive outcomes.[257] This indicates the potential for FMT to effectively treat some of the symptoms associated with HD while also reinforcing the likely involvement of the gut microbiota within HD.

Comparisons of HD patients to controls revealed, that the HD group had lower diversity, while sex-specific differences at the phylum and family level, were only observed for males.[316] Several inferred functional pathways (via PICRUSt) were indicated to be involved in HD; Starch degradation V, Methylerythritol phosphate I and II, and NAD biosynthesis I were all greater (in terms of relative abundance) in HD group. Research investigating the gut microbiome and cytokine levels of HD patients and controls did not reveal any sex-specific effects observed previously, but found several genera (*Intestinimonas, Bilophila, Lactobacillus, Oscillibacter, Gemmiger, and Dialister*) had greater abundance in the HD compared to control group.[317] Additionally, Intestinimonas was positively correlated with total functional capacity scores, while *Lactobacillus* was negatively associated with MMSE score. Across 10 plasma inflammatory markers only IL-4 differed, being greater in the control compared to HD group. Associations between bacteria and cytokines were completed in the HD group only, with *Intestinimonas* positively correlated with plasma IL-4, and *Porphyromonas* positively correlated with IL-4, IL-10, and IL-13, while *Bilophia, Oscillibacter and Gemmiger* negatively associated to IL-6. A review that details much of the above discussed evidence for involvement of the microbiota-gut-brain axis in MS, and incorporates a deeper discussion of potential mechanisms can be found here.[318]

4 Conclusions and Future directions

The microbiome-gut-brain axis research field has expanded horizons significantly since the initial focus on conducting compositional and correlational analyses. While such studies provided important impetus to the field, the past decade has answered the calls for more mechanistic studies and started to address the need to translate promising studies from animal models to humans. Collectively, these studies have further shown the importance of the gut microbiome to neurodevelopment and neurodegeneration.

These advances have seen a proliferation in clinical research in neurodevelopmental disorders including ASD, ADHD and schizophrenia. This research has largely resonated with the insights from earlier animal research, linking time-window specific microbiome configurations to infant temperament and cognition, and animal models work continues to document mechanistic insights, including the role of the vagus nerve.[75] It is anticipated that animal research will ultimately accrue the information necessary to pinpoint of targeted temporal microbial manipulations in neurodevelopmental disorders,[65] although there are still many pieces missing in this puzzle .

In neurodegeneration research, there is now strong causal evidence linking the gut microbiome to symptom expression in Alzheimer's,[246] and PD [252] using human-to-animal FMT. Such approaches could form the basis of new gut-brain animal models of these disorders to allow the assessment of possible microbiota interventions. A greater amount of clinical work investigating Alzheimer's, PD, ALS, HD, and MS has also been conducted, with longitudinal research becoming more prevalent. Some burning questions remain, including the interface between biological sex and the gut microbiome in these disorders, the role of host genetics, the gut virome's role in neurodegeneration including PD,[319] and whether the gut microbiome during prodromal stages could be predictive of future psychopathology and be amenable to prevention strategies.[320] Moreover, the issue of psychiatric comorbidity and possible transdiagnostic microbial profiles also requires further resolution.

While the recent research advances represent important progress, there are still fundamental knowledge gaps and obstacles to overcome. For example, there is still no consensus on what a 'healthy microbiome' is, either during adulthood or at the extremes of life, and developing this understanding is paramount to exploiting the

diagnostic and therapeutic potential of recent observations. Future neurodevelopment research should include more translational human intervention research, such as evaluating the potential of probiotics, prebiotics, postbiotics or FMT interventions. Understanding and overcoming the undesirable impacts of factors disrupting the gut microbiome such as maternal and infant antibiotic usage, and an increased focus on researching the potential benefits and safer application of 'seeding' an infant's microbiome via the mother's vaginal microbiome following C-section are warranted.[81] While ALS, HD and MS research in the microbiota-gut-brain axis is expanding, a clearer focus on mechanism and causation is needed, akin to that demonstrated in in Alzheimer's and PD, and overall, the neurodegeneration field would benefit from longitudinal research assessing humans in prodromal phases prior to and after disease diagnosis, to define disease-relevant gut microbiota changes. In these complex disorders, the microbiota is increasingly recognized as a key pathophysiological component. Knowing how and why the gut microbiome changes at the extremes of life will further develop our mechanistic understanding and help build the evidence base as we strive toward counteracting microbial missteps with precision therapeutic interventions.

Disclosure Statement

SKS has received speaker's and authors' honoraria from Medice Arzneimittel Pütter GmbH Co KG, Janssen and Takeda. J.F.C. has spoken at conferences organized by Bromotech and has received research funding from Reckitt, Nutricia, Dupont/IFF, and Nestle. G.C. received honoraria from Janssen, Probi, and Apsen and research funding from Pharmavite, Fonterra, Reckitt, Tate and Lyle and Nestle and is or has been a paid consultant for Yakult, Zentiva and Heel Pharmaceuticals. This support did not influence or constrain this review. The other authors report there are no competing interests to declare.

Funding

APC Microbiome Ireland is a research centre funded by Science Foundation Ireland (SFI; (SFI/12/RC/2273_P2)). NN receives project funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 101034270.

ORCID

Nathan D. Nuzum http://orcid.org/0000-0002-8627-6192
Clara Deady http://orcid.org/0000-0001-8857-3783
Sarah Kittel-Schneider http://orcid.org/0000-0003-3057-6150
John F. Cryan http://orcid.org/0000-0001-5887-2723
Siobhain M. O'Mahony http://orcid.org/0000-0002-9597-9391
Gerard Clarke http://orcid.org/0000-0001-9771-3979

Data availability statement

Data sharing not applicable to this article as no datasets were generated or analysed during the current study.

References

1. Cryan JF, O'Riordan KJ, Cowan CSM, Sandhu KV, Bastiaanssen TFS, Boehme M, Codagnone MG, Cussotto S, Fulling C, Golubeva AV, et al. The Microbiota-Gut-Brain Axis. Physiol Rev. 2019;99:1877–2013. doi:10.1152/physrev.00018.2018.
2. Goodwin S, McPherson JD, McCombie WR. Coming of age: ten years of next-generation sequencing technologies. Nat Rev Genet. 2016;17:333–351. doi:10.1038/nrg.2016.49.
3. Bastiaanssen TFS, Quinn TP, Loughman A. Bugs as features (part 1): concepts and foundations for the compositional data analysis of the microbiome–gut–brain axis. Nature Mental Health. 2023b;1(12):930–938. doi:10.1038/s44220-023-00148-3.
4. Bastiaanssen TFS, Quinn TP, Loughman A. Bugs as features (part 2): a perspective on enriching microbiome–gut–brain axis analyses. Nature Mental Health. 2023c;1(12):939–949. doi:10.1038/s44220-023-00149-2.
5. Lozupone CA, Stombaugh JI, Gordon JI, Jansson JK, Knight R. Diversity, stability and resilience of the human gut microbiota. Nature. 2012;489:220–230. doi:10.1038/nature11550.
6. Nishijima S, Suda W, Oshima K, Kim S-W, Hirose Y, Morita H, Hattori M. The gut microbiome of healthy Japanese and its microbial and functional uniqueness. DNA Res. 2016;23:125–133. doi:10.1093/dnares/dsw002.
7. Yatsunenko T, Rey FE, Manary MJ, Trehan I, Dominguez-Bello MG, Contreras M, Magris M, Hidalgo G, Baldassano RN, Anokhin AP, et al. Human gut microbiome viewed across age and geography. Nature. 2012;486:222–227. doi:10.1038/nature11053.
8. Odamaki T, Kato K, Sugahara H, Hashikura N, Takahashi S, Xiao J, Abe F, Osawa R. Age-related changes in gut microbiota composition from newborn to centenarian: a cross-sectional study. BMC Microbiol. 2016;16:90. doi:10.1186/s12866-016-0708-5.

9. Ho LKH, Tong VJW, Syn N, Nagarajan N, Tham EH, Tay SK, Shorey S, Tambyah PA, Law ECN. Gut microbiota changes in children with autism spectrum disorder: a systematic review. Gut Pathog. 2020;12:6. doi:10.1186/s13099-020-0346-1.
10. Nuzum ND, Loughman A, Szymlek-Gay EA, Hendy A, Teo W-P, Macpherson H. Gut microbiota differences between healthy older adults and individuals with Parkinson's disease: a systematic review. Neurosci Biobehav Rev. 2020;112:227–241. doi:10.1016/j.neubiorev.2020.02.003.
11. Sochocka M, Donskow-Łysoniewska K, Diniz BS, Kurpas D, Brzozowska E, Leszek J. The Gut Microbiome Alterations and Inflammation-Driven Pathogenesis of Alzheimer's Disease—a Critical Review. Mol Neurobiol. 2019;56:1841–1851. doi:10.1007/s12035-018-1188-4.
12. Vogt NM, Kerby RL, Dill-McFarland KA, Harding SJ, Merluzzi AP, Johnson SC, Carlsson CM, Asthana S, Zetterberg H, Blennow K, et al. Gut microbiome alterations in Alzheimer's disease. Sci Rep. 2017;7:13537. doi:10.1038/s41598-017-13601-y.
13. Brüssow H. Problems with the concept of gut microbiota dysbiosis. Microb Biotechnol. 2020;13(2):423–434. doi:10.1111/1751-7915.13479.
14. Cani PD. Gut microbiota — at the intersection of everything?. Nat Rev Gastroenterol Hepatol. 2017;14(6):321–322. doi:10.1038/nrgastro.2017.54.
15. Fischbach MA. Microbiome: focus on causation and mechanism. Cell. 2018;174:785–790. doi:10.1016/j.cell.2018.07.038.
16. Bokoliya SC, Dorsett Y, Panier H, Zhou Y. Procedures for Fecal Microbiota Transplantation in Murine Microbiome Studies. Front Cell Inf Microbiol. 2021;11. doi:10.3389/fcimb.2021.711055.
17. Gheorghe CE, Ritz NL, Martin JA, Wardill HR, Cryan JF, Clarke G. Investigating causality with fecal microbiota transplantation in rodents: applications, recommendations and pitfalls. Gut Microbes. 2021;13:1941711. doi:10.1080/19490976.2021.1941711.
18. Secombe KR, Al-Qadami GH, Subramaniam CB, Bowen JM, Scott J, Van Sebille YZA, Snelson M, Cowan C, Clarke G, Gheorghe CE, et al. Guidelines for reporting on animal fecal transplantation (GRAFT) studies: recommendations from a systematic review of murine transplantation protocols. Gut Microbes. 2021;13:1979878. doi:10.1080/19490976.2021.1979878.
19. Shtossel O, Turjeman S, Riumin A, Goldberg MR, Elizur A, Bekor Y, Mor H, Koren O, Louzoun Y. Recipient-independent, high-accuracy FMT-response prediction and optimization in mice and humans. Microbiome. 2023;11:181. doi:10.1186/s40168-023-01623-w.
20. Luczynski P, McVey Neufeld K-A, Oriach CS, Clarke G, Dinan TG, Cryan JF. Growing up in a Bubble: using Germ-Free Animals to Assess the Influence of the Gut Microbiota on Brain and Behavior. Int J Neuropsychopharmacol. 2016;19:yw020. doi:10.1093/ijnp/pyw020.
21. Leeming ER, Johnson AJ, Spector TD, Le Roy CI. Effect of Diet on the Gut Microbiota: rethinking Intervention Duration. Nutrients. 2019;11:2862. doi:10.3390/nu11122862.
22. Dziewiecka H, Buttar HS, Kasperska A, Ostapiuk–Karolczuk J, Domagalska M, Cichoń J, Skarpańska-Stejnborn A. Physical activity induced alterations of gut microbiota in humans: a systematic review. BMC Sports Sci Med Rehabil. 2022;14:122. doi:10.1186/s13102-022-00513-2.
23. Gibson GR, Hutkins R, Sanders ME, Prescott SL, Reimer RA, Salminen SJ, Scott K, Stanton C, Swanson KS, Cani PD, et al. Expert consensus document: the International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat Rev Gastroenterol Hepatol. 2017;14:491–502. doi:10.1038/nrgastro.2017.75.
24. Swanson KS, Gibson GR, Hutkins R, Reimer RA, Reid G, Verbeke K, Scott KP, Holscher HD, Azad MB, Delzenne NM, et al. The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of synbiotics. Nat Rev Gastroenterol Hepatol. 2020;17:687–701. doi:10.1038/s41575-020-0344-2.
25. Bastiaanssen TFS, Leigh S-J, Tofani GSS, Gheorghe CE, Clarke G, Cryan JF. Kronos: a computational tool to facilitate biological rhythmicity analysis; 2023a. doi:10.1101/2023.04.21.537503.
26. Gheorghe CE, Leigh S-J, Tofani GSS, Bastiaanssen TFS, Lyte JM, Gardellin E, Govindan A, Strain C, Martinez-Herrero S, Goodson MS, et al. The microbiota drives diurnal rhythms in tryptophan metabolism in the stressed gut. Cell Rep. 2024:43. doi:10.1016/j.celrep.2024.114079.
27. Valles-Colomer M, Falony G, Darzi Y, Tigchelaar EF, Wang J, Tito RY, Schiweck C, Kurilshikov A, Joossens M, Wijmenga C, et al. The neuroactive potential of the human gut microbiota in quality of life and depression. Nat Microbiol. 2019;4:623–632. doi:10.1038/s41564-018-0337-x.
28. Mirzayi C, Renson A, Zohra F, Elsafoury S, Geistlinger L, Kasselman LJ, Eckenrode K, van de Wijgert J, Loughman A, Marques FZ, et al. Reporting guidelines for human microbiome research: the STORMS checklist. Nat Med. 2021;27:1885–1892. doi:10.1038/s41591-021-01552-x.
29. Beghini F, McIver LJ, Blanco-Míguez A, Dubois L, Asnicar F, Maharjan S, Mailyan A, Manghi P, Scholz M, Thomas AM, et al. Integrating taxonomic, functional, and strain-level profiling of diverse microbial communities with bioBakery 3. eLife. 2021;10:e65088. doi:10.7554/eLife.65088.

30. Vieira-Silva S, Falony G, Darzi Y, Lima-Mendez G, Garcia Yunta R, Okuda S, Vandeputte D, Valles-Colomer M, Hildebrand F, Chaffron S, et al. Species–function relationships shape ecological properties of the human gut microbiome. Nat Microbiol. 2016;1:1–8. doi:10.1038/nmicrobiol.2016.88.
31. Ansari F, Neshat M, Pourjafar H, Jafari SM, Samakkhah SA, Mirzakhani E. The role of probiotics and prebiotics in modulating of the gut-brain axis. Front Nutr. 2023;10. doi:10.3389/fnut.2023.1173660.
32. Berding K, Bastiaanssen TFS, Moloney GM, Boscaini S, Strain CR, Anesi A, Long-Smith C, Mattivi F, Stanton C, Clarke G, et al. Feed your microbes to deal with stress: a psychobiotic diet impacts microbial stability and perceived stress in a healthy adult population. Mol Psychiatry. 2023;28(2):601–610. doi:10.1038/s41380-022-01817-y.
33. Allen JM, Mailing LJ, Niemiro GM, Moore R, Cook MD, White BA, Holscher HD, Woods JA. Exercise Alters Gut Microbiota Composition and Function in Lean and Obese Humans. Med Sci Sports Exerc. 2018;50(4):747–757. doi:10.1249/MSS.0000000000001495.
34. Gubert C, Kong G, Renoir T, Hannan AJ. Exercise, diet and stress as modulators of gut microbiota: implications for neurodegenerative diseases. Neurobiol Dis. 2020;134:104621. doi:10.1016/j.nbd.2019.104621.
35. Codagnone MG, Spichak S, O'Mahony SM, O'Leary OF, Clarke G, Stanton C, Dinan TG, Cryan JF. Programming Bugs: microbiota and the Developmental Origins of Brain Health and Disease. Biol Psychiatry. 2019;85:150–163. doi:10.1016/j.biopsych.2018.06.014.
36. Jašarević E, Bale TL. Prenatal and postnatal contributions of the maternal microbiome on offspring programming. Front Neuroendocrinol. 2019;55:100797. doi:10.1016/j.yfrne.2019.100797.
37. Hung -C-C, Chang -C-C, Huang C-W, Nouchi R, Cheng C-H. Gut microbiota in patients with Alzheimer's disease spectrum: a systematic review and meta-analysis. Aging (Albany NY). 2022;14:477–496. doi:10.18632/aging.203826.
38. Jiang C, Li G, Huang P, Liu Z, Zhao B. The Gut Microbiota and Alzheimer's Disease. J Alzheimers Dis. 2017;58:1–15. doi:10.3233/JAD-161141.
39. Nishiwaki H, Ito M, Ishida T, Hamaguchi T, Maeda T, Kashihara K, Tsuboi Y, Ueyama J, Shimamura T, Mori H, et al. Meta-Analysis of Gut Dysbiosis in Parkinson's Disease. Mov Disord. 2020b;35:1626–1635. doi:10.1002/mds.28119.
40. Romano S, Savva GM, Bedarf JR, Charles IG, Hildebrand F, Narbad A. Meta-analysis of the Parkinson's disease gut microbiome suggests alterations linked to intestinal inflammation. npj Parkinsons Dis. 2021;7:1–13. doi:10.1038/s41531-021-00156-z.
41. Morton JT, Jin D-M, Mills RH, Shao Y, Rahman G, McDonald D, Zhu Q, Balaban M, Jiang Y, Cantrell K, et al. Multi-level analysis of the gut–brain axis shows autism spectrum disorder-associated molecular and microbial profiles. Nat Neurosci. 2023;26:1208–1217. doi:10.1038/s41593-023-01361-0.
42. Douglas GM, Maffei VJ, Zaneveld JR, Yurgel SN, Brown JR, Taylor CM, Huttenhower C, Langille MGI. PICRUSt2 for prediction of metagenome functions. Nat Biotechnol. 2020;38:685–688. doi:10.1038/s41587-020-0548-6.
43. Spichak S, Bastiaanssen TFS, Berding K, Vlckova K, Clarke G, Dinan TG, Cryan JF. Mining microbes for mental health: determining the role of microbial metabolic pathways in human brain health and disease. Neurosci Biobehav Rev. 2021;125:698–761. doi:10.1016/j.neubiorev.2021.02.044.
44. Li L, Wang T, Ning Z, Zhang X, Butcher J, Serrana JM, Simopoulos CMA, Mayne J, Stintzi A, Mack DR, et al. Revealing proteome-level functional redundancy in the human gut microbiome using ultra-deep metaproteomics. Nat Commun. 2023;14:3428. doi:10.1038/s41467-023-39149-2.
45. Minkoff NZ, Aslam S, Medina M, Tanner-Smith EE, Zackular JP, Acra S, Nicholson MR, Imdad A. Fecal microbiota transplantation for the treatment of recurrent *Clostridioides difficile* (*Clostridium difficile*). Cochr Database Syst Rev. 2023. doi:10.1002/14651858.CD013871.pub2.
46. Merrick B, Allen L, Masirah M, Zain N, Forbes B, Shawcross DL, Goldenberg SD. Regulation, risk and safety of Faecal Microbiota Transplant. Infect Prev Pract. 2020;2:100069. doi:10.1016/j.infpip.2020.100069.
47. Park S-Y, Seo GS. Fecal Microbiota Transplantation: is It Safe?. Clin Endosc. 2021;54:157–160. doi:10.5946/ce.2021.072.
48. Moon LD, Xiong F. Mechanics of neural tube morphogenesis. Semin. Cell Dev. Biol. 2022;130:56–69. doi:10.1016/j.semcdb.2021.09.009.
49. Avagliano L, Massa V, George TM, Qureshy S, Bulfamante G, Finnell RH. Overview on Neural tube defects: from development to physical characteristics. Birth Defects Research. 2019;111(19):1455–1467. doi:10.1002/bdr2.1380.
50. Gressens P. Mechanisms and Disturbances of Neuronal Migration. Pediatr Res. 2000;48:725–730. doi:10.1203/00006450-200012000-00004.
51. Morell P, Quarles RH. Developmental Biology of Myelin. In: Basic Neurochemistry: molecular, Cellular and Medical Aspects 6th. Lippincott-Raven; 1999.
52. Thomason ME. Development of Brain Networks In Utero: relevance for Common Neural Disorders. Biol Psychiatry. 2020;88:40–50. doi:10.1016/j.biopsych.2020.02.007.
53. Galvez-Contreras AY, Zarate-Lopez D, Torres-Chavez AL, Gonzalez-Perez O. Role of Oligodendrocytes and Myelin in the Pathophysiology of Autism Spectrum Disorder. Brain Sci. 2020;10:951. doi:10.3390/brainsci10120951.

54. Pan Y-H, Wu N, Yuan X-B. Toward a Better Understanding of Neuronal Migration Deficits in Autism Spectrum Disorders. Front Cell Dev Biol. 2019;7:205. doi:10.3389/fcell.2019.00205.
55. McDonald B, McCoy KD. Maternal microbiota in pregnancy and early life. Science. 2019;365:984–985. doi:10.1126/science.aay0618.
56. Mueller NT, Bakacs E, Combellick J, Grigoryan Z, Dominguez-Bello MG. The infant microbiome development: mom matters. Trends Mol Med. 2015;21:109–117. doi:10.1016/j.molmed.2014.12.002.
57. O'Mahony SM, Clarke G, Dinan TG, Cryan JF. Early-life adversity and brain development: is the microbiome a missing piece of the puzzle?. Neuroscience, Early Adversity and Brain Development. 2017;342:37–54. doi:10.1016/j.neuroscience.2015.09.068.
58. Bhagavata Srinivasan SP, Raipuria M, Bahari H, Kaakoush NO, Morris MJ. Impacts of Diet and Exercise on Maternal Gut Microbiota Are Transferred to Offspring. Front Endocrinol. 2018;9. doi:10.3389/fendo.2018.00716.
59. Leyrolle Q, Decoeur F, Briere G, Amadieu C, Quadros ARAA, Voytyuk I, Lacabanne C, Benmamar-Badel A, Bourel J, Aubert A, et al. Maternal dietary omega-3 deficiency worsens the deleterious effects of prenatal inflammation on the gut-brain axis in the offspring across lifetime. Neuropsychopharmacology. 2021;46:579–602. doi:10.1038/s41386-020-00793-7.
60. Sanguinetti E, Guzzardi MA, Tripodi M, Panetta D, Selma-Royo M, Zega A, Telleschi M, Collado MC, Iozzo P. Microbiota signatures relating to reduced memory and exploratory behaviour in the offspring of overweight mothers in a murine model. Sci Rep. 2019;9:12609. doi:10.1038/s41598-019-48090-8.
61. Leclercq S, Mian FM, Stanisz AM, Bindels LB, Cambier E, Ben-Amram H, Koren O, Forsythe P, Bienenstock J. Low-dose penicillin in early life induces long-term changes in murine gut microbiota, brain cytokines and behavior. Nat Commun. 2017;8:15062. doi:10.1038/ncomms15062.
62. Champagne-Jorgensen K, Mian MF, Kay S, Hanani H, Ziv O, McVey Neufeld K-A, Koren O, Bienenstock J. Prenatal low-dose penicillin results in long-term sex-specific changes to murine behaviour, immune regulation, and gut microbiota. Brain Behav Immun. 2020;84:154–163. doi:10.1016/j.bbi.2019.11.020.
63. O'Connor R, Moloney GM, Fulling C, O'Riordan KJ, Fitzgerald P, Bastiaanssen TFS, Schellekens H, Dinan TG, Cryan JF. Maternal antibiotic administration during a critical developmental window has enduring neurobehavioural effects in offspring mice. Behav Brain Res. 2021;404:113156. doi:10.1016/j.bbr.2021.113156.
64. Vuong HE, Pronovost GN, Williams DW, Coley EJL, Siegler EL, Qiu A, Kazantsev M, Wilson CJ, Rendon T, Hsiao EY. The maternal microbiome modulates fetal neurodevelopment in mice. Nature. 2020;586:281–286. doi:10.1038/s41586-020-2745-3.
65. Lynch CMK, Cowan CSM, Bastiaanssen TFS, Moloney GM, Theune N, van de Wouw M, Florensa Zanuy E, Ventura-Silva AP, Codagnone MG, Villalobos-Manríquez F, et al. Critical windows of early-life microbiota disruption on behaviour, neuroimmune function, and neurodevelopment. Brain Behav Immun. 2023;108:309–327. doi:10.1016/j.bbi.2022.12.008.
66. Kayyal M, Javkar T, Firoz Mian M, Binyamin D, Koren O, McVey Neufeld K-A, Forsythe P. Sex dependent effects of post-natal penicillin on brain, behavior and immune regulation are prevented by concurrent probiotic treatment. Sci Rep. 2020;10:10318. doi:10.1038/s41598-020-67271-4.
67. Gur TL, Shay L, Palkar AV, Fisher S, Varaljay VA, Dowd S, Bailey MT. Prenatal stress affects placental cytokines and neurotrophins, commensal microbes, and anxiety-like behavior in adult female offspring. Brain Behav Immun. 2017;64:50–58. doi:10.1016/j.bbi.2016.12.021.
68. Gur TL, Palkar AV, Rajasekera T, Allen J, Niraula A, Godbout J, Bailey MT. Prenatal stress disrupts social behavior, cortical neurobiology and commensal microbes in adult male offspring. Behav Brain Res. 2019;359:886–894. doi:10.1016/j.bbr.2018.06.025.
69. Nicolas S, McGovern AJ, Hueston CM, O'Mahony SM, Cryan JF, O'Leary OF, Nolan YM. Prior maternal separation stress alters the dendritic complexity of new hippocampal neurons and neuroinflammation in response to an inflammatory stressor in juvenile female rats. Brain Behav Immun. 2022;99:327–338. doi:10.1016/j.bbi.2021.10.016.
70. Collins JM, Caputi V, Manurung S, Gross G, Fitzgerald P, Golubeva AV, Popov J, Deady C, Dinan TG, Cryan JF, et al. Supplementation with milk fat globule membrane from early life reduces maternal separation-induced visceral pain independent of enteric nervous system or intestinal permeability changes in the rat. Neuropharmacology. 2022;210:109026. doi:10.1016/j.neuropharm.2022.109026.
71. O'Mahony SM, McVey Neufeld K-A, Waworuntu RV, Pusceddu MM, Manurung S, Murphy K, Strain C, Laguna MC, Peterson VL, Stanton C, et al. The enduring effects of early-life stress on the microbiota-gut-brain axis are buffered by dietary supplementation with milk fat globule membrane and a prebiotic blend. Eur J Neurosci. 2020;51:1042–1058. doi:10.1111/ejn.14514.
72. McVey Neufeld K-A, O'Mahony SM, Hoban AE, Waworuntu RV, Berg BM, Dinan TG, Cryan JF. Neurobehavioural effects of Lactobacillus rhamnosus GG alone and in combination with prebiotics polydextrose and galactooligosaccharide in male rats exposed to early-life stress. Nutr Neurosci. 2019;22:425–434. doi:10.1080/1028415X.2017.1397875.
73. Morais LH, Golubeva AV, Moloney GM, Moya-Pérez A, Ventura-Silva AP, Arboleya S, Bastiaanssen TFS, O'Sullivan O, Rea K, Borre Y, et al. Enduring

Behavioral Effects Induced by Birth by Caesarean Section in the Mouse. Curr Biol. 2020;30:3761–3774.e6. doi:10.1016/j.cub.2020.07.044.
74. Sharon G, Cruz NJ, Kang D-W, Gandal MJ, Wang B, Kim Y-M, Zink EM, Casey CP, Taylor BC, Lane CJ, et al. Human Gut Microbiota from Autism Spectrum Disorder Promote Behavioral Symptoms in Mice. Cell. 2019;177:1600–1618.e17. doi:10.1016/j.cell.2019.05.004.
75. Sgritta M, Dooling SW, Buffington SA, Momin EN, Francis MB, Britton RA, Costa-Mattioli M. Mechanisms Underlying Microbial-Mediated Changes in Social Behavior in Mouse Models of Autism Spectrum Disorder. Neuron. 2019;101:246–259.e6. doi:10.1016/j.neuron.2018.11.018.
76. Dervola KS, Roberg BÅ, Wøien G, Bogen IL, Sandvik TH, Sagvolden T, Drevon CA, Johansen EB, Walaas SI. Marine omega-3 polyunsaturated fatty acids induce sex-specific changes in reinforcer-controlled behaviour and neurotransmitter metabolism in a spontaneously hypertensive rat model of ADHD. Behav Brain Funct. 2012;8:56. doi:10.1186/1744-9081-8-56.
77. Zhu F, Guo R, Wang W, Ju Y, Wang Q, Ma Q, Sun Q, Fan Y, Xie Y, Yang Z, et al. Transplantation of microbiota from drug-free patients with schizophrenia causes schizophrenia-like abnormal behaviors and dysregulated kynurenine metabolism in mice. Mol Psychiatry. 2020a;25:2905–2918. doi:10.1038/s41380-019-0475-4.
78. Slykerman RF, Coomarasamy C, Wickens K, Thompson JMD, Stanley TV, Barthow C, Kang J, Crane J, Mitchell EA. Exposure to antibiotics in the first 24 months of life and neurocognitive outcomes at 11 years of age. Psychopharmacology. 2019;236:1573–1582. doi:10.1007/s00213-019-05216-0.
79. Aatsinki A-K, Keskitalo A, Laitinen V, Munukka E, Uusitupa H-M, Lahti L, Kortesluoma S, Mustonen P, Rodrigues AJ, Coimbra B, et al. Maternal prenatal psychological distress and hair cortisol levels associate with infant fecal microbiota composition at 2.5 months of age. Psychoneuroendocrinology. 2020;119:104754. doi:10.1016/j.psyneuen.2020.104754.
80. Korpela K, Helve O, Kolho K-L, Saisto T, Skogberg K, Dikareva E, Stefanovic V, Salonen A, Andersson S, de Vos WM. Maternal Fecal Microbiota Transplantation in Cesarean-Born Infants Rapidly Restores Normal Gut Microbial Development: a Proof-of-Concept Study. Cell. 2020;183:324–334.e5. doi:10.1016/j.cell.2020.08.047.
81. Zhou L, Qiu W, Wang J, Zhao A, Zhou C, Sun T, Xiong Z, Cao P, Shen W, Chen J, et al. Effects of vaginal microbiota transfer on the neurodevelopment and microbiome of cesarean-born infants: a blinded randomized controlled trial. Cell Host Microbe. 2023;31:1232–1247.e5. doi:10.1016/j.chom.2023.05.022.
82. Wilson BC, Butler ÉM, Grigg CP, Derraik JGB, Chiavaroli V, Walker N, Thampi S, Creagh C, Reynolds AJ, Vatanen T, et al. Oral administration of maternal vaginal microbes at birth to restore gut microbiome development in infants born by caesarean section: a pilot randomised placebo-controlled trial. EBioMedicine. 2021;69:103443. doi:10.1016/j.ebiom.2021.103443.
83. Shaaban SY, El Gendy YG, Mehanna NS, El-Senousy WM, El-Feki HSA, Saad K, El-Asheer OM. The role of probiotics in children with autism spectrum disorder: a prospective, open-label study. Nutr Neurosci. 2018;21:676–681. doi:10.1080/1028415X.2017.1347746.
84. Kang D-W, Adams JB, Gregory AC, Borody T, Chittick L, Fasano A, Khoruts A, Geis E, Maldonado J, McDonough-Means S, et al. Microbiota Transfer Therapy alters gut ecosystem and improves gastrointestinal and autism symptoms: an open-label study. Microbiome. 2017;5:10. doi:10.1186/s40168-016-0225-7.
85. Kumperscak HG, Gricar A, Ülen I, Micetic-Turk D. A Pilot Randomized Control Trial With the Probiotic Strain Lactobacillus rhamnosus GG (LGG) in ADHD: children and Adolescents Report Better Health-Related Quality of Life. Front Psychiatry. 2020;11:181. doi:10.3389/fpsyt.2020.00181.
86. Skott E, Yang LL, Stiernborg M, Söderström Å, Rüegg J, Schalling M, Forsell Y, Giacobini M, Lavebratt C. Effects of a synbiotic on symptoms, and daily functioning in attention deficit hyperactivity disorder – a double-blind randomized controlled trial. Brain Behav Immun. 2020;89:9–19. doi:10.1016/j.bbi.2020.05.056.
87. Wang L-J, Yang C-Y, Kuo H-C, Chou W-J, Tsai C-S, Lee S-Y. Effect of Bifidobacterium bifidum on Clinical Characteristics and Gut Microbiota in Attention-Deficit/Hyperactivity Disorder. J Pers Med. 2022;12:227. doi:10.3390/jpm12020227.
88. Aatsinki A-K, Lahti L, Uusitupa H-M, Munukka E, Keskitalo A, Nolvi S, O'Mahony S, Pietilä S, Elo LL, Eerola E, et al. Gut microbiota composition is associated with temperament traits in infants. Brain Behav Immun. 2019;80:849–858. doi:10.1016/j.bbi.2019.05.035.
89. Delgadillo DR, Pressman SD, Christian LM, Galley JD, Bailey MT. Associations Between Gut Microbes and Social Behavior in Healthy 2-Year-Old Children. Psychosom Med. 2022;84:749–756. doi:10.1097/PSY.0000000000001103.
90. Huovinen V, Aatsinki A-K, Kataja E-L, Munukka E, Keskitalo A, Lamichhane S, Raunioniemi P, Bridgett DJ, Lahti L, O'Mahony SM, et al. Infant gut microbiota and negative and fear reactivity. Dev Psychopathol. 2023:1–16. doi:10.1017/S0954579423001396.
91. Loughman A, Ponsonby A-L, O'Hely M, Symeonides C, Collier F, Tang MLK, Carlin J, Ranganathan S, Allen K, Pezic A, et al. Gut microbiota composition during infancy and subsequent behavioural outcomes. EBioMedicine. 2020;52:102640. doi:10.1016/j.ebiom.2020.102640.
92. Aatsinki A-K, Kataja E-L, Munukka E, Lahti L, Keskitalo A, Korja R, Nolvi S, Häikiö T, Tarro S,

Karlsson H, et al. Infant fecal microbiota composition and attention to emotional faces. Emotion. 2022;22 (6):1159–1170. doi:10.1037/emo0000924.

93. Carlson AL, Xia K, Azcarate-Peril MA, Goldman BD, Ahn M, Styner MA, Thompson AL, Geng X, Gilmore JH, Knickmeyer RC. Infant Gut Microbiome Associated With Cognitive Development. Biol Psychiatry. 2018;83(2):148–159. doi:10.1016/j.biopsych.2017.06.021.
94. Banchi P, Colitti B, Opsomer G, Rota A, Van Soom A. The dogma of the sterile uterus revisited: does microbial seeding occur during fetal life in humans and animals?. Reproduction. 2024;167(1):e230078. doi:10.1530/REP-23-0078.
95. Kennedy KM, de Goffau MC, Perez-Muñoz ME, Arrieta M-C, Bäckhed F, Bork P, Braun T, Bushman FD, Dore J, de Vos WM, et al. Questioning the fetal microbiome illustrates pitfalls of low-biomass microbial studies. Nature. 2023;613:639–649. doi:10.1038/s41586-022-05546-8.
96. Frerichs NM, de Meij TGJ, Niemarkt HJ. Microbiome and its impact on fetal and neonatal brain development: current opinion in pediatrics. Curr Opin Clin Nutr Metab Care. 2024;27:297–303. doi:10.1097/MCO.0000000000001028.
97. Aburto MR, Cryan JF. Gastrointestinal and brain barriers: unlocking gates of communication across the microbiota-gut-brain axis. Nat Rev Gastroenterol Hepatol. 2024;21(4):222–247. doi:10.1038/s41575-023-00890-0.
98. Li Y, Toothaker JM, Ben-Simon S, Ozeri L, Schweitzer R, McCourt BT, McCourt CC, Werner L, Snapper SB, Shouval DS, et al. In utero human intestine harbors unique metabolome, including bacterial metabolites. JCI Insight. 2020;5(e138751):138751. doi:10.1172/jci.insight.138751.
99. Argaw-Denboba A, Schmidt TSB, Di Giacomo M, Ranjan B, Devendran S, Mastrorilli E, Lloyd CT, Pugliese D, Paribeni V, Dabin J, et al. Paternal microbiome perturbations impact offspring fitness. Nature. 2024;629(8012):652–659. doi:10.1038/s41586-024-07336-w.
100. Masson BA, Kiridena P, Lu D, Kleeman EA, Reisinger SN, Qin W, Davies WJ, Muralitharan RR, Jama HA, Antonacci S, et al. Depletion of the paternal gut microbiome alters sperm small RNAs and impacts offspring physiology and behavior in mice. Brain Behav Immun. 2024. doi:10.1016/j.bbi.2024.09.020.
101. Kilama J, Dahlen CR, Reynolds LP, Amat S. Contribution of the seminal microbiome to paternal programming. Biol Reprod. 2024;111:242–268. doi:10.1093/biolre/ioae068.
102. Roigé-Castellví J, Murphy M, Fernández-Ballart J, Canals J. Moderately elevated preconception fasting plasma total homocysteine is a risk factor for psychological problems in childhood. Public Health Nutr. 2019;22:1615–1623. doi:10.1017/S1368980018003610.
103. Murray LK, Smith MJ, Jadavji NM. Maternal oversupplementation with folic acid and its impact on neurodevelopment of offspring. Nutr Rev. 2018;76:708–721. doi:10.1093/nutrit/nuy025.
104. Suzuki T, Nishigori T, Obara T, Masumoto T, Mori M, Murata T, Kyozuka H, Ogata Y, Sato A, Sampei M, et al.; Group, T.J.E. and C.S. Maternal folic acid supplement use/dietary folate intake from preconception to early pregnancy and neurodevelopment in 2-year-old offspring: the Japan Environment and Children's Study. Br J Nutr. 2022;128:2480–2489. doi:10.1017/S000711452200037X.
105. de Lauzon-Guillain B, Marques C, Kadawathagedara M, Bernard JY, Tafflet M, Lioret S, Charles MA. Maternal diet during pregnancy and child neurodevelopment up to age 3.5 years: the nationwide Étude Longitudinale Française depuis l'Enfance (ELFE) birth cohort. Am J Clin Nutr. 2022;116:1101–1111. doi:10.1093/ajcn/nqac206.
106. Veena SR, Gale CR, Krishnaveni GV, Kehoe SH, Srinivasan K, Fall CH. Association between maternal nutritional status in pregnancy and offspring cognitive function during childhood and adolescence; a systematic review. BMC Pregnancy Childbirth. 2016:16. doi:10.1186/s12884-016-1011-z.
107. Dahl WJ, Rivero Mendoza D, Lambert JM. Diet, nutrients and the microbiome. Prog Mol Biol Transl Sci. 2020;171:237–263. doi:10.1016/bs.pmbts.2020.04.006.
108. Trakman GL, Fehily S, Basnayake C, Hamilton AL, Russell E, Wilson-O'Brien A, Kamm MA. Diet and gut microbiome in gastrointestinal disease. J Gastroenterol Hepatol. 2022;37:237–245. doi:10.1111/jgh.15728.
109. Kyozuka H, Murata T, Fukuda T, Yamaguchi A, Kanno A, Yasuda S, Suzuki D, Takahashi T, Go H, Maeda H, et al. Association between preconception dietary inflammatory index and neurodevelopment of offspring at 3 years of age. Japan Env Children's Study Nutr. 2022;102:111708. doi:10.1016/j.nut.2022.111708.
110. Adane AA, Mishra GD, Tooth LR. Maternal pre-pregnancy obesity and childhood physical and cognitive development of children: a systematic review. Int J Obes. 2016;40(11):1608–1618. doi:10.1038/ijo.2016.140.
111. Liu B-N, Liu X-T, Liang Z-H, Wang J-H. Gut microbiota in obesity. World J Gastroenterol. 2021;27:3837. doi:10.3748/wjg.v27.i25.3837.
112. de Jonge L, Bos HJ, van Langen IM, de Jong-van den Berg LTW, Bakker MK. Antibiotics prescribed before, during and after pregnancy in the Netherlands: a drug utilization study. Pharmacoepidemiol Drug Saf. 2014;23:60–68. doi:10.1002/pds.3492.
113. Faas MM, Liu Y, Wekema L, Weiss GA, van Loo-Bouwman CA, Silva Lagos L. The Effect of Antibiotics Treatment on the Maternal Immune Response and Gut Microbiome in Pregnant and Non-Pregnant Mice. Nutrients. 2023;15:2723. doi:10.3390/nu15122723.

114. González-Arancibia C, Collio V, Silva-Olivares F, Montaña-Collao P, Martínez-Pinto J, Julio-Pieper M, Sotomayor-Zárate R, Bravo JA. Early-Life Exposure to Non-Absorbable Broad-Spectrum Antibiotics Affects the Dopamine Mesocorticolimbic Pathway of Adult Rats in a Sex-Dependent Manner. Front Pharmacol. 2022;13:837652. doi:10.3389/fphar.2022.837652.
115. Tochitani S, Ikeno T, Ito T, Sakurai A, Yamauchi T, Matsuzaki H. Administration of Non-Absorbable Antibiotics to Pregnant Mice to Perturb the Maternal Gut Microbiota Is Associated with Alterations in Offspring Behavior. PLOS ONE. 2016;11:e0138293. doi:10.1371/journal.pone.0138293.
116. Sarna JR, Furtado S, Brownell AKW. Neurologic Complications of Metronidazole. Can J Neurol Sci. 2013;40:768–776. doi:10.1017/S0317167100015870.
117. Njotto LL, Simin J, Fornes R, Odsbu I, Mussche I, Callens S, Engstrand L, Bruyndonckx R, Brusselaers N. Maternal and Early-Life Exposure to Antibiotics and the Risk of Autism and Attention-Deficit Hyperactivity Disorder in Childhood: a Swedish Population-Based Cohort Study. Drug Saf. 2023;46:467–478. doi:10.1007/s40264-023-01297-1.
118. Straughen JK, Sitarik AR, Wegienka G, Johnson CC, Johnson-Hooper TM, Cassidy-Bushrow AE. Association between prenatal antimicrobial use and offspring attention deficit hyperactivity disorder. PLOS ONE. 2023;18:e0285163. doi:10.1371/journal.pone.0285163.
119. Tao Q, Shen Y, Li Y, Luo H, Yuan M, Gan J. Prenatal exposure to antibiotics and risk of neurodevelopmental disorders in offspring: a systematic review and meta-analysis. Front Neurol. 2022;13:1045865. doi:10.3389/fneur.2022.1045865.
120. Tripathi N, Cotten CM, Smith PB. Antibiotic Use and Misuse in the Neonatal Intensive Care Unit. Clin Perinatol. 2012;39:61–68. doi:10.1016/j.clp.2011.12.003.
121. Stocker M, Klingenberg C, Navér L, Nordberg V, Berardi A, El Helou S, Fusch G, Bliss JM, Lehnick D, Dimopoulou V, et al. Less is more: antibiotics at the beginning of life. Nat Commun. 2023;14:2423. doi:10.1038/s41467-023-38156-7.
122. Slykerman RF, Neumann D, Underwood L, Hobbs M, Waldie KE. Age at first exposure to antibiotics and neurodevelopmental outcomes in childhood. Psychopharmacology. 2023;240:1143–1150. doi:10.1007/s00213-023-06351-5.
123. Gibson MK, Crofts TS, Dantas G. Antibiotics and the developing infant gut microbiota and resistome. Curr Opin Microbiol. 2015;27:51–56. doi:10.1016/j.mib.2015.07.007.
124. Morowitz MJ, Katheria AC, Polin RA, Pace E, Huang DT, Chang -C-CH, Yabes JG. The NICU Antibiotics and Outcomes (NANO) trial: a randomized multicenter clinical trial assessing empiric antibiotics and clinical outcomes in newborn preterm infants. Trials. 2022;23:428. doi:10.1186/s13063-022-06352-3.
125. Russell JT, Lauren Ruoss J, de la Cruz D, Li N, Bazacliu C, Patton L, McKinley KL, Garrett TJ, Polin RA, Triplett EW, et al. Antibiotics and the developing intestinal microbiome, metabolome and inflammatory environment in a randomized trial of preterm infants. Sci Rep. 2021;11:1943. doi:10.1038/s41598-021-80982-6.
126. Sherman MP, Zaghouani H, Niklas V. Gut microbiota, the immune system, and diet influence the neonatal gut-brain axis. Pediatr Res. 2015;77:127–135. doi:10.1038/pr.2014.161.
127. Slykerman RF, Thompson J, Waldie KE, Murphy R, Wall C, Mitchell EA. Antibiotics in the first year of life and subsequent neurocognitive outcomes. Acta Paediatrica. 2017;106:87–94. doi:10.1111/apa.13613.
128. Wimberley T, Agerbo E, Pedersen CB, Dalsgaard S, Horsdal HT, Mortensen PB, Thompson WK, Köhler-Forsberg O, Yolken RH. Otitis media, antibiotics, and risk of autism spectrum disorder. Autism Res. 2018;11:1432–1440. doi:10.1002/aur.2015.
129. Hamad AF, Alessi-Severini S, Mahmud SM, Brownell M, Kuo IF. Early childhood antibiotics use and autism spectrum disorders: a population-based cohort study. Int J Epidemiol. 2018;47:1497–1506. doi:10.1093/ije/dyy162.
130. Sandler RH, Finegold SM, Bolte ER, Buchanan CP, Maxwell AP, Väisänen M-L, Nelson MN, Wexler HM. Short-Term Benefit From Oral Vancomycin Treatment of Regressive-Onset Autism. J Child Neurol. 2000;15:429–435. doi:10.1177/088307380001500701.
131. Otten K, Keller L, Puiu AA, Herpertz-Dahlmann B, Seitz J, Kohn N, Edgar JC, Wagels L, Konrad K. Pre- and postnatal antibiotic exposure and risk of developing attention deficit hyperactivity disorder-A systematic review and meta-analysis combining evidence from human and animal studies. Neurosci Biobehav Rev. 2022;140:104776. doi:10.1016/j.neubiorev.2022.104776.
132. Galley JD, Chen HJ, Antonson AM, Gur TL. Prenatal stress-induced disruptions in microbial and host tryptophan metabolism and transport. Behav Brain Res. 2021;414:113471. doi:10.1016/j.bbr.2021.113471.
133. Sherwin E, Bordenstein SR, Quinn JL, Dinan TG, Cryan JF. Microbiota and the social brain. Science. 2019;366:eaar2016. doi:10.1126/science.aar2016.
134. Van Den Bergh BRH, Van Den Heuvel MI, Lahti M, Braeken M, De Rooij SR, Entringer S, Hoyer D, Roseboom T, Räikkönen K, King S, et al. Prenatal developmental origins of behavior and mental health: the influence of maternal stress in pregnancy. Neurosci Biobehav Rev. 2020;117:26–64. doi:10.1016/j.neubiorev.2017.07.003.
135. O'Mahony SM, Hyland NP, Dinan TG, Cryan JF. Maternal separation as a model of brain–gut axis dysfunction. Psychopharmacology. 2011;214:71–88.
136. Clarke G, O'Mahony SM, Hennessy AA, Ross P, Stanton C, Cryan JF, Dinan TG. Chain reactions: early-life stress alters the metabolic profile of plasma

polyunsaturated fatty acids in adulthood. Behav Brain Res. 2009;205(1):319–321. doi:10.1016/j.bbr.2009.07.008.
137. O'Mahony S, Chua ASB, Quigley EMM, Clarke G, Shanahan F, Keeling PWN, Dinan TG. Evidence of an enhanced central 5HT response in irritable bowel syndrome and in the rat maternal separation model. Neurogastroenterol Motil. 2008;20:680–688. doi:10.1111/j.1365-2982.2007.01065.x.
138. De Palma G, Blennerhassett P, Lu J, Deng Y, Park AJ, Green W, Denou E, Silva MA, Santacruz A, Sanz Y, et al. Microbiota and host determinants of behavioural phenotype in maternally separated mice. Nat Commun. 2015;6:7735. doi:10.1038/ncomms8735.
139. O'Mahony SM, Marchesi JR, Scully P, Codling C, Ceolho A-M, Quigley EMM, Cryan JF, Dinan TG. Early Life Stress Alters Behavior, Immunity, and Microbiota in Rats: implications for Irritable Bowel Syndrome and Psychiatric Illnesses. Biol Psychiatry. 2009;65:263–267. doi:10.1016/j.biopsych.2008.06.026.
140. Conti MG, Natale F, Stolfi I, Pedicino R, Boscarino G, Ajassa C, Cardilli V, Ciambra GL, Guadalupi L, Favata P, et al. Consequences of Early Separation of Maternal-Newborn Dyad in Neonates Born to SARS-CoV-2 Positive Mothers: an Observational Study. Int J Environ Res Public Health. 2021;18:5899. doi:10.3390/ijerph18115899.
141. WHO, 2023. Preterm birth [WWW Document]. [accessed 2024 April 05]. https://www.who.int/news-room/fact-sheets/detail/preterm-birth. (accessed 4.5.24).
142. Matthews LG, Walsh BH, Knutsen C, Neil JJ, Smyser CD, Rogers CE, Inder TE. Brain growth in the NICU: critical periods of tissue-specific expansion. Pediatr Res. 2018;83:976–981. doi:10.1038/pr.2018.4.
143. Seki D, Mayer M, Hausmann B, Pjevac P, Giordano V, Goeral K, Unterasinger L, Klebermaß-Schrehof K, De Paepe K, Van de Wiele T, et al. Aberrant gut-microbiota-immune-brain axis development in premature neonates with brain damage. Cell Host Microbe. 2021;29:1558–1572.e6. doi:10.1016/j.chom.2021.08.004.
144. Aguilar-Lopez M, Dinsmoor AM, Ho TTB, Donovan SM. A systematic review of the factors influencing microbial colonization of the preterm infant gut. Gut Microbes. 2021;13(1):1884514. doi:10.1080/19490976.2021.1884514.
145. Clarke G, Aatsinki A, O'Mahony SM. Brain development in premature infants: a bug in the programming system?. Cell Host Microbe. 2021;29(10):1477–1479. doi:10.1016/j.chom.2021.09.015.
146. Betrán AP, Ye J, Moller A-B, Zhang J, Gülmezoglu AM, Torloni MR. The Increasing Trend in Caesarean Section Rates: global. Regional and National Estimates: 1990-2014. PLOS ONE 11. 2016:e0148343. doi:10.1371/journal.pone.0148343.
147. Zhang T, Sidorchuk A, Sevilla-Cermeño L, Vilaplana-Pérez A, Chang Z, Larsson H, Mataix-Cols D, Fernández de la Cruz L. Association of Cesarean Delivery With Risk of Neurodevelopmental and Psychiatric Disorders in the Offspring. JAMA Network Open. 2019;2:e1910236. doi:10.1001/jamanetworkopen.2019.10236.
148. Zhang Y, Ogbu D, Garrett S, Xia Y, Sun J. Aberrant enteric neuromuscular system and dysbiosis in amyotrophic lateral sclerosis. Gut Microbes. 2021;13:1996848. doi:10.1080/19490976.2021.1996848.
149. Curran EA, Dalman C, Kearney PM, Kenny LC, Cryan JF, Dinan TG, Khashan AS. Association Between Obstetric Mode of Delivery and Autism Spectrum Disorder: a Population-Based Sibling Design Study. JAMA Psychiatry. 2015;72:935–942. doi:10.1001/jamapsychiatry.2015.0846.
150. Zheng X, Li R, Wang L, Yang H, Li L, Cui J, Zhao W, Yang Z, Zhang Q, Xu T, et al. The association of cesarean section with overweight and neurodevelopment of Chinese children aged 1–5 months. Front Pediatr. 2022;10:940422. doi:10.3389/fped.2022.940422.
151. Sznajder KK, Teti D, Hackman NM, Massare B, Kjerulff KH. Cesarean Section and Child Development at 3 Years: a Prospective Cohort Study of First Births in Pennsylvania. Matern Child Health J. 2022;26:2526–2535. doi:10.1007/s10995-022-03525-z.
152. Cabré S, Ratsika A, Rea K, Stanton C, Cryan JF. Animal models for assessing impact of C-section delivery on biological systems. Neurosci Biobehav Rev. 2022;135:104555. doi:10.1016/j.neubiorev.2022.104555.
153. Martinez KA, Devlin JC, Lacher CR, Yin Y, Cai Y, Wang J, Dominguez-Bello MG. Increased weight gain by C-section: functional significance of the primordial microbiome. Sci Adv. 2017;3:eaao1874. doi:10.1126/sciadv.aao1874.
154. Wong WSW, Sabu P, Deopujari V, Levy S, Shah AA, Clemency N, Provenzano M, Saadoon R, Munagala A, Baker R, et al. Prenatal and Peripartum Exposure to Antibiotics and Cesarean Section Delivery Are Associated with Differences in Diversity and Composition of the Infant Meconium Microbiome. Microorganisms. 2020;8:179. doi:10.3390/microorganisms8020179.
155. Mueller NT, Dominguez-Bello MG, Appel LJ, Hourigan SK. 'Vaginal seeding' after a caesarean section provides benefits to newborn children: FOR: does exposing caesarean-delivered newborns to the vaginal microbiome affect their chronic disease risk? The critical need for trials of 'vaginal seeding' during caesarean section. Bjog. 2020;127:301–311. doi:10.1111/1471-0528.15979.
156. Dominguez-Bello MG, De Jesus-Laboy KM, Shen N, Cox LM, Amir A, Gonzalez A, Bokulich NA, Song SJ, Hoashi M, Rivera-Vinas JI, et al. Partial restoration of the microbiota of cesarean-born infants via vaginal microbial transfer. Nat Med. 2016;22:250–253. doi:10.1038/nm.4039.

157. Cunnington AJ, Sim K, Deierl A, Kroll JS, Brannigan E, Darby J. "Vaginal seeding" of infants born by caesarean section. BMJ. 2016;352:i227. doi:10.1136/bmj.i227.
158. Kielbratowska B, Kazmierczak M, Michalek J, Preis K. Temperament and the Mother–Infant Dyad: associations with Breastfeeding and Formula Feeding with a Bottle. Infant Mental Health J. 2015;36:243–250. doi:10.1002/imhj.21508.
159. Taut C, Kelly A, Zgaga L. The Association Between Infant Temperament and Breastfeeding Duration: a Cross-Sectional Study. Breastfeed Med. 2016;11:111–118. doi:10.1089/bfm.2015.0184.
160. WHO, 2003. Global strategy for infant and young child feeding [WWW Document]. [accessed 2024 Mar 12]. https://www.who.int/publications-detail-redirect/9241562218.
161. Mosca F, Giannì ML. Human milk: composition and health benefits. Pediatr Med Chir. 2017:39. doi:10.4081/pmc.2017.155.
162. Savage JH, Lee-Sarwar KA, Sordillo JE, Lange NE, Zhou Y, O'Connor GT, Sandel M, Bacharier LB, Zeiger R, Sodergren E, et al. Diet during Pregnancy and Infancy and the Infant Intestinal Microbiome. J Pediatr. 2018;203:47–54.e4. doi:10.1016/j.jpeds.2018.07.066.
163. Davis EC, Castagna VP, Sela DA, Hillard MA, Lindberg S, Mantis NJ, Seppo AE, Järvinen KM. Gut Microbiome and Breast-feeding: implications for Early Immune Regulation. J Allergy Clin Immunol. 2022;150:523–534. doi:10.1016/j.jaci.2022.07.014.
164. Shenhav L, Fehr K, Reyna ME, Petersen C, Dai DLY, Dai R, Breton V, Rossi L, Smieja M, Simons E, et al. Microbial colonization programs are structured by breastfeeding and guide healthy respiratory development. Cell. 2024;187:5431–5452.e20. doi:10.1016/j.cell.2024.07.022.
165. Milani C, Duranti S, Bottacini F, Casey E, Turroni F, Mahony J, Belzer C, Delgado Palacio S, Arboleya Montes S, Mancabelli L, et al. The First Microbial Colonizers of the Human Gut: composition. Activities, and Health Implications of the Infant Gut Microbiota Microbiol Mol Biol Rev. 2017:81. doi:10.1128/mmbr.00036-17.
166. Al-Farsi YM, Al-Sharbati MM, Waly MI, Al-Farsi OA, Al-Shafaee MA, Al-Khaduri MM, Trivedi MS, Deth RC. Effect of suboptimal breast-feeding on occurrence of autism: a case–control study. Nutrition. 2012;28(7–8):e27–32. doi:10.1016/j.nut.2012.01.007.
167. Bar S, Milanaik R, Adesman A. Long-term neurodevelopmental benefits of breastfeeding. Curr Opin Pediatr. 2016;28(4):559–566. doi:10.1097/MOP.0000000000000389.
168. Sørensen HJ, Mortensen EL, Reinisch JM, Mednick SA. Breastfeeding and risk of schizophrenia in the Copenhagen Perinatal Cohort. Acta Psychiatr Scand. 2005;112:26–29. doi:10.1111/j.1600-0447.2005.00548.x.
169. Tseng P-T, Chen Y-W, Stubbs B, Carvalho AF, Whiteley P, Tang C-H, Yang W-C, Chen T-Y, Li D-J, Chu C-S, et al. Maternal breastfeeding and autism spectrum disorder in children: a systematic review and meta-analysis. Nutr Neurosci. 2019a;22:354–362. doi:10.1080/1028415X.2017.1388598.
170. Tseng P-T, Yen C-F, Chen Y-W, Stubbs B, Carvalho AF, Whiteley P, Chu C-S, Li D-J, Chen T-Y, Yang W-C, et al. Maternal breastfeeding and attention-deficit/hyperactivity disorder in children: a meta-analysis. Eur Child Adolesc Psychiatry. 2019b;28:19–30. doi:10.1007/s00787-018-1182-4.
171. Brahm P, Valdés V. Beneficios de la lactancia materna y riesgos de no amamantar. Revista Chilena de Pediatría. 2017;88(1):07–14. doi:10.4067/S0370-41062017000100001.
172. Mortensen EL, Michaelsen KF, Sanders SA, Reinisch JM. The Association Between Duration of Breastfeeding and Adult Intelligence. JAMA. 2002;287:2365–2371. doi:10.1001/jama.287.18.2365.
173. Ottolini KM, Andescavage N, Keller S, Limperopoulos C. Nutrition and the developing brain: the road to optimizing early neurodevelopment: a systematic review. Pediatr Res. 2020;87:194–201. doi:10.1038/s41390-019-0508-3.
174. Jones HJ, Stergiakouli E, Tansey KE, Hubbard L, Heron J, Cannon M, Holmans P, Lewis G, Linden DEJ, Jones PB, et al. Phenotypic Manifestation of Genetic Risk for Schizophrenia During Adolescence in the General Population. JAMA Psychiatry. 2016;73:221–228. doi:10.1001/jamapsychiatry.2015.3058.
175. Palladino VS, McNeill R, Reif A, Kittel-Schneider S. Genetic risk factors and gene–environment interactions in adult and childhood attention-deficit/hyperactivity disorder. Psychiatr Genet. 2019;29:63. doi:10.1097/YPG.0000000000000220.
176. Wei H, Zhu Y, Wang T, Zhang X, Zhang K, Zhang Z. Genetic risk factors for autism-spectrum disorders: a systematic review based on systematic reviews and meta-analysis. J Neural Transm. 2021;128:717–734. doi:10.1007/s00702-021-02360-w.
177. Ghafouri-Fard S, Pourtavakoli A, Hussen BM, Taheri M, Ayatollahi SA. A Review on the Role of Genetic Mutations in the Autism Spectrum Disorder. Mol Neurobiol. 2023;60:5256–5272. doi:10.1007/s12035-023-03405-9.
178. Demontis D, Walters GB, Athanasiadis G, Walters R, Therrien K, Nielsen TT, Farajzadeh L, Voloudakis G, Bendl J, Zeng B, et al. Genome-wide analyses of attention deficit hyperactivity disorder identify 27 risk loci, refine the genetic architecture, and implicate several cognitive domains. Nat Genet. 2023;55:198–208. doi:10.1038/s41588-022-01285-8.
179. Freitag CM, Rohde LA, Lempp T, Romanos M. Phenotypic and measurement influences on heritability estimates in childhood ADHD. Eur Child Adolesc Psychiatry. 2010;19:311–323. doi:10.1007/s00787-010-0097-5.

180. Hilker R, Helenius D, Fagerlund B, Skytthe A, Christensen K, Werge TM, Nordentoft M, Glenthøj B. Heritability of Schizophrenia and Schizophrenia Spectrum Based on the Nationwide Danish Twin Register. Biol Psychiatry. 2018;83:492–498. doi:10.1016/j.biopsych.2017.08.017.
181. Alamoudi MU, Hosie S, Shindler AE, Wood JL, Franks AE, Hill-Yardin EL. Comparing the Gut Microbiome in Autism and Preclinical Models: a Systematic Review. Front Cell Infect Microbiol. 2022;12:905841. doi:10.3389/fcimb.2022.905841.
182. Saurman V, Margolis KG, Luna RA. Autism Spectrum Disorder as a Brain□Gut□Microbiome Axis Disorder. Dig Dis Sci. 2020;65:818–828. doi:10.1007/s10620-020-06133-5.
183. Sivamaruthi BS, Suganthy N, Kesika P, Chaiyasut C. The Role of Microbiome, Dietary Supplements, and Probiotics in Autism Spectrum Disorder. Int J Environ Res Public Health. 2020;17:2647. doi:10.3390/ijerph17082647.
184. Sauer AK, Bockmann J, Steinestel K, Boeckers TM, Grabrucker AM. Altered Intestinal Morphology and Microbiota Composition in the Autism Spectrum Disorders Associated SHANK3 Mouse Model. Int J Mol Sci. 2019;20:2134. doi:10.3390/ijms20092134.
185. Yap CX, Henders AK, Alvares GA, Wood DLA, Krause L, Tyson GW, Restuadi R, Wallace L, McLaren T, Hansell NK, et al. Autism-related dietary preferences mediate autism-gut microbiome associations. Cell. 2021;184:5916–5931.e17. doi:10.1016/j.cell.2021.10.015.
186. Kang D-W, Adams JB, Coleman DM, Pollard EL, Maldonado J, McDonough-Means S, Caporaso JG, Krajmalnik-Brown R. Long-term benefit of Microbiota Transfer Therapy on autism symptoms and gut microbiota. Sci Rep. 2019;9:5821. doi:10.1038/s41598-019-42183-0.
187. Kang D-W, Adams JB, Vargason T, Santiago M, Hahn J, Krajmalnik-Brown R. Distinct Fecal and Plasma Metabolites in Children with Autism Spectrum Disorders and Their Modulation after Microbiota Transfer Therapy. mSphere. 2020;5:e00314–20. doi:10.1128/mSphere.00314-20.
188. Cickovski T, Mathee K, Aguirre G, Tatke G, Hermida A, Narasimhan G, Stollstorff M, Šiler BT. Attention Deficit Hyperactivity Disorder (ADHD) and the gut microbiome: an ecological perspective. PLOS ONE. 2023a;18(8):e0273890. doi:10.1371/journal.pone.0273890.
189. Pinto S, Correia-de-Sá T, Sampaio-Maia B, Vasconcelos C, Moreira P, Ferreira-Gomes J. Eating Patterns and Dietary Interventions in ADHD: a Narrative Review. Nutrients. 2022;14:4332. doi:10.3390/nu14204332.
190. Stiernborg M, Debelius J, Yang LL, Skott E, Millischer V, Giacobini M, Melas PA, Boulund F, Lavebratt C. Bacterial gut microbiome differences in adults with ADHD and in children with ADHD on psychostimulant medication. Brain Behav Immun. 2023;110:310–321. doi:10.1016/j.bbi.2023.03.012.
191. Wang Q, Yang Q, Liu X. The microbiota–gut–brain axis and neurodevelopmental disorders. Protein Cell. 2023;14:762–775. doi:10.1093/procel/pwad026.
192. Gkougka D, Mitropoulos K, Tzanakaki G, Panagouli E, Psaltopoulou T, Thomaidis L, Tsolia M, Sergentanis TN, Tsitsika A. Gut microbiome and attention deficit/hyperactivity disorder: a systematic review. Pediatr Res. 2022a;92:1507–1519. doi:10.1038/s41390-022-02027-6.
193. Sukmajaya AC, Lusida MI, Soetjipto N, Setiawati Y. Systematic review of gut microbiota and attention-deficit hyperactivity disorder (ADHD). Ann Gen Psychiatry. 2021a;20:12. doi:10.1186/s12991-021-00330-w.
194. Kalenik A, Kardaś K, Rahnama A, Sirojć K, Wolańczyk T. Gut microbiota and probiotic therapy in ADHD: a review of current knowledge. Prog Neuro Psychopharmacol Biol Psychiatry. 2021;110:110277. doi:10.1016/j.pnpbp.2021.110277.
195. Hooi SL, Dwiyanto J, Rasiti H, Toh KY, Wong RKM, Lee JWJ. A case report of improvement on ADHD symptoms after fecal microbiota transplantation with gut microbiome profiling pre- and post-procedure. Curr Med Res Opin. 2022;38:1977–1982. doi:10.1080/03007995.2022.2129232.
196. McGuinness AJ, Davis JA, Dawson SL, Loughman A, Collier F, O'Hely M, Simpson CA, Green J, Marx W, Hair C, et al. A systematic review of gut microbiota composition in observational studies of major depressive disorder, bipolar disorder and schizophrenia. Mol Psychiatry. 2022;27:1920–1935. doi:10.1038/s41380-022-01456-3.
197. Murray N, Al Khalaf S, Bastiaanssen TFS, Kaulmann D, Lonergan E, Cryan JF, Clarke G, Khashan AS, O'Connor K. Compositional and Functional Alterations in Intestinal Microbiota in Patients with Psychosis or Schizophrenia: a Systematic Review and Meta-analysis. Schizophrenia Bulletin. 2023;49:1239–1255. doi:10.1093/schbul/sbad049.
198. Nikolova VL, Smith MRB, Hall LJ, Cleare AJ, Stone JM, Young AH. Perturbations in Gut Microbiota Composition in Psychiatric Disorders: a Review and Meta-analysis. JAMA Psychiatry. 2021;78:1343–1354. doi:10.1001/jamapsychiatry.2021.2573.
199. Li S, Zhuo M, Huang X, Huang Y, Zhou J, Xiong D, Li J, Liu Y, Pan Z, Li H, et al. Altered gut microbiota associated with symptom severity in schizophrenia. PeerJ. 2020;8:e9574. doi:10.7717/peerj.9574.
200. Nguyen TT, Kosciolek T, Maldonado Y, Daly RE, Martin AS, McDonald D, Knight R, Jeste DV. Differences in Gut Microbiome Composition Between Persons with Chronic Schizophrenia and Healthy Comparison Subjects. Schizophr Res. 2019;204:23–29. doi:10.1016/j.schres.2018.09.014.
201. Zhu F, Ju Y, Wang W, Wang Q, Guo R, Ma Q, Sun Q, Fan Y, Xie Y, Yang Z, et al. Metagenome-wide

association of gut microbiome features for schizophrenia. Nat Commun. 2020b;11:1612. doi:10.1038/s41467-020-15457-9.
202. Dickerson FB, Stallings C, Origoni A, Katsafanas E, Savage CLG, Schweinfurth LAB, Goga J, Khushalani S, Yolken RH. Effect of Probiotic Supplementation on Schizophrenia Symptoms and Association With Gastrointestinal Functioning: a Randomized, Placebo-Controlled Trial. Prim Care Companion CNS Disord. 2014;16:CC.13m01579. doi:10.4088/PCC.13m01579.
203. Tomasik J, Yolken RH, Bahn S, Dickerson FB. Immunomodulatory Effects of Probiotic Supplementation in Schizophrenia Patients: a Randomized, Placebo-Controlled Trial. Biomark Insights. 2015;10:47–54. doi:10.4137/BMI.S22007.
204. Marder SR, Umbricht D. Negative symptoms in schizophrenia: newly emerging measurements, pathways, and treatments. Schizophr Res. 2023;258:71–77. doi:10.1016/j.schres.2023.07.010.
205. Nocera A, Nasrallah HA. The Association of the Gut Microbiota with Clinical Features in Schizophrenia. Behav Sci (Basel). 2022;12:89. doi:10.3390/bs12040089.
206. Schwarz E, Maukonen J, Hyytiäinen T, Kieseppä T, Orešič M, Sabunciyan S, Mantere O, Saarela M, Yolken R, Suvisaari J. Analysis of microbiota in first episode psychosis identifies preliminary associations with symptom severity and treatment response. Schizophr Res. 2018;192:398–403. doi:10.1016/j.schres.2017.04.017.
207. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. Hallmarks of aging: an expanding universe. Cell. 2023;186:243–278. doi:10.1016/j.cell.2022.11.001.
208. Ghosh TS, Shanahan F, O'Toole PW. The gut microbiome as a modulator of healthy ageing. Nat Rev Gastroenterol Hepatol. 2022a;19:565–584. doi:10.1038/s41575-022-00605-x.
209. Biagi E, Franceschi C, Rampelli S, Severgnini M, Ostan R, Turroni S, Consolandi C, Quercia S, Scurti M, Monti D, et al. Gut Microbiota and Extreme Longevity. Curr Biol. 2016;26(11):1480–1485. doi:10.1016/j.cub.2016.04.016.
210. Wu L, Zeng T, Zinellu A, Rubino S, Kelvin DJ, Carru C. A Cross-Sectional Study of Compositional and Functional Profiles of Gut Microbiota in Sardinian Centenarians. mSystems. 2019;4:e00325–19. doi:10.1128/mSystems.00325-19.
211. Ghosh TS, Shanahan F, O'Toole PW. Toward an improved definition of a healthy microbiome for healthy aging. Nat Aging. 2022b;2:1054–1069. doi:10.1038/s43587-022-00306-9.
212. Phillips S, Watt R, Atkinson T, Rajan S, Hayhoe A, Savva GM, Hornberger M, Burton BJL, Saada J, Cambell-Kelly M, et al. A protocol paper for the MOTION Study—A longitudinal study in a cohort aged 60 years and older to obtain mechanistic knowledge of the role of the gut microbiome during normal healthy ageing in order to develop strategies that will improve lifelong health and wellbeing. PLOS ONE. 2022;17:e0276118. doi:10.1371/journal.pone.0276118.
213. Pang S, Chen X, Lu Z, Meng L, Huang Y, Yu X, Huang L, Ye P, Chen X, Liang J, et al. Longevity of centenarians is reflected by the gut microbiome with youth-associated signatures. Nat Aging. 2023;3:436–449. doi:10.1038/s43587-023-00389-y.
214. Jazwinski SM, Kim S. Examination of the Dimensions of Biological Age. Front Genet. 2019;10:263. doi:10.3389/fgene.2019.00263.
215. Maltoni R, Ravaioli S, Bronte G, Mazza M, Cerchione C, Massa I, Balzi W, Cortesi M, Zanoni M, Bravaccini S. Chronological age or biological age: what drives the choice of adjuvant treatment in elderly breast cancer patients?. Transl Oncol. 2021;15:101300. doi:10.1016/j.tranon.2021.101300.
216. Rutledge J, Oh H, Wyss-Coray T. Measuring biological age using omics data. Nat Rev Genet. 2022;23:715–727. doi:10.1038/s41576-022-00511-7.
217. Xue Q-L. The Frailty Syndrome: definition and Natural History. Clin Geriatr Med. 2011;27:1–15. doi:10.1016/j.cger.2010.08.009.
218. Larson PJ, Zhou W, Santiago A, Driscoll S, Fleming E, Voigt AY, Chun OK, Grady JJ, Kuchel GA, Robison JT, et al. Associations of the skin, oral and gut microbiome with aging, frailty and infection risk reservoirs in older adults. Nat Aging. 2022;2:941–955. doi:10.1038/s43587-022-00287-9.
219. Haran JP, Bucci V, Dutta P, Ward D, McCormick B. The nursing home elder microbiome stability and associations with age, frailty, nutrition and physical location. J Med Microbiol. 2018;67:40–51. doi:10.1099/jmm.0.000640.
220. Boehme M, Guzzetta KE, Bastiaanssen TFS, van de Wouw M, Moloney GM, Gual-Grau A, Spichak S, Olavarría-Ramírez L, Fitzgerald P, Morillas E, et al. Microbiota from young mice counteracts selective age-associated behavioral deficits. Nat Aging. 2021;1(8):666–676. doi:10.1038/s43587-021-00093-9.
221. Mossad O, Nent E, Woltemate S, Folschweiller S, Buescher JM, Schnepf D, Erny D, Staeheli P, Bartos M, Szalay A, et al. Microbiota-dependent increase in δ-valerobetaine alters neuronal function and is responsible for age-related cognitive decline. Nat Aging. 2021;1:1127–1136. doi:10.1038/s43587-021-00141-4.
222. Parker A, Romano S, Ansorge R, Aboelnour A, Le Gall G, Savva GM, Pontifex MG, Telatin A, Baker D, Jones E, et al. Fecal microbiota transfer between young and aged mice reverses hallmarks of the aging gut, eye, and brain. Microbiome. 2022;10:68. doi:10.1186/s40168-022-01243-w.
223. Armstrong RA. Risk factors for Alzheimer's disease. Folia Neuropathol. 2019;57(2):87–105. doi:10.5114/fn.2019.85929.
224. Hou Y, Dan X, Babbar M, Wei Y, Hasselbalch SG, Croteau DL, Bohr VA. Ageing as a risk factor for

neurodegenerative disease. Nat Rev Neurol. 2019;15:565–581. doi:10.1038/s41582-019-0244-7.
225. Reeve A, Simcox E, Turnbull D. Ageing and Parkinson's disease: why is advancing age the biggest risk factor?. Ageing Res Rev. 2014;14:19. doi:10.1016/j.arr.2014.01.004.
226. Jagaraj CJ, Shadfar S, Kashani SA, Saravanabavan S, Farzana F, Atkin JD. Molecular hallmarks of ageing in amyotrophic lateral sclerosis. Cell Mol Life Sci. 2024;81:111. doi:10.1007/s00018-024-05164-9.
227. Niccoli T, Partridge L, Isaacs AM. Ageing as a risk factor for ALS/FTD. Human Mol Genet. 2017;26:R105–R113. doi:10.1093/hmg/ddx247.
228. Graves JS, Krysko KM, Hua LH, Absinta M, Franklin RJM, Segal BM. Ageing and multiple sclerosis. Lancet Neurol. 2023;22:66–77. doi:10.1016/S1474-4422(22)00184-3.
229. Machiela E, Southwell AL. Biological Aging and the Cellular Pathogenesis of Huntington's Disease. J Huntingtons Dis. 2020;9:115–128. doi:10.3233/JHD-200395.
230. Alexopoulos P, Grimmer T, Perneczky R, Domes G, Kurz A. Progression to Dementia in Clinical Subtypes of Mild Cognitive Impairment. Dementia Geriatric Cognit Disord. 2006;22(1):27–34. doi:10.1159/000093101.
231. Petersen RC, Doody R, Kurz A, Mohs RC, Morris JC, Rabins PV, Ritchie K, Rossor M, Thal L, Winblad B. Current Concepts in Mild Cognitive Impairment. Arch Neurol. 2001;58:1985–1992. doi:10.1001/archneur.58.12.1985.
232. Łuc M, Misiak B, Pawłowski M, Stańczykiewicz B, Zabłocka A, Szcześniak D, Pałęga A, Rymaszewska J. Gut microbiota in dementia. Critical review of novel findings and their potential application. Prog Neuro Psychopharmacol Biol Psychiatry. 2021;104:110039. doi:10.1016/j.pnpbp.2020.110039.
233. Loughman A, Adler CJ, Macpherson H. Unlocking Modifiable Risk Factors for Alzheimer's Disease: does the Oral Microbiome Hold Some of the Keys?. JAD. 2023;92:1111–1129. doi:10.3233/JAD-220760.
234. Connell E, Le Gall G, Pontifex MG, Sami S, Cryan JF, Clarke G, Müller M, Vauzour D. Microbial-derived metabolites as a risk factor of age-related cognitive decline and dementia. Mol Neurodegener. 2022;17(1):43. doi:10.1186/s13024-022-00548-6.
235. Den H, Dong X, Chen M, Zou Z. Efficacy of probiotics on cognition, and biomarkers of inflammation and oxidative stress in adults with Alzheimer's disease or mild cognitive impairment — a meta-analysis of randomized controlled trials. Aging (Albany NY). 2020;12:4010–4039. doi:10.18632/aging.102810.
236. Krüger JF, Hillesheim E, Pereira ACSN, Camargo CQ, Rabito EI. Probiotics for dementia: a systematic review and meta-analysis of randomized controlled trials. Nutr Rev. 2021;79:160–170. doi:10.1093/nutrit/nuaa037.
237. Ferreiro AL, Choi J, Ryou J, Newcomer EP, Thompson R, Bollinger RM, Hall-Moore C, Ndao IM, Sax L, Benzinger TLS, et al. Gut microbiome composition may be an indicator of preclinical Alzheimer's disease. Sci, trans med. 2023;15:eabo2984. doi:10.1126/scitranslmed.abo2984.
238. Wu L, Han Y, Zheng Z, Peng G, Liu P, Yue S, Zhu S, Chen J, Lv H, Shao L, et al. Altered Gut Microbial Metabolites in Amnestic Mild Cognitive Impairment and Alzheimer's Disease: signals in Host–Microbe Interplay. Nutrients. 2021;13:228. doi:10.3390/nu13010228.
239. Ling Z, Zhu M, Yan X, Cheng Y, Shao L, Liu X, Jiang R, Wu S. Structural and Functional Dysbiosis of Fecal Microbiota in Chinese Patients With Alzheimer's Disease. Front Cell Develop Biol. 2021;8 doi:https://doi.org/10.3389/fcell.2020.634069.
240. Fernández-Veledo S, Vendrell J. Gut microbiota-derived succinate: friend or foe in human metabolic diseases?. Rev Endocr Metab Disord. 2019;20:439–447. doi:10.1007/s11154-019-09513-z.
241. Tannahill G, Curtis A, Adamik J, Palsson-McDermott E, McGettrick A, Goel G, Frezza C, Bernard N, Kelly B, Foley N, et al. Succinate is a danger signal that induces IL-1β via HIF-1α. Nature. 2013;496:238–242. doi:10.1038/nature11986.
242. Kowalski K, Mulak A. Brain-Gut-Microbiota Axis in Alzheimer's Disease. J Neurogastroenterol Motil. 2019;25:48–60. doi:10.5056/jnm18087.
243. Killingsworth J, Sawmiller D, Shytle RD. Propionate and Alzheimer's Disease. Front Aging Neurosci. 2021;12:580001. doi:10.3389/fnagi.2020.580001.
244. Rei D, Saha S, Haddad M, Rubio AH, Perlaza BL, Berard M, Ungeheuer M-N, Sokol H, Lledo P-M. Age-associated gut microbiota impairs hippocampus-dependent memory in a vagus-dependent manner. JCI Insight. 2022:7. doi:10.1172/jci.insight.147700.
245. Kim M-S, Kim Y, Choi H, Kim W, Park S, Lee D, Kim DK, Kim HJ, Choi H, Hyun D-W, et al. Transfer of a healthy microbiota reduces amyloid and tau pathology in an Alzheimer's disease animal model. Gut. 2020;69:283–294. doi:10.1136/gutjnl-2018-317431.
246. Grabrucker S, Marizzoni M, Silajdžić E, Lopizzo N, Mombelli E, Nicolas S, Dohm-Hansen S, Scassellati C, Moretti DV, Rosa M, et al. Microbiota from Alzheimer's patients induce deficits in cognition and hippocampal neurogenesis. Brain. 2023:awad303. doi:10.1093/brain/awad303.
247. Park S-H, Lee JH, Shin J, Kim J-S, Cha B, Lee S, Kwon KS, Shin YW, Choi SH. Cognitive function improvement after fecal microbiota transplantation in Alzheimer's dementia patient: a case report. Curr Med Res Opin. 2021;37:1739–1744. doi:10.1080/03007995.2021.1957807.
248. Kim J-S, Park H, Lee J-H, Shin J, Cha B, Kwon KS, Shin YW, Kim Y, Kim Y, Bae JS, et al. Effect of altered gene expression in lipid metabolism on cognitive

improvement in patients with Alzheimer's dementia following fecal microbiota transplantation: a preliminary study. Ther Adv Neurol Disord. 2024;17:17562864231218181. doi:10.1177/17562864231218181.
249. Ağagündüz D, Kocaadam-Bozkurt B, Bozkurt O, Sharma H, Esposito R, Özoğul F, Capasso R. Microbiota alteration and modulation in Alzheimer's disease by gerobiotics: the gut-health axis for a good mind. Biomed Pharmacother. 2022;153:113430. doi:10.1016/j.biopha.2022.113430.
250. Zhao Z, Ning J, Bao X, Shang M, Ma J, Li G, Zhang D. Fecal microbiota transplantation protects rotenone-induced Parkinson's disease mice via suppressing inflammation mediated by the lipopolysaccharide-TLR4 signaling pathway through the microbiota-gut-brain axis. Microbiome. 2021;9:226. doi:10.1186/s40168-021-01107-9.
251. Sun M-F, Zhu Y-L, Zhou Z-L, Jia X-B, Xu Y-D, Yang Q, Cui C, Shen Y-Q. Neuroprotective effects of fecal microbiota transplantation on MPTP-induced Parkinson's disease mice: gut microbiota, glial reaction and TLR4/TNF-α signaling pathway. Brain Behav Immun. 2018;70:48–60. doi:10.1016/j.bbi.2018.02.005.
252. Xie Z, Zhang M, Luo Y, Jin D, Guo X, Yang W, Zheng J, Zhang H, Zhang L, Deng C, et al. Healthy Human Fecal Microbiota Transplantation into Mice Attenuates MPTP-Induced Neurotoxicity via AMPK/SOD2 Pathway. Aging Dis. 2023;14:2193–2214. doi:10.14336/AD.2023.0309.
253. Zhang Y-G, Wu S, Yi J, Xia Y, Jin D, Zhou J, Sun J. Target Intestinal Microbiota to Alleviate Disease Progression in Amyotrophic Lateral Sclerosis. Clin Ther. 2017;39:322–336. doi:10.1016/j.clinthera.2016.12.014.
254. Li K, Wei S, Hu L, Yin X, Mai Y, Jiang C, Peng X, Cao X, Huang Z, Zhou H, et al. Protection of Fecal Microbiota Transplantation in a Mouse Model of Multiple Sclerosis. Mediators Inflam. 2020;2020:e2058272. doi:10.1155/2020/2058272.
255. Kong G, Ellul S, Narayana VK, Kanojia K, Ha HTT, Li S, Renoir T, Cao K-AL, Hannan AJ. An integrated metagenomics and metabolomics approach implicates the microbiota-gut-brain axis in the pathogenesis of Huntington's disease. Neurobiol Dis. 2021;148:105199. doi:10.1016/j.nbd.2020.105199.
256. Gubert C, Kong G, Costello C, Adams CD, Masson BA, Qin W, Choo J, Narayana VK, Rogers G, Renoir T, et al. Dietary fibre confers therapeutic effects in a preclinical model of Huntington's disease. Brain Behav Immun. 2024;116:404–418. doi:10.1016/j.bbi.2023.12.023.
257. Gubert C, Choo JM, Love CJ, Kodikara S, Masson BA, Liew JJM, Wang Y, Kong G, Narayana VK, Renoir T, et al. Faecal microbiota transplant ameliorates gut dysbiosis and cognitive deficits in Huntington's disease mice. Brain Comm. 2022;4:fcac205. doi:10.1093/braincomms/fcac205.
258. DuPont HL, Suescun J, Jiang Z-D, Brown EL, Essigmann HT, Alexander AS, DuPont AW, Iqbal T, Utay NS, Newmark M, et al. Fecal microbiota transplantation in Parkinson's disease—A randomized repeat-dose, placebo-controlled clinical pilot study. Front Neurol. 2023;14:1104759. doi:10.3389/fneur.2023.1104759.
259. Xue L-J, Yang X-Z, Tong Q, Shen P, Ma S-J, Wu S-N, Zheng J-L, Wang H-G. Fecal microbiota transplantation therapy for Parkinson's disease. Medicine (Baltimore). 2020;99:e22035. doi:10.1097/MD.0000000000022035.
260. Fratiglioni L. Epidemiology of Alzheimer's disease and current possibilities for prevention. Acta neurol Scand. 1996;94:33–40. doi:10.1111/j.1600-0404.1996.tb05870.x.
261. Tilley L, Morgan K, Kalsheker N. Genetic risk factors in Alzheimer's disease. Mol Pathol. 1998;51:293–304.
262. Billingsley KJ, Bandres-Ciga S, Saez-Atienzar S, Singleton AB. Genetic risk factors in Parkinson's disease. Cell Tissue Res. 2018;373(1):9–20. doi:10.1007/s00441-018-2817-y.
263. Iwaki H, Blauwendraat C, Leonard HL, Liu G, Maple-Grødem J, Corvol J-C, Pihlstrøm L, van Nimwegen M, Hutten SJ, Nguyen K-DH, et al. Genetic risk of Parkinson disease and progression. In: Neurology Genetics. Vol. 5. Van Hilten, J.J. Scherzer, C.R. Singleton, A.B. Nalls, M.A: B.P; 2019. e348. van de Warrenburg. doi:10.1212/NXG.0000000000000348.
264. Ingre C, Roos PM, Piehl F, Kamel F, Fang F. Risk factors for amyotrophic lateral sclerosis. Clin Epidemiol. 2015;7:181–193. doi:10.2147/CLEP.S37505.
265. Waubant E, Lucas R, Mowry E, Graves J, Olsson T, Alfredsson L, Langer-Gould A. Environmental and genetic risk factors for MS: an integrated review. Ann. Clin. Transl. Neurol. 2019;6:1905–1922. doi:10.1002/acn3.50862.
266. Olsson T, Barcellos LF, Alfredsson L. Interactions between genetic, lifestyle and environmental risk factors for multiple sclerosis. Nat Rev Neurol. 2017;13:25–36. doi:10.1038/nrneurol.2016.187.
267. Myers RH. Huntington's disease genetics. Neurotherapeutics. 2004;1:255–262. doi:10.1602/neurorx.1.2.255.
268. Lee J-M, Wheeler VC, Chao MJ, Vonsattel JPG, Pinto RM, Lucente D, Abu-Elneel K, Ramos EM, Mysore JS, Gillis T, et al., Cell, 2015 Identification of Genetic Factors that Modify Clinical Onset of Huntington's Disease. ;162:516–526. doi:10.1016/j.cell.2015.07.003.
269. Mo C, Hannan AJ, Renoir T. Environmental factors as modulators of neurodegeneration: insights from gene–environment interactions in Huntington's disease. Neurosci Biobehav Rev. 2015;52:178–192. doi:10.1016/j.neubiorev.2015.03.003.
270. Fasano A, Visanji NP, Liu LW, Lang AE, Pfeiffer RF. Gastrointestinal dysfunction in Parkinson's disease. Lancet Neurol. 2015;14:625–639.

271. Hawkes CH, Del Tredici K, Braak H. Parkinson's disease: the dual hit theory revisited. Ann NY Acad Sci. 2009;1170:615–622.
272. O'Neill C. Gut microbes metabolize Parkinson's disease drug. Science. 2019;364:1030–1031. doi:10.1126/science.aax8937.
273. La Reau AJ, Strom NB, Filvaroff E, Mavrommatis K, Ward TL, Knights D. Shallow shotgun sequencing reduces technical variation in microbiome analysis. Sci Rep. 2023;13:7668. doi:10.1038/s41598-023-33489-1.
274. Liu P, Hu S, He Z, Feng C, Dong G, An S, Liu R, Xu F, Chen Y, Ying X. Towards Strain-Level Complexity: sequencing Depth Required for Comprehensive Single-Nucleotide Polymorphism Analysis of the Human Gut Microbiome. Front Microbiol. 2022;13 doi:https://doi.org/10.3389/fmicb.2022.828254.
275. Wallen ZD, Demirkan A, Twa G, Cohen G, Dean MN, Standaert DG, Sampson TR, Payami H. Metagenomics of Parkinson's disease implicates the gut microbiome in multiple disease mechanisms. Nat Commun. 2022;13:6958. doi:10.1038/s41467-022-34667-x.
276. Bedarf JR, Hildebrand F, Coelho LP, Sunagawa S, Bahram M, Goeser F, Bork P, Wüllner U. Functional implications of microbial and viral gut metagenome changes in early stage L-DOPA-naïve Parkinson's disease patients. Genome Med. 2017;9(1):39. doi:10.1186/s13073-017-0428-y.
277. Lin A, Zheng W, He Y, Tang W, Wei X, He R, Huang W, Su Y, Huang Y, Zhou H, et al. Gut microbiota in patients with Parkinson's disease in southern China. Parkinsonism Related Disord. 2018;53:82–88. doi:10.1016/j.parkreldis.2018.05.007.
278. Chen S-J, Chen -C-C, Liao H-Y, Lin Y-T, Wu Y-W, Liou J-M, Wu M-S, Kuo C-H, Lin C-H. Association of Fecal and Plasma Levels of Short-Chain Fatty Acids With Gut Microbiota and Clinical Severity in Patients With Parkinson Disease. Neurology. 2022;98(8):e848–e858. doi:10.1212/WNL.0000000000013225.
279. Unger MM, Spiegel J, Dillmann K-U, Grundmann D, Philippeit H, Bürmann J, Faßbender K, Schwiertz A, Schäfer K-H. Short chain fatty acids and gut microbiota differ between patients with Parkinson's disease and age-matched controls. Parkinsonism Related Disord. 2016;32:66–72. doi:10.1016/j.parkreldis.2016.08.019.
280. Yang X, Ai P, He X, Mo C, Zhang Y, Xu S, Lai Y, Qian Y, Xiao Q. Parkinson's Disease Is Associated with Impaired Gut–Blood Barrier for Short-Chain Fatty Acids. Mov Disord. 2022;37:1634–1643. doi:10.1002/mds.29063.
281. Dalile B, Van Oudenhove L, Vervliet B, Verbeke K. The role of short-chain fatty acids in microbiota–gut–brain communication. Nat Rev Gastroenterol Hepatol. 2019;16:461–478. doi:10.1038/s41575-019-0157-3.
282. Nuzum ND, Szymlek-Gay EA, Loke S, Dawson SL, Teo W-P, Hendy AM, Loughman A, Macpherson H. Differences in the gut microbiome across typical ageing and in Parkinson's disease. Neuropharmacology. 2023:109566. doi:10.1016/j.neuropharm.2023.109566.
283. Aho VTE, Pereira PAB, Voutilainen S, Paulin L, Pekkonen E, Auvinen P, Scheperjans F. Gut microbiota in Parkinson's disease: temporal stability and relations to disease progression. EBioMedicine. 2019;44:691–707. doi:10.1016/j.ebiom.2019.05.064.
284. Cilia R, Piatti M, Cereda E, Bolliri C, Caronni S, Ferri V, Cassani E, Bonvegna S, Ferrarese C, Zecchinelli AL, et al. Does Gut Microbiota Influence the Course of Parkinson's Disease? A 3-Year Prospective Exploratory Study in de novo Patients. Journal of Parkinson's Disease. 2021;11(1):159–170. doi:10.3233/JPD-202297.
285. Lubomski M, Xu X, Holmes AJ, Muller S, Yang JYH, Davis RL, Sue CM. The Gut Microbiome in Parkinson's Disease: a Longitudinal Study of the Impacts on Disease Progression and the Use of Device-Assisted Therapies. Front Aging Neurosci. 2022;14 doi:10.3389/fnagi.2022.875261.
286. Minato T, Maeda T, Fujisawa Y, Tsuji H, Nomoto K, Ohno K, Hirayama M. Progression of Parkinson's disease is associated with gut dysbiosis: two-year follow-up study. PLOS ONE. 2017:12. doi:10.1371/journal.pone.0187307.
287. Huang B, Chau SWH, Liu Y, Chan JWY, Wang J, Ma SL, Zhang J, Chan PKS, Yeoh YK, Chen Z, et al. Gut microbiome dysbiosis across early Parkinson's disease, REM sleep behavior disorder and their first-degree relatives. Nat Commun. 2023;14:2501. doi:10.1038/s41467-023-38248-4.
288. Borghammer P, Van Den Berge N, van Laar T. Brain-First versus Gut-First Parkinson's Disease: a Hypothesis. Journal of Parkinson's Disease. 2019;9(s2):S281–S295. doi:10.3233/JPD-191721.
289. Horsager J, Andersen KB, Knudsen K, Skjærbæk C, Fedorova TD, Okkels N, Schaeffer E, Bonkat SK, Geday J, Otto M, et al. Brain-first versus body-first Parkinson's disease: a multimodal imaging case-control study. Brain. 2020. doi:10.1093/brain/awaa238.
290. Nishiwaki H, Hamaguchi T, Ito M, Ishida T, Maeda T, Kashihara K, Tsuboi Y, Ueyama J, Shimamura T, Mori H, et al. Short-Chain Fatty Acid-Producing Gut Microbiota Is Decreased in Parkinson's Disease but Not in Rapid-Eye-Movement Sleep Behavior Disorder. mSystems. 2020a;5:e00797–20. doi:10.1128/mSystems.00797-20.
291. Nishiwaki H, Ito M, Hamaguchi T, Maeda T, Kashihara K, Tsuboi Y, Ueyama J, Yoshida T, Hanada H, Takeuchi I, et al. Short chain fatty acids-producing and mucin-degrading intestinal bacteria predict the progression of early Parkinson's disease. npj Parkinsons Dis. 2022;8:1–12. doi:10.1038/s41531-022-00328-5.
292. Chen SG, Stribinskis V, Rane MJ, Demuth DR, Gozal E, Roberts AM, Jagadapillai R, Liu R, Choe K, Shivakumar B, et al. Exposure to the Functional Bacterial Amyloid Protein Curli Enhances Alpha-Synuclein Aggregation

in Aged Fischer 344 Rats and Caenorhabditis elegans. Sci Rep. 2016;6(1):34477. doi:10.1038/srep34477.
293. Huynh VA, Takala TM, Murros KE, Diwedi B, Saris PEJ. Desulfovibrio bacteria enhance alpha-synuclein aggregation in a Caenorhabditis elegans model of Parkinson's disease. Front Cell Infect Microbiol. 2023;13:1181315. doi:10.3389/fcimb.2023.1181315.
294. O'Donovan SM, Crowley EK, Brown JR-M, O'Sullivan O, O'Leary OF, Timmons S, Nolan YM, Clarke DJ, Hyland NP, Joyce SA, et al. Nigral overexpression of α-synuclein in a rat Parkinson's disease model indicates alterations in the enteric nervous system and the gut microbiome. Neurogastroenterol Motil. 2020;32: e13726. doi:10.1111/nmo.13726.
295. Shojaie A, Rota S, Al Khleifat A, Ray Chaudhuri K, Al-Chalabi A. Non-motor symptoms in amyotrophic lateral sclerosis: lessons from Parkinson's disease. Amyotrophic Lateral Scler Fronttemporal Degener. 2023;0:1–10. doi:10.1080/21678421.2023.2220748.
296. Longstreth WT, Meschke JS, Davidson SK, Smoot LM, Smoot JC, Koepsell TD. Hypothesis: a motor neuron toxin produced by a clostridial species residing in gut causes ALS. Med Hypotheses. 2005;64:1153–1156. doi:10.1016/j.mehy.2004.07.041.
297. Blacher E, Bashiardes S, Shapiro H, Rothschild D, Mor U, Dori-Bachash M, Kleimeyer C, Moresi C, Harnik Y, Zur M, et al. Potential roles of gut microbiome and metabolites in modulating ALS in mice. Nature. 2019;572(7770):474–480. doi:10.1038/s41586-019-1443-5.
298. Di Gioia D, Bozzi Cionci N, Baffoni L, Amoruso A, Pane M, Mogna L, Gaggìa F, Lucenti MA, Bersano E, Cantello R, et al. A prospective longitudinal study on the microbiota composition in amyotrophic lateral sclerosis. BMC Med. 2020;18:153. doi:10.1186/s12916-020-01607-9.
299. Holmstrøm K, Collins MD, Møller T, Falsen E, Lawson PA. Subdoligranulum variabile gen. nov. sp. nov. from human feces. Anaerobe. 2004;10:197–203. doi:10.1016/j.anaerobe.2004.01.004.
300. Niccolai E, Di Pilato V, Nannini G, Baldi S, Russo E, Zucchi E, Martinelli I, Menicatti M, Bartolucci G, Mandrioli J, et al. The Gut Microbiota-Immunity Axis in ALS: a Role in Deciphering Disease Heterogeneity?. Biomedicines. 2021;9:753. doi:10.3390/biomedicines9070753.
301. Brenner D, Hiergeist A, Adis C, Mayer B, Gessner A, Ludolph AC, Weishaupt JH. The fecal microbiome of ALS patients. Neurobiol Aging. 2018;61:132–137. doi:10.1016/j.neurobiolaging.2017.09.023.
302. Fang X, Wang X, Yang S, Meng F, Wang X, Wei H, Chen T. Evaluation of the Microbial Diversity in Amyotrophic Lateral Sclerosis Using High-Throughput Sequencing. Front Microbiol. 2016;7 doi:10.3389/fmicb.2016.01479.
303. Rowin J, Xia Y, Jung B, Sun J. Gut inflammation and dysbiosis in human motor neuron disease. Physiol Rep. 2017;5:e13443. doi:10.14814/phy2.13443.
304. Henkel JS, Beers DR, Wen S, Bowser R, Appel SH. Decreased mRNA expression of tight junction proteins in lumbar spinal cords of patients with ALS. Neurology. 2009;72:1614–1616. doi:10.1212/WNL.0b013e3181a41228.
305. Mirza A, Forbes JD, Zhu F, Bernstein CN, Van Domselaar G, Graham M, Waubant E, Tremlett H. The multiple sclerosis gut microbiota: a systematic review. Mult Scler Relat Disord. 2020;37:101427. doi:10.1016/j.msard.2019.101427.
306. Correale J, Hohlfeld R, Baranzini SE. The role of the gut microbiota in multiple sclerosis. Nat Rev Neurol. 2022;18:544–558. doi:10.1038/s41582-022-00697-8.
307. Borody T, Leis S, Campbell J, Torres M, Nowak A. Fecal Microbiota Transplantation (FMT) in Multiple Sclerosis (MS): 942. Off J Am College Gastroenterol. 2011;106:S352.
308. Makkawi S, Camara-Lemarroy C, Metz L. Fecal microbiota transplantation associated with 10 years of stability in a patient with SPMS. Neurol Neuroimmunol Neuroinflamm. 2018;5:e459. doi:10.1212/NXI.0000000000000459.
309. Al KF, Craven LJ, Gibbons S, Parvathy SN, Wing AC, Graf C, Parham KA, Kerfoot SM, Wilcox H, Burton JP, et al. Fecal microbiota transplantation is safe and tolerable in patients with multiple sclerosis: a pilot randomized controlled trial. Multiple Sclerosis J. 2022;8(2):20552173221086662. doi:10.1177/20552173221086662.
310. Cantoni C, Lin Q, Dorsett Y, Ghezzi L, Liu Z, Pan Y, Chen K, Han Y, Li Z, Xiao H, et al. Alterations of host-gut microbiome interactions in multiple sclerosis. EBioMedicine. 2022;76:103798. doi:10.1016/j.ebiom.2021.103798.
311. Devolder L, Pauwels A, Van Remoortel A, Falony G, Vieira-Silva S, Nagels G, De Keyser J, Raes J, D'Hooghe MB. Gut microbiome composition is associated with long-term disability worsening in multiple sclerosis. Gut Microbes. 2023;15:2180316. doi:10.1080/19490976.2023.2180316.
312. Montgomery TL, Wang Q, Mirza A, Dwyer D, Wu Q, Dowling CA, Martens JW, Yang J, Krementsov DN, Mao-Draayer Y. Identification of commensal gut microbiota signatures as predictors of clinical severity and disease progression in multiple sclerosis; 2023. medRxiv 2023.06.26.23291875. doi:10.1101/2023.06.26.23291875.
313. Rosas HD, Doros G, Bhasin S, Thomas B, Gevorkian S, Malarick K, Matson W, Hersch SM. A systems-level "misunderstanding": the plasma metabolome in Huntington's disease. Ann Clin Transl Neurol. 2015;2:756–768. doi:10.1002/acn3.214.

314. Kong G, Cao K-AL, Judd LM, Li S, Renoir T, Hannan AJ. Microbiome profiling reveals gut dysbiosis in a transgenic mouse model of Huntington's disease. Neurobiol Dis. 2020;135:104268. doi:10.1016/j.nbd.2018.09.001.
315. Anand S, Kaur H, Mande SS. Comparative In silico Analysis of Butyrate Production Pathways in Gut Commensals and Pathogens. Front Microbiol. 2016;7. doi:10.3389/fmicb.2016.01945.
316. Wasser CI, Mercieca E-C, Kong G, Hannan AJ, McKeown SJ, Glikmann-Johnston Y, Stout JC. Gut dysbiosis in Huntington's disease: associations among gut microbiota, cognitive performance and clinical outcomes. Brain Comm. 2020;2:fcaa110. doi:10.1093/braincomms/fcaa110.
317. Du G, Dong W, Yang Q, Yu X, Ma J, Gu W, Huang Y. Altered Gut Microbiota Related to Inflammatory Responses in Patients With Huntington's Disease. Front Immunol. 2021;11:603594. doi:10.3389/fimmu.2020.603594.
318. Ekwudo MN, Gubert C, Hannan AJ. The microbiota–gut–brain axis in Huntington's disease: pathogenic mechanisms and therapeutic targets. FEBS J. 2024. doi:10.1111/febs.17102.
319. Stockdale SR, Draper LA, O'Donovan SM, Barton W, O'Sullivan O, Volpicelli-Daley LA, Sullivan AM, O'Neill C, Hill C. Alpha-synuclein alters the faecal viromes of rats in a gut-initiated model of Parkinson's disease. Commun Biol. 2021;4:1–12. doi:10.1038/s42003-021-02666-1.
320. Williams ZAP, Lang L, Nicolas S, Clarke G, Cryan J, Vauzour D, Nolan YM. Do microbes play a role in Alzheimer's disease?. Microb Biotechnol. 2024;17:e14462. doi:10.1111/1751-7915.14462.
321. Zhang S, Zeng B, Chen Y, Yang M, Kong F, Wei L, Li F, Zhao J, Li Y. Gut microbiota in healthy and unhealthy long-living people. Gene. 2021;779:145510. doi:10.1016/j.gene.2021.145510.
322. Zhang T, Brander G, Mantel Ä, Kuja-Halkola R, Stephansson O, Chang Z, Larsson H, Mataix-Cols D, Fernández de la Cruz L. Assessment of Cesarean Delivery and Neurodevelopmental and Psychiatric Disorders in the Children of a Population-Based Swedish Birth Cohort. JAMA Network Open. 2021;4:e210837. doi:10.1001/jamanetworkopen.2021.0837.
323. Cimino S, Cerniglia L Impact of maternal-newborn separation in Italian women with suspected COVID-19 infection on psychopathological symptoms and quality of interactions during breastfeeding. Psychopathology. 2023;56(5):335–341. 10.1159/000528410.