Gut Microbiota: A Perspective for Psychiatrists

Kieran Rea^a Timothy G. Dinan^{a, b} John F. Cryan^{a, c}

^aAPC Microbiome Ireland, University College Cork, Cork, Ireland; ^bDepartment of Psychiatry and Neurobehavioural Science, University College Cork, Cork, Ireland; ^cDepartment of Anatomy and Neuroscience, University College Cork, Cork, Ireland

Keywords

Microbes · Gut · Mental health

Abstract

There is mounting evidence that the trillions of microbes that inhabit our gut are a substantial contributing factor to mental health and, equally, to the progression of neuropsychiatric disorders. The extraordinary complexity of the gut ecosystem, and how it interacts with the intestinal epithelium to manifest physiological changes in the brain to influence mood and behaviour, has been the subject of intense scientific scrutiny over the last 2 decades. To further complicate matters, we each harbour a unique microbiota community that is subject to change by a number of factors including diet, exercise, stress, health status, genetics, medication, and age, amongst others. The microbiota-gut-brain axis is a dynamic matrix of tissues and organs including the gastrointestinal (GI) microbiota, immune cells, gut tissue, glands, the autonomic nervous system (ANS), and the brain that communicate in a complex multidirectional manner through a number of anatomically and physiologically distinct systems. Long-term perturbations to this homeostatic environment may contribute to the progression of a number of disorders by altering physiological processes including hypothalamic-pituitary-adrenal axis activation, neurotransmitter

systems, immune function, and the inflammatory response. While an appropriate, co-ordinated physiological response, such as an immune or stress response, is necessary for survival, a dysfunctional response can be detrimental to the host, contributing to the development of a number of central nervous system disorders.

© 2019 S. Karger AG, Basel

Introduction

Over thousands of years, the gut ecosystem has evolved to contain a diverse population of microorganisms including yeasts, archaea, parasites, helminth, viruses, and protozoa, but the bacterial population is currently the most well characterized [1–5]. Current estimates suggest that our microbial cells outnumber human cells by a ratio of 1.3:1 [6], while at a genetic level more than 99% of the genes in our bodies are microbial, comprising over 10 million microbial genes [7–9]. The contribution of these microbes to human health and disease cannot be understated, as they play key roles in such functions as metabolism, satiety, and immune regulation and more recently they have been shown to play a role in mood and behaviour. In certain circumstances, our microbes possess the genetic machinery for the metabolism of important di-

etary compounds that would otherwise be non-digestible to the host [8, 10]. Interestingly, while our inherited genome (that we inherited from our parents) is relatively stable throughout our lifetimes, the microbial genome is immensely diverse [11, 12], dynamic [13], and responsive to external inputs including diet, exercise, stress, health status, medication, and age amongst others.

The composition of the bacteria that inhabit our GI tract throughout our adult life is established early in our first few years of life and at this early-life stage they are particularly sensitive to manipulation by a number of environmental factors including mode of delivery (vaginal or C-section), whether we are breastfed or bottle-fed, diet, medication (in particular antibiotic medication), and exposure to viral or bacterial infections and stress [14]. It is worth noting that this critical seeding of our core microbiota occurs in parallel with the growth, maturation, and sprouting of neurons in the young brain [14, 15], and a similar profile is evident in old age where a decline in microbiota complexity and diversity occurs in parallel with a decrease in neuronal complexity [16], and this may contribute to the onset or progression of mood disorders including anxiety, depressive-like symptoms, mania, anhedonia, irritability, or suicidal ideology.

Concomitantly with the birth process or C-section delivery (or perhaps even prenatally [17, 18], our GI tract comes into contact with, and maintains constant communication with, the microbes that inhabit our gut, either through direct physical contact or the release of secreted compounds. Current hypotheses suggest that these microbiota-host interactions at the level of the gut release cytokines, chemokines, neurotransmitters, neuropeptides, endocrine messengers, and microbial by-products that can infiltrate the blood and lymphatic systems, or influence neural messages carried by the vagal and spinal afferent neurons to constantly communicate with the brain and update as to the health status and possibly to regulate mood and behaviour. However, it seems unlikely that any single class of compounds is wholly responsible for mediation of microbiota-gut-brain interactions and these will be discussed below.

Communication along the Microbiota-Gut-Brain Axis

The gut microbiota is part of a complex network termed the microbiota-gut-brain axis along with the sympathetic and parasympathetic divisions of the ANS, the enteric nervous system (ENS), and the neuroendocrine and neuroimmune components of the central nervous

system (CNS) [19–21]. However, the mechanisms underpinning this bi-directional communication in the microbiota-gut-brain axis remain, as yet, unresolved [22]. Modes of communication likely involve neural mechanisms, immune response, neurotransmitter and neuropeptide release, and/or microbial by-products (Fig. 1), which will be discussed below. The consequential dynamic molecules that are released may infiltrate multiple anatomical environments to communicate within their local environment to regulate complex co-ordinated responses in multiple physiological systems. Molecular candidates thought to be involved in these processes include neurotransmitters, neuropeptides, short-chain fatty acids (SCFA), bile moieties, endocrine hormones, and immunomodulators amongst others.

Gut-Microbe Interactions

Despite the many beneficial functions of the gut microbes, the host has to maintain a physical barrier between the host and the microbiota to prevent infection. The epithelial lining of the gut contains predominantly secretory cells, enterocytes, chemosensory cells, and gut-associated lymphoid tissue [23] and is interspersed with tight junction proteins [194] that maintain protection against a “leaky” gut. A key function of the single-celled epithelial layer of our gut is to limit the direct contact of intestinal microbiota with the visceral tissue, which it does by secreting a protective viscous mucus layer from goblet cells lining the gut wall that increases in thickness as we travel from proximal to distal gut. This luminal-mucosal interface is where the majority of host-microbe interactions occur, and the exchange of molecules back and forth through the mucous layer and epithelium serve to facilitate communication between the microbiota, the gut, the immune, endocrine and ANS and the brain [24].

Endothelial secretory cells play a key role in gut function. Bacterial colonization in the gut is kept in check by antimicrobial peptides, expressed by secretory Paneth cells within small intestinal crypts. Further host protection from the gut microbes is guaranteed by protective mucus layer from secretory goblet cells, defensins, and anti-bacterial lectins shielding the epithelium from direct contact with the microbes as well as the innate (immune cells in the lamina propria of the ENS) and adaptive (immunoglobulin A-secreting plasma B cells) roles of the mucosal immune system. The secretory cells also control of the release of substances from enteroendocrine cells (EEC) such as ghrelin, somatostatin, cholecystokinin, peptide YY and serotonin amongst others [25].

Fig. 1. a Bi-directional communication along the microbiota-gut-brain axis, involving neural, endocrine, immune, and other pathways. Proposed mechanisms of communication include: (1) microbial by-products including SCFA, lipopolysaccharides, and peptidoglycans amongst others; (2) neuropeptides and neurotransmitters; (3) cytokines and immune cell activation; (4) neural networks including ENS and vagal nerve activation; (5) and

HPA axis modulation. **b** Dysregulation of the microbiota-gut-brain axis has been linked to a number of psychiatric disorders including stress, anxiety, depression, autism, schizophrenia, Parkinson's disease, and Alzheimer's disease. NT, neuropeptide; NT, neurotransmitters; DC, dendritic cell; CRF, corticosterone-releasing factor; ACTH, adrenocorticotrophic hormone.

There are many microbes that are only found in the mucosal layer, while some are restricted to the lumen of the gut. Host-microorganism interactions from luminal and mucosal microbiota occur, and are highly dependent on immune response. The enterocytes express innate immune receptors and once activated prompt the release of cytokines and chemokines, chemosensory cells play a key role in defence against helminths [26], while the gut-associated lymphoid tissue utilize lymphocytes to mount a more specific immune response if warranted.

GI bacteria possess a polysaccharide coating that protects them from degradation but also serves to identify the multitude of bacteria to the host. In turn, the host immune system can monitor and regulate commensal bacteria and identify invading pathogens. Specific pattern recognition receptors, of which members of the Toll-like receptor family are the most studied, line the epithelium and are capable of recognizing specific molecular signatures unique to bacteria [27] and other microorganisms

(pathogen-associated molecular patterns, or PAMP) [28, 29]. Once activated, these receptors can initiate a cascade of events including the recruitment of inflammatory mediators, cytokine production and chemokine-mediated recruitment of acute inflammatory cells [30, 31]. This in turn helps to regulate the microbial population and diversity in the gut. Whilst this innate immune response is necessary to maintain gut homeostasis, a dysfunctional or prolonged response (e.g., to invading pathogens or as a consequence of a chronically high inflammatory tone) can weaken the integrity of the intestinal barrier and the mucosal layer, facilitating an increase in plasma lipopolysaccharide levels as a consequence of increased bacteria infiltration across the gut [32].

Microbe-Mediated Synthesis and Release of Bioactive Compounds

Along with activation of the innate immune system, commensal bacteria can also directly or indirectly

(through endothelial cells) synthesize and release other bioactive compounds including neurotransmitters, neuromodulators, bacteriocins, bile acids, choline, and SCFA that are integral to host health.

Two ongoing large collaborative efforts from the Human Microbiome Project (HMP) [33, 34] and MetaHIT [8, 35] have been instrumental in surveying and describing the gut microbiota at a population level. Current combined HMP and MetaHIT data estimate that there are at least 2,776 species that have been isolated from human faecal matter. These have been classified into 11 different phyla, with Proteobacteria, Firmicutes, Actinobacteria, and Bacteroidetes comprising over 90% of the microbiome [35–37], while Fusobacteria and Verrucomicrobia phyla are present in low abundance [2]. Furthermore, the indigenous microbiota diversity changes as one moves from proximal to distal gut [38, 39]. These commensal bacteria are capable of synthesizing and releasing many neurotransmitters and neuromodulators themselves or evoke EEC into the synthesis and release of neuropeptides. For example, *Lactobacillus* and *Bifidobacterium* species can produce γ -aminobutyric acid (GABA); *Escheridia*, *Bacillus*, and *Saccharomyces* spp. can produce norepinephrine; *Bacillus* can produce dopamine; *Lactobacillus* can produce acetylcholine; and *Candida*, *Streptococcus*, *Escheridia*, and *Enterococcus* spp. can produce serotonin [40–42]. Serotonin is a key neurotransmitter implicated in many psychiatric disorders. It has been estimated that approximately 95% of the serotonin in the body is compartmentalized in the gut, predominantly in enterochromaffin cells of the mucosa and in nerve terminals of the ENS. Studies using germ-free mice have implicated GI microbiota in impacting on tryptophan metabolism, the precursor of serotonin [43]. These neurotransmitters are critical in centrally mediated events and bodily functions, and it is possible that these microbially synthesized neurotransmitters can cross the mucosal layer of the intestines, and possibly mediate effects locally in the gut, or enter the bloodstream and impact on physiological events in the brain.

GI bacteria can also secrete bioactive chemicals such as bacteriocins, bile acids, choline, and SCFA that are integral to host health and disease. Bacteriocins are antibiotic proteins that inhibit the growth of other bacteria in the vicinity, while bile acids are best known for facilitating the absorption of dietary lipids and lipid-soluble vitamins from the gut lumen but can play a role in regulating the number of bacteria in the small intestine [44–46]. Strikingly, all major gut-associated bacterial divisions, including *Lactobacillus*, *Bifidobacterium*, and *Bacteroidetes*

taxa, were shown to express bile salt hydrolase [47] enzymes which allow de-conjugation of bile acid from taurine and glycine [48, 49], which are beneficial for the survival of the microbiota secreting them. SCFA including butyric acid, acetic acid, propionic acid, and lactic acid are dietary by-products derived from the fermentation of polysaccharides [50] by colonic bacteria. These fatty acids can enter the blood and activate free fatty acid receptors (FFAR) in the gut, periphery, and brain and have been reported to have neuroactive properties [51–53] and to play a role in anorexia nervosa [54], Parkinson's disease [55], and animal models of chronic stress [56] and Alzheimer's disease [57]. Conversely, increased SCFA levels have been associated with obesity [58–60], autism spectrum disorder [61], and chronic psychosocial stress in children [62]. Such data implicates the potential of SCFA to be a key player in microbiota-gut-brain axis communication. Interestingly, SCFA can pass directly through the epithelium via transporters [63–66] or indirectly by diffusing through epithelia as un-ionized SCFA [67]; and they have receptors along the epithelium [68–72], in the periphery [47], along the vagus nerve and ANS ganglia [73–76], and even in the brain epithelium [52, 77].

As well as influencing local immune responses at the epithelium, and synthesizing and releasing neurotransmitters and SCFA, the gut microbes can influence the release of neuropeptides and hormones from EEC of the intestines. Gut peptides such as ghrelin, gastrin, orexin, galanin, cholecystokinin, leptin, and neuropeptide Y are thought to influence peripheral neural communication and can also act centrally to influence behaviour. Though EEC represent only 1% of the epithelial cells in the GI tract, they are critically important for the maintenance of gut homeostasis due to the pleiotropic effects of secreted signalling molecules. To date, about 10 different types of EEC have been characterized, all of which are sensory cells that coordinate changes in the gut-nutrient luminal content with metabolic and behavioural responses, such as the regulation of insulin secretion or food intake [78]. The receptors to many of these peptides are expressed locally in the gut enteric neurons and vagal afferents and in the CNS, including the brainstem and the hypothalamus [79, 80].

From Gut to Brain

Under homeostatic conditions there exists a healthy, resting inflammatory tone where the microbiota stimulate cytokines and chemokine release, which in turn regulates local levels of bacteria in the gut. As well as influencing local immune responses at the epithelium, microbio-

ta can synthesize and release neurotransmitters and SCFA and influence the release of neuropeptides and hormones from EEC of the intestines. Gut peptides such as ghrelin, gastrin, orexin, galanin, cholecystokinin, leptin, and neuropeptide Y are thought to influence peripheral neural communication and can also act centrally to influence behaviour. Current hypotheses suggest that these circulating cytokines, chemokines, endocrine messengers, and microbial by-products can infiltrate the blood and lymphatic systems or influence neural messages carried by the vagal and spinal afferent neurons to impact on centrally mediated events, including regulation of hypothalamic-pituitary-adrenal (HPA) axis activity and neuroinflammation.

At the interface between the microbiota and the host lies a network of neurons, i.e., the ENS, positioned to respond, either directly or indirectly, to the microbiota and/or microbiota-derived metabolites. The ENS can be crudely sub-divided into the submucosal plexus and the myenteric plexus and is largely responsible for the coordination of gut functions such as motility and control of fluid movement [81]. The ANS is best described as a neural relay network within the central and peripheral nervous systems that governs bodily functions that are normally controlled without a conscious effort (autonomously), such as breathing, regulation of our heartbeat, and digestion. The vagus nerve is the principal neuron within the ANS and innervates the stomach, the small intestine, and the proximal portion of the colon and works in concert with the neural network lining the layers of the gut, the ENS, and the CNS to regulate GI functions such as gut motility and permeability, epithelial fluid maintenance, luminal osmolarity, secretion of bile, carbohydrate levels, mechanical distortion of the mucosa, and bicarbonate and mucus production as well as the mucosal immune response and intestinal-fluid handling [82] and as such can regulate microbiota diversity and complexity in the gut. Conversely, local events at the level of the gut including homeostasis, and potential harmful pathogens, are communicated via the ENS and the vagal nerve from the gut to the brain.

From an anatomical perspective, the current consensus is that there are 2 neuroanatomical routes for neural signalling from the intestine to the brain. Primary afferents from the gut can be crudely separated into “non-painful” (satiety, distension, motility, and other homeostatic functions) mediated predominantly by vagal/pelvic innervation and “painful” sensory stimuli conveyed by splanchnic innervation [83]. Physiological information relating to the status of the gut including distension, mo-

tility, inflammation, pain, pH change, cellular damage, or temperature generates neural signals [84] via the release of bioactive chemicals including globulin, protein kinases, serine proteases, arachidonic acid, prostaglandins, cytokines, histamine, nerve growth factor, substance P, calcitonin gene-related peptide, serotonin, acetylcholine, and ATP as well as changes in pH [83, 85, 86]. These neural messages are transmitted systematically from the enteric neural networks that include the glial cells, myenteric and submucosal ganglia in the gut [87, 88] to the prevertebral (dorsal root) ganglia that regulate peripheral visceral reflex responses [89–91], to the spinal cord and the nucleus tractus solitarius of the brainstem [92, 93], and to higher centres in the brain.

Regulation of the Stress Response by GI Microbiota

Stress, and the subsequent activation of the HPA axis is an evolutionary conserved response to a perturbation of the homeostatic environment of the organism in response to an actual or perceived threat [94]. Upon activation, the HPA axis ultimately results in the release of behaviour-altering endocrine messengers including glucocorticoids and mineralocorticoids, as well as catecholamines, and it returns to normal homeostatic levels once the threat or perceived threat has subsided. While a deficient or blunted HPA axis is commonly observed in the clinic in a wide range of autoimmune and inflammatory diseases; chronic or prolonged exposure to stress can contribute to the precipitation, maintenance, and/or neuroprogression of a number of neuroinflammatory and stress-related disorders including anxiety [95, 96], depression [95, 96], bipolar disorder [97], post-traumatic stress disorder [98], Alzheimer’s disease [99, 100], and schizophrenia [101] amongst others. A clear reciprocal role for microbiota and stress response has been extensively reviewed over the last 2 decades. It has been shown in animal models that different types of psychological stress including maternal separation, chronic social defeat, restraint conditions, crowding, heat stress, and acoustic stress can alter the composition of GI microbiota [102–109]. Conversely, a number of experimental models and conditions have been employed, demonstrating a key role of the microbiota in regulating the stress response including prebiotic and probiotic [110–116] intervention, antibiotic administration [116–119], faecal transplantation, and the use of germ-free and specific pathogen-free animals [43, 106, 120–123]. Limited studies have also seen beneficial health reports in humans [124, 125]. Whilst the precise mechanisms behind GI microbiota and stress interactions remain to be elucidated,

the intricate relationship of the immune system with both the stress response and GI microbiota makes it a likely biochemical candidate.

Regulation of the Neuroinflammatory Response by GI Microbiota

The passage of immune cells, cytokines, chemokines, endocrine (stress) messengers, and microbial by-products to the brain is tightly regulated by the blood-brain barrier, which is under vigilant surveillance by resident macrophage and microglia [126]. Recently, it was determined that a diverse GI microbiota is necessary the maintenance and maturation of microglia in a healthy functional state [47], and that this is regulated by microbially synthesized SCFA. Furthermore, the integrity of the blood-brain barrier is under the control of GI microbiota [127]. As with the epithelium of the gut, pattern recognition receptors on these immune cells recognize microbial by-products or respond to local changes in the environment as a consequence of cytokines, chemokines, and endocrine messengers ultimately resulting in neuroinflammation and recruit peripheral monocytes to traffic from the spleen to the brain [128]. Interestingly, the spleen expresses a high density of FFAR2 [48], the receptor for SCFA, on trafficking lymphocytes [129]. Moreover, these FFAR2-expressing trafficking lymphocytes also express glucocorticoid receptors and respond to stress stimuli. Collectively, the evidence suggests a multimodal influence of microbiota on physiological events at the level of the CNS via intervention in the recruitment of local immune regulators from the periphery to the brain.

Thus far we have introduced some of the proposed mechanisms of action of microbially driven control of the gut-brain axis and their role in regulating the stress and immune response. We will further address the role of the GI microbiota in psychiatric disorders including Alzheimer's disease, Parkinson's disease, autism, schizophrenia, anxiety disorders, depression, and bipolar disorder.

The Role of GI Microbiota in Psychiatric Disorders

Given the myriad of complex physiological processes by which gut microbes could affect brain function, it is hardly surprising that current research has linked dysregulation of gut microbes to various psychiatric illnesses. Given the limitations of current treatments for psychiatric disorders, and the rates of remission, treatment resistance, and side effects, the possibility of low-risk but po-

tentially microbiome-modulating treatments represents an attractive target for manipulation.

However, despite the persuasive data from animal studies for a role of the microbiota in regulating mood, cognition, stress, and social behaviour, amongst others, relevant studies from human cohorts are extremely limited. From a translational perspective, double blind, crossover studies in naive subjects are required in order to advance the field in this research area.

Clinical evidence for a role of the microbiome in psychiatric disorders is provided by an alteration in microbiota diversity and complexity when compared to healthy controls as determined for autism [130–135], schizophrenia [136–139], and attention deficit disorder [140], as well as mood disorders including bipolar disorder [141–144], anxiety, stress, and depression [138, 145–150] and neuroinflammatory linked disorders including Alzheimer's [151–153] and Parkinson's [55, 154–157] diseases. However, whether these alterations are causal in these disorders or the changes are an appropriate response to a shift in their host environment remains, as yet, unresolved.

Positive Modulation of the Microbiota to Improve Psychiatric Disorders

Positive modulation of the gut microbiota with the introduction of living microorganisms has been proven to be successful in ameliorating some negative effects in various disorders, yet many technical and pragmatic difficulties exist with this therapeutic approach. Once ingested, the fate of these organisms is somewhat uncertain as it traverses a number of somewhat hostile physiological environments including the oropharyngeal area with its high density of digestive enzymes, the acidic pH levels of the stomach, the alkali bile environment of the small intestine, the immune-active interface with the epithelium, and the competitive local environment within the lumen of the gut.

Recently, studies investigating the effects of a probiotic mix of *Bifidobacterium anamalis* subsp. *Lactis*, *Streptococcus thermophiles*, *Lactobacillus bulgaricus*, and *Lactococcus lactis* subsp. *Lactis* on mood reported a positive effect in the brains of healthy female volunteers using fMRI after 4 weeks [159]. Another 4-week double-blind, placebo-controlled randomized fMRI study demonstrated that a probiotic formulation containing *L. casei*, *L. acidophilus*, *L. paracasei*, *B. lactis*, *L. salivarius*, *L. lactis*, *B. lactis*, *L. plantarum*, and *B. bifidum* altered brain activation patterns in response to emotional memory and emotional decision-making tasks, which were also accompanied by subtle shifts in the gut microbiome profile

[159]. A 3-week study in a healthy, aged population reported a positive effect of *L. casei* Shirota [160] on mood, whilst the same probiotic was effective in ameliorating anxiety in chronic fatigue patients following 2 months treatment [161] and reduced abdominal discomfort and sleep quality in healthy medical students exposed to exam stress [162, 163]. Similarly, a *L. plantarum* 299v probiotic was shown to decrease exam stress levels in healthy controls following a 2-week administration [164]. Anxiety and depressive readouts were decreased with a concomitant decrease in cortisol levels following 30 days probiotic (*L. helveticus* and *B. longum*) treatment in healthy adults [165], while a mix of *S. thermophiles*, *L. bulgaricus*, *L. lactis* subsp. *Lactis*, *L. acidophilus*, *S. thermophiles*, *L. plantarum*, *B. lactis*, and *L. reuteri* decreased anxiety behaviour in healthy controls after 3 weeks [166]. *L. gasseri* CP2305 ameliorated stress-related symptoms and sleep quality in healthy male and female volunteers after 5 weeks of treatment [167]. *B. longum* 1714, already proven to reduce anxiety and stress responses during acute stress in mice [168], similarly reduced stress and anxiety measures after 4 weeks treatment in a population of healthy adults [169], and it improved the cognitive performance [113, 169]. However, not all probiotics that show beneficial effects pre-clinically translate to a beneficial effect in human studies. A study in healthy volunteers undergoing exam stress demonstrated that *L. rhamnosus* (JB-1) had no effect on mood, anxiety, stress, or sleep [170].

Alterations in GI microbiota have also been linked to bipolar disorder, although the data is somewhat limited [141–144] and intervention studies have mostly been mostly inconclusive [136, 171]. Further evidence for a role of the microbiota in bipolar disorder is provided by a study where patients presenting with bipolar mania were nearly twice as likely as other patients to have been recently treated with systemic antibiotics [172]. Specifically, bipolar disorder has been linked to decreased Firmicutes, specifically *Faecalibacterium* [142], which also correlated with self-reporting symptom severity. In females undergoing atypical antipsychotic treatment, a decreased species diversity, in particular Lachnospiraceae, *Akkermansia*, and *Sutterella*, was observed compared to treatment-naïve controls [143]. Recent clinical probiotic studies in bipolar disorder suggest that probiotic therapy could reduce the rate of re-hospitalization of patients over a 24-week adjunctive probiotic treatment with *L. rhamnosus* strain GG and *B. animalis* subsp. *lactis* strain Bb12 who were recently discharged following hospitalization for mania [136], and a subtle effect on “cognitive reactivity to sad mood” following 3 month treatment with

L. casei, *L. acidophilus*, *L. paracasei*, *B. lactis*, *L. salivarius*, *L. lactis*, *B. lactis*, *L. plantarum*, and *B. bifidum* [171].

While a number of studies have reported an altered microbiota in individuals with autism, schizophrenia, or ADHD, there is limited or no evidence to demonstrate whether targeting the microbiota through probiotic or dietary interventions can improve their symptoms in humans. However, a small open-label clinical intervention study using faecal matter transplant of a standardized microbiota cocktail to children with ASD was efficacious in improving the GI and behavioural symptoms [173]. Although the sample size was small and the experimental design lacked a randomized double-blind structure, the study provided promising preliminary evidence to demonstrate that the microbiota may indeed be targeted as a potential treatment strategy for ASD. Similarly, clinical studies investigating the role of probiotics in ADHD are lacking, but one promising study where children aged up to 6 months were given *L. rhamnosus* GG (ATCC 53103) and followed until 13 years of age and, while 6 of 35 control students presented with ADHD, none of the 40 probiotic-treated students presented with ADHD symptoms [174].

A limited number of studies investigating the therapeutic role of probiotics in individuals suffering from mood disorders including stress, anxiety, bipolar disorder, and depression have been conducted. In a recent 8-week, randomized, double-blind, placebo-controlled clinical trial including 40 patients with a diagnosis of MDD based on DSM-IV criteria whose age ranged between 20 and 55 years, a triple-strain probiotic (*L. acidophilus*, *L. casei*, and *B. bifidum*) resulted in improvements in depression scores in addition to beneficial metabolic effects in an MDD cohort [175]. Another 8-week, randomized, double-blind, placebo-controlled clinical trial included 81 patients and examined the effect of a probiotic mix of *L. helveticus* and *B. longum* on mild to moderate MDD in a placebo-controlled parallel study [176] and reported a decrease in depression scores as well as improvements in tryptophan signalling. Interestingly, a randomized, placebo-controlled, double-blind study with 380 participants reported that daily *L. rhamnosus* HN001 during pregnancy (gestational weeks 14–16) and into the post-partum period (6 months post-partum if breastfeeding) significantly decreased post-natal anxiety and depression scores [177]. However, other studies in the literature failed to show any benefit for depression scores following the administration of probiotics in patients presenting with depression [178]. Overall, systematic reviews of probiotics as a potential adjunct therapy in

MDD are encouraging, describing that probiotics effectively improve mood in humans [179–181]. However, another recent meta-analysis called into question the significance of these findings, highlighting the confounding comparability of studies due to strain differences and disease severity [182].

In patients suffering from Alzheimer's disease or Parkinson's disease, there are as yet no published studies reporting the effects of probiotics in ameliorating the symptoms of these disorders.

Psychiatric Medication and the Microbiome

A growing body of evidence suggests that medication used in the treatment of psychiatric disorders can also affect the composition of the gut microbiota, with potential implications for behaviour, as well as the involvement of the microbiota in drug pharmacokinetics in general [183]. Benzodiazepines and antidepressants were listed as co-variables influencing microbiota in the Belgian Flemish Gut Flora Project [184]. Similarly, in another population-based study with Dutch participants, i.e., LifeLines-DEEP, deep-sequencing of the gut microbiotas revealed a relationship between the medication used in the treatment of psychiatric disorders, particularly antidepressants, and the diversity and complexity of the gut microbiota [185].

Given the comorbidity of psychiatric disorders with other ailments, polypharmacy (the concurrent use of multiple medications by a patient) has also been investigated. One study demonstrated that there was a significant negative correlation between the number of different drugs consumed and microbial diversity, although it is unknown whether the lower diversity resulted in a reduced cognitive function [186]. Specifically, the drug classes that had the strongest association with single taxa abundance were PPI, anti-depressants, and anti-psychotics. Several studies have been performed in vitro in order to determine the anti-microbial activity of non-antibiotic drugs [187–190] and again antidepressants, benzodiazepines, and antipsychotics and they have been shown to possess antimicrobial activity, with a strong potential for subsequent functional neural effects.

While the microbes in our gut are susceptible to modulation by treatments in psychiatric disorders, they in turn also play a key role in metabolizing orally administered natural products and drugs [191, 192]. This area of research investigating the role of the gut microbiota in the kinetic profile of xenobiotics is termed “pharmacomicrobiomics” and it is in its infancy. However, the reciprocal influence of medication used in the treatment of psychi-

atric disorders and gut microbiota diversity and complexity may unlock information on personalized therapeutic treatment – in terms of onset of action, dose, treatment resistance, and relapse.

The Prospects for Microbiota Manipulation as a Therapy for Psychiatric Disorders

Given the role that the microbiome plays in health maintenance, and with the evidence from preclinical studies, microbiota manipulation represents a promising tool or adjunct therapy in the treatment of a number of disorders and/or their associated symptoms. By and large, the rather limited clinical studies in this research field have been performed in healthy adults, at 1 dose and with only 1 or 2 probiotic treatment groups – and often the cohorts are quite small. However, the majority of these studies have reported positive effects. Clinicians, while considering the potentially beneficial effects of these microbiota manipulations, cannot neglect the treatment of their patients with the appropriate psychiatric medicine and, as such, the true beneficial outcomes of this research field will remain somewhat stunted until we better understand the effects of the psychiatric treatments on the microbiota and the role of the microbiota in the efficacy of the psychiatric treatment.

A profound advantage of microbiota manipulation as a treatment strategy is that the microbiota is extremely dynamic and can be positively altered quite readily by a number of factors including diet, exercise, and stress reduction. A quote from Hippocrates, “Let food be thy medicine and medicine be thy food,” remains as true today as it was over 2,000 years ago. Research into positive modulation of the microbiota through diet with a view to improving health has dramatically increased in the last 2 decades, with various dietary compounds including carbohydrates (sugars, oligosaccharides, and inulin), proteins (branched chain amino acids, SCFA, and essential amino acids), linoleic acids, bile acids, polyunsaturated fatty acids, and polyphenols being explored for health promotion [193].

Given the recent advances in sequencing technologies, and computational approaches to data mining, the integration of data from clinical datasets from taxonomic profiles of the microbiota, pharmacomicrobiomics, metabolic and immune readouts from metagenomics, proteomics, and lipidomics combined with functional imaging and behavioural readouts should allow significant leaps forward in this research field. Given the unique signature of the trillions of microbiota we each harbour in our gut, perhaps these tiny microbes may explain the

range of responders and non-responders in treatment resistance, side effect profiles, and/or tolerance. Indeed, personalized medicine based on our unique microbiome signature may represent the future of psychiatric treatment or an adjunct therapy.

Acknowledgement

This research was conducted with the financial support of Science Foundation Ireland (SFI) under grant No. SFI/12/RC/2273.

References

- Lankelma JM, Nieuwdorp M, de Vos WM, Wiersinga WJ. The gut microbiota in internal medicine: implications for health and disease. *Neth J Med*. 2015 Feb;73(2):61–8.
- Eckburg PB, Bik EM, Bernstein CN, Purdom E, Dethlefsen L, Sargent M, et al. Diversity of the human intestinal microbial flora. *Science*. 2005 Jun;308(5728):1635–8.
- Scarpellini E, Ianiro G, Attili F, Bassanelli C, De Santis A, Gasbarrini A. The human gut microbiota and virome: potential therapeutic implications. *Dig Liver Dis*. 2015 Dec;47(12):1007–12.
- Gaci N, Borrel G, Tottey W, O'Toole PW, Brugère JF. Archaea and the human gut: new beginning of an old story. *World J Gastroenterol*. 2014 Nov;20(43):16062–78.
- Williamson LL, McKenney EA, Holzkecht ZE, Belliveau C, Rawls JF, Poulton S, et al. Got worms? Perinatal exposure to helminths prevents persistent immune sensitization and cognitive dysfunction induced by early-life infection. *Brain Behav Immun*. 2016 Jan;51:14–28.
- Sender R, Fuchs S, Milo R. Are We Really Vastly Outnumbered? Revisiting the Ratio of Bacterial to Host Cells in Humans. *Cell*. 2016 Jan;164(3):337–40.
- Nicholson JK, Holmes E, Wilson ID. Gut microorganisms, mammalian metabolism and personalized health care. *Nat Rev Microbiol*. 2005 May;3(5):431–8.
- Qin J, Li R, Raes J, Arumugam M, Burgdorf KS, Manichanh C, et al.; MetaHIT Consortium. A human gut microbial gene catalogue established by metagenomic sequencing. *Nature*. 2010 Mar;464(7285):59–65.
- Gill SR, Pop M, Deboy RT, Eckburg PB, Turnbaugh PJ, Samuel BS, et al. Metagenomic analysis of the human distal gut microbiome. *Science*. 2006 Jun;312(5778):1355–9.
- Flint HJ, Scott KP, Duncan SH, Louis P, Forano E. Microbial degradation of complex carbohydrates in the gut. *Gut Microbes*. 2012 Jul-Aug;3(4):289–306.
- Mosca A, Leclerc M, Hugot JP. Gut microbiota diversity and human diseases: should we reintroduce key predators in our ecosystem? *Front Microbiol*. 2016 Mar;7:455.
- Avershina E, Slangsvold S, Simpson MR, Storø O, Johnsen R, Øien T, et al. Diversity of vaginal microbiota increases by the time of labor onset. *Sci Rep*. 2017 Dec;7(1):17558.
- Lloyd-Price J, Mahurkar A, Rahnavard G, Crabtree J, Orvis J, Hall AB, et al. Strains, functions and dynamics in the expanded Human Microbiome Project. *Nature*. 2017 Oct;550(7674):61–6.
- Borre YE, O'Keefe GW, Clarke G, Stanton C, Dinan TG, Cryan JF. Microbiota and neurodevelopmental windows: implications for brain disorders. *Trends Mol Med*. 2014 Sep;20(9):509–18.
- Borre YE, Moloney RD, Clarke G, Dinan TG, Cryan JF. The impact of microbiota on brain and behavior: mechanisms & therapeutic potential. *Adv Exp Med Biol*. 2014;817:373–403.
- Biagi E, Franceschi C, Rampelli S, Severgnini M, Ostan R, Turrioni S, et al. Gut Microbiota and Extreme Longevity. *Curr Biol*. 2016 Jun;26(11):1480–5.
- Slattery J, MacFabe DF, Frye RE. The Significance of the enteric microbiome on the development of childhood disease: a review of prebiotic and probiotic therapies in disorders of childhood. *Clin Med Insights Pediatr*. 2016 Oct;10:91–107.
- Pelzer E, Gomez-Arango LF, Barrett HL, Nitert MD. Review: maternal health and the placental microbiome. *Placenta*. 2017 Jun;54:30–7.
- Cryan JF, Dinan TG. More than a gut feeling: the microbiota regulates neurodevelopment and behavior. *Neuropsychopharmacology*. 2015 Jan;40(1):241–2.
- Mayer EA, Knight R, Mazmanian SK, Cryan JF, Tillisch K. Gut microbes and the brain: paradigm shift in neuroscience. *J Neurosci*. 2014 Nov;34(46):15490–6.
- Rhee SH, Pothoulakis C, Mayer EA. Principles and clinical implications of the brain-gut-enteric microbiota axis. *Nat Rev Gastroenterol Hepatol*. 2009 May;6(5):306–14.
- Cryan JF, O'Riordan KJ, Cowan CS, Sandhu KV, Bastiaansen TF, Boehme M, et al. The Microbiota-Gut-Brain Axis. *Physiol Rev*. 2019 Oct;99(4):1877–2013.
- Pott J, Horneff M. Innate immune signalling at the intestinal epithelium in homeostasis and disease. *EMBO Rep*. 2012 Aug;13(8):684–98.
- Fasano A, Shea-Donohue T. Mechanisms of disease: the role of intestinal barrier function in the pathogenesis of gastrointestinal autoimmune diseases. *Nat Clin Pract Gastroenterol Hepatol*. 2005 Sep;2(9):416–22.
- Cani PD, Everard A, Duparc T. Gut microbiota, enteroendocrine functions and metabolism. *Curr Opin Pharmacol*. 2013 Dec;13(6):935–40.
- Allaire JM, Crowley SM, Law HT, Chang SY, Ko HJ, Vallance BA. The intestinal epithelium: central coordinator of mucosal immunity. *Trends Immunol*. 2018 Sep;39(9):677–96.
- Royet J, Gupta D, Dziarski R. Peptidoglycan recognition proteins: modulators of the microbiome and inflammation. *Nat Rev Immunol*. 2011 Nov;11(12):837–51.
- Vaishnava S, Behrendt CL, Hooper LV. Innate immune responses to commensal bacteria in the gut epithelium. *J Pediatr Gastroenterol Nutr*. 2008 Apr;46 Suppl 1:E10–1.
- Duerkop BA, Vaishnava S, Hooper LV. Immune responses to the microbiota at the intestinal mucosal surface. *Immunity*. 2009 Sep;31(3):368–76.
- Takeda K, Akira S. Microbial recognition by Toll-like receptors. *J Dermatol Sci*. 2004 Apr;34(2):73–82.
- Vaishnava S, Behrendt CL, Ismail AS, Eckmann L, Hooper LV. Paneth cells directly sense gut commensals and maintain homeostasis at the intestinal host-microbial interface. *Proc Natl Acad Sci USA*. 2008 Dec;105(52):20858–63.
- Souza DG, Vieira AT, Soares AC, Pinho V, Nicoli JR, Vieira LQ, et al. The essential role of the intestinal microbiota in facilitating acute inflammatory responses. *J Immunol*. 2004 Sep;173(6):4137–46.
- Huttenhower C, Gevers D, Knight R, Abubucker S, Badger JH, Chinwalla AT, et al.; Human Microbiome Project Consortium. Structure, function and diversity of the healthy human microbiome. *Nature*. 2012 Jun;486(7402):207–14.
- Nelson KE, Weinstock GM, Highlander SK, Worley KC, Creasy HH, Wortman JR, et al.; Human Microbiome Jumpstart Reference Strains Consortium. A catalog of reference genomes from the human microbiome. *Science*. 2010 May;328(5981):994–9.
- Li J, Jia H, Cai X, Zhong H, Feng Q, Sunagawa S, et al.; MetaHIT Consortium; MetaHIT Consortium. An integrated catalog of reference genes in the human gut microbiome. *Nat Biotechnol*. 2014 Aug;32(8):834–41.
- Hugon P, Dufour JC, Colson P, Fournier PE, Sallah K, Raoult D. A comprehensive repertoire of prokaryotic species identified in human beings. *Lancet Infect Dis*. 2015 Oct;15(10):1211–9.
- Bilen M, Dufour JC, Lagier JC, Cadoret F, Daoud Z, Dubourg G, et al. The contribution of culturomics to the repertoire of isolated human bacterial and archaeal species. *Microbiome*. 2018 May;6(1):94.
- Lozupone CA, Stombaugh JI, Gordon JI, Jansson JK, Knight R. Diversity, stability and resilience of the human gut microbiota. *Nature*. 2012 Sep;489(7415):220–30.

- 39 Gevers D, Pop M, Schloss PD, Huttenhower C. Bioinformatics for the human microbiome project. *PLoS Comput Biol*. 2012;8(11):e1002779.
- 40 Lyte M. Microbial endocrinology in the microbiome-gut-brain axis: how bacterial production and utilization of neurochemicals influence behavior. *PLoS Pathog*. 2013; 9(11):e1003726.
- 41 Lyte M. Microbial endocrinology: host-microbiota neuroendocrine interactions influencing brain and behavior. *Gut Microbes*. 2014 May-Jun;5(3):381–9.
- 42 Rea K, Dinan TG, Cryan JF. The microbiome: a key regulator of stress and neuroinflammation. *Neurobiol Stress*. 2016 Mar;4:23–33.
- 43 Clarke G, Grenham S, Scully P, Fitzgerald P, Moloney RD, Shanahan F, et al. The microbiome-gut-brain axis during early life regulates the hippocampal serotonergic system in a sex-dependent manner. *Mol Psychiatry*. 2013 Jun; 18(6):666–73.
- 44 Begley M, Gahan CG, Hill C. The interaction between bacteria and bile. *FEMS Microbiol Rev*. 2005 Sep;29(4):625–51.
- 45 Inagaki T, Moschetta A, Lee YK, Peng L, Zhao G, Downes M, et al. Regulation of antibacterial defense in the small intestine by the nuclear bile acid receptor. *Proc Natl Acad Sci USA*. 2006 Mar;103(10):3920–5.
- 46 Joyce SA, MacSharry J, Casey PG, Kinsella M, Murphy EF, Shanahan F, et al. Regulation of host weight gain and lipid metabolism by bacterial bile acid modification in the gut. *Proc Natl Acad Sci USA*. 2014 May;111(20):7421–6.
- 47 Erny D, Hrabě de Angelis AL, Jaitin D, Wieghofer P, Staszewski O, David E, et al. Host microbiota constantly control maturation and function of microglia in the CNS. *Nat Neurosci*. 2015 Jul;18(7):965–77.
- 48 Begley M, Hill C, Gahan CG. Bile salt hydrolase activity in probiotics. *Appl Environ Microbiol*. 2006 Mar;72(3):1729–38.
- 49 Jones BV, Begley M, Hill C, Gahan CG, Marchesi JR. Functional and comparative metagenomic analysis of bile salt hydrolase activity in the human gut microbiome. *Proc Natl Acad Sci USA*. 2008 Sep;105(36):13580–5.
- 50 Cummings JH, Macfarlane GT. Role of intestinal bacteria in nutrient metabolism. *J Parenter Enteral Nutr*. 1997 Nov-Dec;21(6):357–65.
- 51 Koh A, De Vadder F, Kovatcheva-Datchary P, Bäckhed F. From Dietary Fiber to Host Physiology: Short-Chain Fatty Acids as Key Bacterial Metabolites. *Cell*. 2016 Jun;165(6):1332–45.
- 52 Hoyles L, Fernandez-Real JM, Federici M, Serino M, Abbott J, Charpentier J, et al. Molecular phenomics and metagenomics of hepatic steatosis in non-diabetic obese women. *Nat Med*. 2018;24(7):1070–80.
- 53 Russell WR, Hoyles L, Flint HJ, Dumas ME. Colonic bacterial metabolites and human health. *Curr Opin Microbiol*. 2013 Jun;16(3):246–54.
- 54 Morita C, Tsuji H, Hata T, Gondo M, Takakura S, Kawai K, et al. Gut Dysbiosis in Patients with Anorexia Nervosa. *PLoS One*. 2015 Dec; 10(12):e0145274.
- 55 Unger MM, Spiegel J, Dillmann KU, Grundmann D, Philippeit H, Bürmann J, et al. Short chain fatty acids and gut microbiota differ between patients with Parkinson's disease and age-matched controls. *Parkinsonism Relat Disord*. 2016 Nov;32:66–72.
- 56 Maltz RM, Keirsey J, Kim SC, Mackos AR, Gharabeh RZ, Moore CC, et al. Prolonged restraint stressor exposure in outbred CD-1 mice impacts microbiota, colonic inflammation, and short chain fatty acids. *PLoS One*. 2018 May;13(5):e0196961.
- 57 Zhang L, Wang Y, Xiayu X, Shi C, Chen W, Song N, et al. Altered Gut Microbiota in a Mouse Model of Alzheimer's Disease. *J Alzheimers Dis*. 2017;60(4):1241–57.
- 58 Rahat-Rozenbloom S, Fernandes J, Gloor GB, Wolever TM. Evidence for greater production of colonic short-chain fatty acids in overweight than lean humans. *Int J Obes*. 2014 Dec;38(12):1525–31.
- 59 Fernandes J, Su W, Rahat-Rozenbloom S, Wolever TM, Comelli EM. Adiposity, gut microbiota and faecal short chain fatty acids are linked in adult humans. *Nutr Diabetes*. 2014 Jun;4(6):e121.
- 60 Schwirtz A, Taras D, Schäfer K, Beijer S, Bos NA, Donus C, et al. Microbiota and SCFA in lean and overweight healthy subjects. *Obesity (Silver Spring)*. 2010 Jan;18(1):190–5.
- 61 Wang L, Christophersen CT, Sorich MJ, Gerber JP, Angley MT, Conlon MA. Elevated fecal short chain fatty acid and ammonia concentrations in children with autism spectrum disorder. *Dig Dis Sci*. 2012 Aug;57(8):2096–102.
- 62 Michels N, Van de Wiele T, De Henauw S. Chronic Psychosocial Stress and Gut Health in Children: Associations With Calprotectin and Fecal Short-Chain Fatty Acids. *Psychosom Med*. 2017 Oct;79(8):927–35.
- 63 Kekuda R, Manoharan P, Baseler W, Sundaram U. Monocarboxylate 4 mediated butyrate transport in a rat intestinal epithelial cell line. *Dig Dis Sci*. 2013 Mar;58(3):660–7.
- 64 Tamai I, Takanaga H, Maeda H, Sai Y, Ogihara T, Higashida H, et al. Participation of a proton-cotransporter, MCT1, in the intestinal transport of monocarboxylic acids. *Biochem Biophys Res Commun*. 1995 Sep; 214(2):482–9.
- 65 Shin HJ, Anzai N, Enomoto A, He X, Kim DK, Endou H, et al. Novel liver-specific organic anion transporter OAT7 that operates the exchange of sulfate conjugates for short chain fatty acid butyrate. *Hepatology*. 2007 Apr; 45(4):1046–55.
- 66 Miyauchi S, Gopal E, Fei YJ, Ganapathy V. Functional identification of SLC5A8, a tumor suppressor down-regulated in colon cancer, as a Na(+)-coupled transporter for short-chain fatty acids. *J Biol Chem*. 2004 Apr; 279(14):13293–6.
- 67 Mascolo N, Rajendran VM, Binder HJ. Mechanism of short-chain fatty acid uptake by apical membrane vesicles of rat distal colon. *Gastroenterology*. 1991 Aug;101(2):331–8.
- 68 Karaki S, Tazoe H, Hayashi H, Kashiwabara H, Tooyama K, Suzuki Y, et al. Expression of the short-chain fatty acid receptor, GPR43, in the human colon. *J Mol Histol*. 2008 Apr; 39(2):135–42.
- 69 Karaki S, Mitsui R, Hayashi H, Kato I, Sugiyama H, Iwanaga T, et al. Short-chain fatty acid receptor, GPR43, is expressed by enteroendocrine cells and mucosal mast cells in rat intestine. *Cell Tissue Res*. 2006 Jun;324(3):353–60.
- 70 Tazoe H, Otomo Y, Kaji I, Tanaka R, Karaki SI, Kuwahara A. Roles of short-chain fatty acids receptors, GPR41 and GPR43 on colonic functions. *J Physiol Pharmacol*. 2008 Aug;59 Suppl 2:251–62.
- 71 Priori D, Colombo M, Clavenzani P, Jansman AJ, Lallès JP, Trevisi P, et al. The Olfactory Receptor OR51E1 Is Present along the Gastrointestinal Tract of Pigs, Co-Localizes with Enteroendocrine Cells and Is Modulated by Intestinal Microbiota. *PLoS One*. 2015 Jun; 10(6):e0129501.
- 72 Fleischer J, Bumbalo R, Bautze V, Strotmann J, Breer H. Expression of odorant receptor Olfr78 in enteroendocrine cells of the colon. *Cell Tissue Res*. 2015 Sep;361(3):697–710.
- 73 Kaji I, Akiba Y, Konno K, Watanabe M, Kimura S, Iwanaga T, et al. Neural FFA3 activation inversely regulates anion secretion evoked by nicotinic ACh receptor activation in rat proximal colon. *J Physiol*. 2016 Jun; 594(12):3339–52.
- 74 De Vadder F, Kovatcheva-Datchary P, Gonçalves D, Vinera J, Zitoun C, Duchamp A, et al. Microbiota-generated metabolites promote metabolic benefits via gut-brain neural circuits. *Cell*. 2014 Jan;156(1–2):84–96.
- 75 Nøhr MK, Egerod KL, Christiansen SH, Gille A, Offermanns S, Schwartz TW, et al. Expression of the short chain fatty acid receptor GPR41/FFAR3 in autonomic and somatic sensory ganglia. *Neuroscience*. 2015 Apr;290:126–37.
- 76 Won YJ, Lu VB, Puhl HL 3rd, Ikeda SR. β -Hydroxybutyrate modulates N-type calcium channels in rat sympathetic neurons by acting as an agonist for the G-protein-coupled receptor FFA3. *J Neurosci*. 2013 Dec; 33(49):19314–25.
- 77 Hoyles L, Snelling T, Umlai UK, Nicholson JK, Carding SR, Glen RC, et al. Microbiome-host systems interactions: protective effects of propionate upon the blood-brain barrier. *Microbiome*. 2018 Mar;6(1):55.
- 78 Gribble FM, Reimann F. Enteroendocrine Cells: Chemosensors in the Intestinal Epithelium. *Annu Rev Physiol*. 2016;78(1):277–99.
- 79 De Silva A, Bloom SR. Gut Hormones and Appetite Control: A Focus on PYY and GLP-1 as Therapeutic Targets in Obesity. *Gut Liver*. 2012 Jan;6(1):10–20.

- 80 Richards P, Parker HE, Adriaenssens AE, Hodgson JM, Cork SC, Trapp S, et al. Identification and characterization of GLP-1 receptor-expressing cells using a new transgenic mouse model. *Diabetes*. 2014 Apr;63(4):1224–33.
- 81 Furness JB. The enteric nervous system and neurogastroenterology. *Nat Rev Gastroenterol Hepatol*. 2012 Mar;9(5):286–94.
- 82 Wehrwein EA, Oler HS, Barman SM. Overview of the Anatomy, Physiology, and Pharmacology of the Autonomic Nervous System. *Compr Physiol*. 2016 Jun;6(3):1239–78.
- 83 Vermeulen W, De Man JG, Pelckmans PA, De Winter BY. Neuroanatomy of lower gastrointestinal pain disorders. *World J Gastroenterol*. 2014 Jan;20(4):1005–20.
- 84 Brookes S, Chen N, Humenick A, Spencer NJ, Costa M. Extrinsic sensory innervation of the gut: structure and function. *Adv Exp Med Biol*. 2016;891:63–9.
- 85 Rosenbaum T, Simon SA. TRPV1 receptors and signal transduction. In: Liedtke WB, Heller S, editors. TRP ion channel function in sensory transduction and cellular signaling cascades. Boca Raton: CRC Press/Taylor & Francis; 2007.
- 86 Cortright DN and Szallasi A: TRP channels and pain. *Curr Pharm Des*. 2009;15:1736–49.
- 87 Schemann M, Neunlist M. The human enteric nervous system. *Neurogastroenterol Motil*. 2004 Apr;16(s1 Suppl 1):55–9.
- 88 Anlauf M, Schäfer MK, Eiden L, Weihe E. Chemical coding of the human gastrointestinal nervous system: cholinergic, VIPergic, and catecholaminergic phenotypes. *J Comp Neurol*. 2003 Apr;459(1):90–111.
- 89 Spencer NJ, Hibberd TJ, Lagerström M, Otsuka Y, Kelley N. Visceral pain - Novel approaches for optogenetic control of spinal afferents. *Brain Res*. 2018 Aug;1693 Pt B:159–64.
- 90 Hibberd TJ, Zagorodnyuk VP, Spencer NJ, Brookes SJ. Identification and mechanosensitivity of viscerofugal neurons. *Neuroscience*. 2012 Dec;225:118–29.
- 91 Szurszewski JH. Physiology of mammalian prevertebral ganglia. *Annu Rev Physiol*. 1981; 43(1):53–68.
- 92 Christianson JA, Bielefeldt K, Altier C, Cenac N, Davis BM, Gebhart GF, et al. Development, plasticity and modulation of visceral afferents. *Brain Res Brain Res Rev*. 2009 Apr; 60(1):171–86.
- 93 Ren K, Randich A, Gebhart GF. Vagal afferent modulation of spinal nociceptive transmission in the rat. *J Neurophysiol*. 1989 Aug; 62(2):401–15.
- 94 De Kloet ER, Oitzl MS, Schöbitz B. Cytokines and the brain corticosteroid receptor balance: relevance to pathophysiology of neuroendocrine-immune communication. *Psychoneuroendocrinology*. 1994;19(2):121–34.
- 95 Furtado M, Katzman MA. Examining the role of neuroinflammation in major depression. *Psychiatry Res*. 2015 Sep;229(1-2):27–36.
- 96 Eley TC, Stevenson J. Specific life events and chronic experiences differentially associated with depression and anxiety in young twins. *J Abnorm Child Psychol*. 2000 Aug;28(4):383–94.
- 97 Berk M, Brnabic A, Dodd S, Kelin K, Tohen M, Malhi GS, et al. Does stage of illness impact treatment response in bipolar disorder? Empirical treatment data and their implication for the staging model and early intervention. *Bipolar Disord*. 2011 Feb;13(1):87–98.
- 98 Bauer ME, Wieck A, Lopes RP, Teixeira AL, Grassi-Oliveira R. Interplay between neuro-immunoendocrine systems during post-traumatic stress disorder: a minireview. *Neuroimmunomodulation*. 2010; 17(3): 192–5.
- 99 Rogers J, Shen Y. A perspective on inflammation in Alzheimer's disease. *Ann N Y Acad Sci*. 2000;924(1):132–5.
- 100 Ricci S, Fuso A, Ippoliti F, Businaro R. Stress-induced cytokines and neuronal dysfunction in Alzheimer's disease. *J Alzheimers Dis*. 2012;28(1):11–24.
- 101 Schultze-Lutter F, Ruhmann S, Fusar-Poli P, Bechdolf A, Schimmelmann BG, Klosterkötter J. Basic symptoms and the prediction of first-episode psychosis. *Curr Pharm Des*. 2012;18(4):351–7.
- 102 Bailey MT. Influence of stressor-induced nervous system activation on the intestinal microbiota and the importance for immunomodulation. *Adv Exp Med Biol*. 2014;817: 255–76.
- 103 Bailey MT, Dowd SE, Galley JD, Hufnagle AR, Allen RG, Lyte M. Exposure to a social stressor alters the structure of the intestinal microbiota: implications for stressor-induced immunomodulation. *Brain Behav Immun*. 2011 Mar;25(3):397–407.
- 104 Bharwani A, Mian MF, Foster JA, Surette MG, Bienenstock J, Forsythe P. Structural & functional consequences of chronic psychosocial stress on the microbiome & host. *Psychoneuroendocrinology*. 2016 Jan;63:217–27.
- 105 De Palma G, Collins SM, Bercik P. The microbiota-gut-brain axis in functional gastrointestinal disorders. *Gut Microbes*. 2014 May-Jun;5(3):419–29.
- 106 De Palma G, Blennerhassett P, Lu J, Deng Y, Park AJ, Green W, et al. Microbiota and host determinants of behavioural phenotype in maternally separated mice. *Nat Commun*. 2015 Jul;6(1):7735.
- 107 Moloney RD, O'Mahony SM, Dinan TG, Cryan JF. Stress-induced visceral pain: toward animal models of irritable-bowel syndrome and associated comorbidities. *Front Psychiatry*. 2015 Feb;6:15.
- 108 Golubeva AV, Crampton S, Desbonnet L, Edge D, O'Sullivan O, Lomasney KW, et al. Prenatal stress-induced alterations in major physiological systems correlate with gut microbiota composition in adulthood. *Psychoneuroendocrinology*. 2015 Oct;60:58–74.
- 109 Galley JD, Bailey MT. Impact of stressor exposure on the interplay between commensal microbiota and host inflammation. *Gut Microbes*. 2014 May-Jun;5(3):390–6.
- 110 Desbonnet L, Garrett L, Clarke G, Kiely B, Cryan JF, Dinan TG. Effects of the probiotic *Bifidobacterium infantis* in the maternal separation model of depression. *Neuroscience*. 2010 Nov;170(4):1179–88.
- 111 Gareau MG, Jury J, MacQueen G, Sherman PM, Perdue MH. Probiotic treatment of rat pups normalises corticosterone release and ameliorates colonic dysfunction induced by maternal separation. *Gut*. 2007 Nov;56(11): 1522–8.
- 112 McKernan DP, Fitzgerald P, Dinan TG, Cryan JF: The probiotic *Bifidobacterium infantis* 35624 displays visceral antinociceptive effects in the rat. *Neurogastroenterol Motil*. 2010;22: 1029–35.
- 113 Savignac HM, Kiely B, Dinan TG, Cryan JF. *Bifidobacteria* exert strain-specific effects on stress-related behavior and physiology in BALB/c mice. *Neurogastroenterol Motil*. 2014 Nov;26(11):1615–27.
- 114 Palomar MM, Maldonado Galdeano C, Perdigón G. Influence of a probiotic lactobacillus strain on the intestinal ecosystem in a stress model mouse. *Brain Behav Immun*. 2014 Jan;35:77–85.
- 115 Bravo JA, Forsythe P, Chew MV, Escaravage E, Savignac HM, Dinan TG, et al. Ingestion of *Lactobacillus* strain regulates emotional behavior and central GABA receptor expression in a mouse via the vagus nerve. *Proc Natl Acad Sci USA*. 2011 Sep;108(38): 16050–5.
- 116 Ait-Belgnaoui A, Durand H, Cartier C, Chaumaz G, Eutamene H, Ferrier L, et al. Prevention of gut leakiness by a probiotic treatment leads to attenuated HPA response to an acute psychological stress in rats. *Psychoneuroendocrinology*. 2012 Nov;37(11): 1885–95.
- 117 Desbonnet L, Clarke G, Traplin A, O'Sullivan O, Crispie F, Moloney RD, et al. Gut microbiota depletion from early adolescence in mice: implications for brain and behaviour. *Brain Behav Immun*. 2015 Aug;48:165–73.
- 118 Matsuo K, Zhang X, Ono Y, Nagatomi R. Acute stress-induced colonic tissue HSP70 expression requires commensal bacterial components and intrinsic glucocorticoid. *Brain Behav Immun*. 2009 Jan;23(1):108–15.
- 119 O'Mahony SM, Felice VD, Nally K, Savignac HM, Claesson MJ, Scully P, et al. Disturbance of the gut microbiota in early-life selectively affects visceral pain in adulthood without impacting cognitive or anxiety-related behaviors in male rats. *Neuroscience*. 2014 Sep;277:885–901.
- 120 Sudo N, Chida Y, Aiba Y, Sonoda J, Oyama N, Yu XN, et al. Postnatal microbial colonization programs the hypothalamic-pituitary-adrenal system for stress response in mice. *J Physiol*. 2004 Jul;558(Pt 1):263–75.
- 121 Neufeld KM, Kang N, Bienenstock J, Foster JA. Reduced anxiety-like behavior and central neurochemical change in germ-free mice. *Neurogastroenterol Motil*. 2011 Mar; 23(3):255–64.

- 122 Asano Y, Hiramoto T, Nishino R, Aiba Y, Kimura T, Yoshihara K, et al. Critical role of gut microbiota in the production of biologically active, free catecholamines in the gut lumen of mice. *Am J Physiol Gastrointest Liver Physiol*. 2012 Dec;303(11):G1288–95.
- 123 Nishino R, Mikami K, Takahashi H, Tomonaga S, Furuse M, Hiramoto T, et al. Commensal microbiota modulate murine behaviors in a strictly contamination-free environment confirmed by culture-based methods. *Neurogastroenterol Motil*. 2013 Jun;25(6):521–8.
- 124 Messaoudi S, Manai M, Kergourlay G, Prévost H, Connil N, Chobert JM, et al. Lactobacillus salivarius: bacteriocin and probiotic activity. *Food Microbiol*. 2013 Dec;36(2):296–304.
- 125 Clarke G, Cryan JF, Dinan TG, Quigley EM. Review article: probiotics for the treatment of irritable bowel syndrome—focus on lactic acid bacteria. *Aliment Pharmacol Ther*. 2012 Feb;35(4):403–13.
- 126 Nayak D, Zinselmeyer BH, Corps KN, McGavern DB. In vivo dynamics of innate immune sentinels in the CNS. *Intravital*. 2012;1(2):95–106.
- 127 Braniste V, Al-Asmakh M, Kowal C, Anuar F, Abbaspour A, Tóth M, et al. The gut microbiota influences blood-brain barrier permeability in mice. *Sci Transl Med*. 2014 Nov;6(263):263ra158.
- 128 Wohleb ES, McKim DB, Sheridan JF, Godbout JP. Monocyte trafficking to the brain with stress and inflammation: a novel axis of immune-to-brain communication that influences mood and behavior. *Front Neurosci*. 2015 Jan;8:447.
- 129 Nilsson NE, Kotarsky K, Owman C, Olde B. Identification of a free fatty acid receptor, FFA2R, expressed on leukocytes and activated by short-chain fatty acids. *Biochem Biophys Res Commun*. 2003 Apr;303(4):1047–52.
- 130 Tomova A, Husarova V, Lakatosova S, Bakos J, Vlkova B, Babinska K, et al. Gastrointestinal microbiota in children with autism in Slovakia. *Physiol Behav*. 2015 Jan;138:179–87.
- 131 Finegold SM, Summanen PH, Downes J, Corbett K, Komoriya T. Detection of Clostridium perfringens toxin genes in the gut microbiota of autistic children. *Anaerobe*. 2017 Jun;45:133–7.
- 132 Parracho HM, Bingham MO, Gibson GR, McCartney AL. Differences between the gut microflora of children with autistic spectrum disorders and that of healthy children. *J Med Microbiol*. 2005 Oct;54(Pt 10):987–91.
- 133 Wang L, Angley MT, Gerber JP, Young RL, Abarno DV, McKinnon RA, et al. Is urinary indolyl-3-acryloylglycine a biomarker for autism with gastrointestinal symptoms? *Biomarkers*. 2009 Dec;14(8):596–603.
- 134 Wang L, Conlon MA, Christophersen CT, Sorich MJ, Angley MT. Gastrointestinal microbiota and metabolite biomarkers in children with autism spectrum disorders. *Biomarkers Med*. 2014;8(3):331–44.
- 135 Critchfield JW, van Hemert S, Ash M, Mulder L, Ashwood P. The potential role of probiotics in the management of childhood autism spectrum disorders. *Gastroenterol Res Pract*. 2011;2011:161358.
- 136 Dickerson F, Severance E, Yolken R. The microbiome, immunity, and schizophrenia and bipolar disorder. *Brain Behav Immun*. 2017 May;62:46–52.
- 137 Severance EG, Yolken RH, Eaton WW. Autoimmune diseases, gastrointestinal disorders and the microbiome in schizophrenia: more than a gut feeling. *Schizophr Res*. 2016 Sep;176(1):23–35.
- 138 Lv F, Chen S, Wang L, Jiang R, Tian H, Li J, et al. The role of microbiota in the pathogenesis of schizophrenia and major depressive disorder and the possibility of targeting microbiota as a treatment option. *Oncotarget*. 2017 Sep;8(59):100899–907.
- 139 Schwarz E, Maukonen J, Hyytiäinen T, Kieseppä T, Orešič M, Sabuncuyan S, et al. Analysis of microbiota in first episode psychosis identifies preliminary associations with symptom severity and treatment response. *Schizophr Res*. 2018 Feb;192:398–403.
- 140 Cenit MC, Nuevo IC, Codoñer-Franch P, Dinan TG, Sanz Y. Gut microbiota and attention deficit hyperactivity disorder: new perspectives for a challenging condition. *Eur Child Adolesc Psychiatry*. 2017 Sep;26(9):1081–92.
- 141 Painold A, Mörl S, Kashofer K, Halwachs B, Dalkner N, Bengesser S, et al. A step ahead: exploring the gut microbiota in inpatients with bipolar disorder during a depressive episode. *Bipolar Disord*. 2019 Feb;21(1):40–9.
- 142 Evans SJ, Bassis CM, Hein R, Assari S, Flowers SA, Kelly MB, et al. The gut microbiome composition associates with bipolar disorder and illness severity. *J Psychiatr Res*. 2017 Apr;87:23–9.
- 143 Flowers SA, Evans SJ, Ward KM, McInnis MG, Ellingrod VL. Interaction Between Atypical Antipsychotics and the Gut Microbiome in a Bipolar Disease Cohort. *Pharmacotherapy*. 2017 Mar;37(3):261–7.
- 144 Bengesser SA, Mörl S, Painold A, Dalkner N, Birner A, Fellendorf FT, et al. Epigenetics of the molecular clock and bacterial diversity in bipolar disorder. *Psychoneuroendocrinology*. 2019 Mar;101:160–6.
- 145 Jiang H, Ling Z, Zhang Y, Mao H, Ma Z, Yin Y, et al. Altered fecal microbiota composition in patients with major depressive disorder. *Brain Behav Immun*. 2015 Aug;48:186–94.
- 146 Liu Y, Zhang L, Wang X, Wang Z, Zhang J, Jiang R, Wang X, Wang K, Liu Z, et al. Similar fecal microbiota signatures in patients with diarrhea-predominant irritable bowel syndrome and patients with depression. *Clin Gastroenterol Hepatol*. 2016;14:1602–11.
- 147 Aizawa E, Tsuji H, Asahara T, Takahashi T, Teraishi T, Yoshida S, et al. Possible association of Bifidobacterium and Lactobacillus in the gut microbiota of patients with major depressive disorder. *J Affect Disord*. 2016 Sep;202:254–7.
- 148 Naseribafrouei A, Hestad K, Avershina E, Sekelja M, Linløkken A, Wilson R, et al. Correlation between the human fecal microbiota and depression. *Neurogastroenterol Motil*. 2014 Aug;26(8):1155–62.
- 149 Jeffery IB, Quigley EM, Öhman L, Simrén M, O'Toole PW. The microbiota link to irritable bowel syndrome: an emerging story. *Gut Microbes*. 2012 Nov-Dec;3(6):572–6.
- 150 Hemmings SM, Malan-Müller S, van den Heuvel LL, Demmitt BA, Stanislawski MA, Smith DG, et al. The Microbiome in Post-traumatic Stress Disorder and Trauma-Exposed Controls: An Exploratory Study. *Psychosom Med*. 2017 Oct;79(8):936–46.
- 151 Köhler CA, Maes M, Slyepchenko A, Berk M, Solmi M, Lanctôt KL, et al. The Gut-Brain Axis, Including the Microbiome, Leaky Gut and Bacterial Translocation: Mechanisms and Pathophysiological Role in Alzheimer's Disease. *Curr Pharm Des*. 2016;22(40):6152–66.
- 152 Vogt NM, Kerby RL, Dill-McFarland KA, Harding SJ, Merluzzi AP, Johnson SC, et al. Gut microbiome alterations in Alzheimer's disease. *Sci Rep*. 2017 Oct;7(1):13537.
- 153 Zhuang ZQ, Shen LL, Li WW, Fu X, Zeng F, Gui L, et al. Gut Microbiota is Altered in Patients with Alzheimer's Disease. *J Alzheimers Dis*. 2018;63(4):1337–46.
- 154 Scheperjans F, Aho V, Pereira PA, Koskinen K, Paulin L, Pekkonen E, et al. Gut microbiota are related to Parkinson's disease and clinical phenotype. *Mov Disord*. 2015 Mar;30(3):350–8.
- 155 Keshavarzian A, Green SJ, Engen PA, Voigt RM, Naqib A, Forsyth CB, et al. Colonic bacterial composition in Parkinson's disease. *Mov Disord*. 2015 Sep;30(10):1351–60.
- 156 Hill-Burns EM, Debelius JW, Morton JT, Wissemann WT, Lewis MR, Wallen ZD, et al. Parkinson's disease and Parkinson's disease medications have distinct signatures of the gut microbiome. *Mov Disord*. 2017 May;32(5):739–49.
- 157 Bedarf JR, Hildebrand F, Coelho LP, Sunagawa S, Bahram M, Goer F, et al. Functional implications of microbial and viral gut metagenome changes in early stage L-DOPA-naïve Parkinson's disease patients. *Genome Med*. 2017 Apr;9(1):39.
- 158 Tillisch K, Labus J, Kilpatrick L, Jiang Z, Stains J, Ebrat B, Guyonnet D, Legrain-Raspaud S, Trotin B, et al. Consumption of fermented milk product with probiotic modulates brain activity. *Gastroenterology*. 2013;144:1394–401.
- 159 Bagga D, Reichert JL, Koschutnig K, Aigner CS, Holzer P, Koskinen K, et al. Probiotics drive gut microbiome triggering emotional brain signatures. *Gut Microbes*. 2018 Nov;9(6):486–96.

- 160 Benton D, Williams C, Brown A. Impact of consuming a milk drink containing a probiotic on mood and cognition. *Eur J Clin Nutr*. 2007 Mar;61(3):355–61.
- 161 Rao AV, Bsted AC, Beaulne TM, Katzman MA, Iorio C, Berardi JM, et al. A randomized, double-blind, placebo-controlled pilot study of a probiotic in emotional symptoms of chronic fatigue syndrome. *Gut Pathog*. 2009 Mar;1(1):6.
- 162 Kato-Kataoka A, Nishida K, Takada M, Suda K, Kawai M, Shimizu K, et al. Fermented milk containing *Lactobacillus casei* strain Shirota prevents the onset of physical symptoms in medical students under academic examination stress. *Benef Microbes*. 2016; 7(2):153–6.
- 163 Takada M, Nishida K, Gondo Y, Kikuchi-Hayakawa H, Ishikawa H, Suda K, et al. Beneficial effects of *Lactobacillus casei* strain Shirota on academic stress-induced sleep disturbance in healthy adults: a double-blind, randomised, placebo-controlled trial. *Benef Microbes*. 2017 Apr;8(2):153–62.
- 164 Andersson H, Tullberg C, Ahnén S, Hamberg K, Lazou Ahnén I, Molin G, et al. Oral administration of *Lactobacillus plantarum* 299v reduces cortisol levels in human saliva during examination induced stress: a randomized, double-blind controlled trial. *Int J Microbiol*. 2016;2016:8469018.
- 165 Messaoudi M, Violle N, Bisson JF, Desor D, Javelot H, Rougeot C. Beneficial psychological effects of a probiotic formulation (*Lactobacillus helveticus* R0052 and *Bifidobacterium longum* R0175) in healthy human volunteers. *Gut Microbes*. 2011 Jul-Aug;2(4): 256–61.
- 166 Colica C, Avolio E, Bollero P, Costa de Miranda R, Ferraro S, Sinibaldi Salimei P, et al. Evidences of a new psychobiotic formulation on body composition and anxiety. *Mediators Inflamm*. 2017;2017:5650627.
- 167 Nishida K, Sawada D, Kawai T, Kuwano Y, Fujiwara S, Rokutan K. Para-psychobiotic *Lactobacillus gasseri* CP2305 ameliorates stress-related symptoms and sleep quality. *J Appl Microbiol*. 2017 Dec;123(6):1561–70.
- 168 Savignac HM, Tramullas M, Kiely B, Dinan TG, Cryan JF. *Bifidobacteria* modulate cognitive processes in an anxious mouse strain. *Behav Brain Res*. 2015;287:59–72.
- 169 Allen AP, Hutch W, Borre YE, Kennedy PJ, Temko A, Boylan G, et al. *Bifidobacterium longum* 1714 as a translational psychobiotic: modulation of stress, electrophysiology and neurocognition in healthy volunteers. *Transl Psychiatry*. 2016 Nov;6(11):e939.
- 170 Kelly JR, Allen AP, Temko A, Hutch W, Kennedy PJ, Farid N, et al. Lost in translation? The potential psychobiotic *Lactobacillus rhamnosus* (JB-1) fails to modulate stress or cognitive performance in healthy male subjects. *Brain Behav Immun*. 2017 Mar;61: 50–9.
- 171 Reininghaus EZ, Wetzlmair LC, Fellendorf FT, Platzer M, Queissner R, Birner A, et al. The impact of probiotic supplements on cognitive parameters in euthymic individuals with bipolar disorder: a pilot study. *Neuropsychobiology*. 2018 Sep;18:1–8.
- 172 Yolken R, Adamos M, Katsafanas E, Khushalani S, Origoni A, Savage C, et al. Individuals hospitalized with acute mania have increased exposure to antimicrobial medications. *Bipolar Disord*. 2016 Aug;18(5): 404–9.
- 173 Kang DW, Adams JB, Gregory AC, Borody T, Chittick L, Fasano A, et al. Microbiota Transfer Therapy alters gut ecosystem and improves gastrointestinal and autism symptoms: an open-label study. *Microbiome*. 2017 Jan;5(1):10.
- 174 Pärtty A, Kalliomäki M, Wacklin P, Salminen S, Isolauri E. A possible link between early probiotic intervention and the risk of neuropsychiatric disorders later in childhood: a randomized trial. *Pediatr Res*. 2015 Jun; 77(6):823–8.
- 175 Akkasheh G, Kashani-Poor Z, Tajabadi-Ebrahimi M, Jafari P, Akbari H, Taghizadeh M, et al. Clinical and metabolic response to probiotic administration in patients with major depressive disorder: A randomized, double-blind, placebo-controlled trial. *Nutrition*. 2016 Mar;32(3):315–20.
- 176 Kazemi A, Noorbala AA, Azam K, Eskandari MH, Djafarian K. Effect of probiotic and prebiotic vs placebo on psychological outcomes in patients with major depressive disorder: a randomized clinical trial. *Clin Nutr*. 2019 Apr;38(2):522–8.
- 177 Slykerman RF, Hood F, Wickens K, Thompson JM, Barthow C, Murphy R, et al.; Probiotic in Pregnancy Study Group. Effect of *Lactobacillus rhamnosus* HN001 in pregnancy on postpartum symptoms of depression and anxiety: a randomised double-blind placebo-controlled trial. *EBioMedicine*. 2017 Oct;24:159–65.
- 178 Romijn AR, Rucklidge JJ, Kuijter RG, Frampton C. A double-blind, randomized, placebo-controlled trial of *Lactobacillus helveticus* and *Bifidobacterium longum* for the symptoms of depression. *Aust N Z J Psychiatry*. 2017 Aug;51(8):810–21.
- 179 Huang R, Wang K, Hu J. Effect of Probiotics on Depression: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. *Nutrients*. 2016 Aug;8(8):8.
- 180 Pirbaglou M, Katz J, de Souza RJ, Stearns JC, Motamed M, Ritvo P. Probiotic supplementation can positively affect anxiety and depressive symptoms: a systematic review of randomized controlled trials. *Nutr Res*. 2016 Sep;36(9):889–98.
- 181 Wallace CJ, Milev R. The effects of probiotics on depressive symptoms in humans: a systematic review. *Ann Gen Psychiatry*. 2017 Feb;16(1):14.
- 182 Ng QX, Peters C, Ho CY, Lim DY, Yeo WS. A meta-analysis of the use of probiotics to alleviate depressive symptoms. *J Affect Disord*. 2018 Mar;228:13–9.
- 183 Clarke G, Sandhu KV, Griffin BT, Dinan TG, Cryan JF, Hyland NP. Gut Reactions: Breaking Down Xenobiotic-Microbiome Interactions. *Pharmacol Rev*. 2019 Apr; 71(2):198–224.
- 184 Falony G, Joossens M, Vieira-Silva S, Wang J, Darzi Y, Faust K, et al. Population-level analysis of gut microbiome variation. *Science*. 2016 Apr;352(6285):560–4.
- 185 Zhernakova A, Kurilshikov A, Bonder MJ, Tigchelaar EF, Schirmer M, Vatanen T, et al.; LifeLines cohort study. Population-based metagenomics analysis reveals markers for gut microbiome composition and diversity. *Science*. 2016 Apr;352(6285):565–9.
- 186 Ticinesi A, Milani C, Lauretani F, Nouvenne A, Mancabelli L, Lugli GA, et al. Gut microbiota composition is associated with polypharmacy in elderly hospitalized patients. *Sci Rep*. 2017 Sep;7(1):11102.
- 187 Kruszewska H, Zareba T, Tyski S. Search of antimicrobial activity of selected non-antibiotic drugs. *Acta Pol Pharm*. 2002 Nov-Dec;59(6):436–9.
- 188 Kruszewska H, Zareba T, Tyski S. Antimicrobial activity of selected non-antibiotics—activity of methotrexate against *Staphylococcus aureus* strains. *Acta Pol Pharm*. 2000 Nov;57 Suppl:117–9.
- 189 Gunics G, Motohashi N, Molnár J, Farkas S, Kawase M, Saito S, et al. Enhanced antibacterial effect of erythromycin in the presence of 3,5-dibenzoyl-1,4-dihydropyridines. *Anticancer Res*. 2001 Jan-Feb;21(1A):269–73.
- 190 Gunics G, Farkas S, Motohashi N, Shah A, Harsukh G, Kawase M, et al. Interaction between 3,5-diacetyl-1,4-dihydropyridines and ampicillin, and erythromycin on different *E. coli* strains. *Int J Antimicrob Agents*. 2002 Sep;20(3):227–9.
- 191 Zhang J, Zhang J, Wang R. Gut microbiota modulates drug pharmacokinetics. *Drug Metab Rev*. 2018 Aug;50(3):357–68.
- 192 Cusotto S, Clarke G, Dinan TG, Cryan JF. Psychotropics and the Microbiome: a Chamber of Secrets.... *Psychopharmacology (Berl)*. 2019 May;236(5):1411–32.
- 193 Sandhu KV, Sherwin E, Schellekens H, Stanton C, Dinan TG, Cryan JF. Feeding the microbiota-gut-brain axis: diet, microbiome, and neuropsychiatry. *Transl Res*. 2017 Jan; 179:223–44.
- 194 Madara JL. Regulation of the movement of solutes across tight junctions. *Annu Rev Physiol*. 1998;60(1):143–59.