Bachelor Degree Project

UNIVERSITY OF SKÖVDE

Is there a Connection Between the Gut-Microbiota and Major Depression?

Bachelor Degree Project in Cognitive Neuroscience
Basic level 22.5 ECTS
Spring term 2020

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Abstract

Major depressive disorder (MDD) is rapidly growing and one of the most common causes of disability and mortality worldwide. People with MDD often display brain changes such as a disrupted balance in neurotransmitters, impaired neurogenesis and neuroplasticity. Traditionally has MDD been treated with medications and talking therapies (psychotherapy). Studies have shown that just around 50 % of people with MDD get improvements from common traditional treatments. Therefore is there a great need for a better understanding of MDD and new treatments. There is now an emerging field of research that indicates that the gut microbiota plays a crucial role in disturbing normal brain functioning in MDD. This connection between the gut and the brain is called the gut-brain axis.

The thesis aims to investigate if there is a connection between gut-microbiota disruption and MDD and if gut-microbiota restoration can be a potential effective future treatment for MDD.

Key findings of the thesis were, studies show that people with MDD often display gut-microbiota disruption and chronic low-grade inflammation. Studies also indicate that this inflammation can cause the specific brain change often displayed in people with MDD. One of the most critical findings in the thesis was that gut-brain treatments affect tryptophan metabolism, which affects the risk of MDD. The research area of the gut-brain axis is still new and many more studies are needed, particularly in humans.

Keywords: Gut-brain axis, Major depressive disorder, Probiotics, Gut-microbiota, low-grade inflammation, Tryptophan.

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Introduction

To start up the day with an energized feeling and no worries in mind is what mental health is for many people, but what makes up the foundation of mental health and mental illnesses such as depression? All people feel probably at some time in life that their mental health has been affected by something and feel tired, gloomy or even depressed. For the majority of the population, these negative feelings are relatively brief and not very long-lasting. For people diagnosed with *Major depressive disorder* (MDD), these negative feelings are long-lasting and negatively affect their daily lives (Liang, Wu, Hu, Wang, & Jin, 2018). MDD is one of the most common causes of disability and mortality the world over. Around 350,000,000 people are already affected by MDD and as many as every fifth people suffer from it in episodes throughout their lives (Rot, Mathew, & Charney, 2009). A majority of people with MDD suffer from suicidal thoughts and around 15–20% die because of suicide (Liang et al., 2018). MDD is according to modern psychology and biology, not just a mental disorder but also a physiological disease. It is often accompanied by brain changes like a disrupted balance in neurotransmitters, impaired neurogenesis and neuroplasticity (Cryan & Dinan, 2012). The majority of researchers agree that depression is caused by a complex mix of genetics and environmental stress. Large genome studies have estimated the heritability of MDD to range from 37 % to 48 %. Certain genes are discussed to be able to predispose some people to depression. Stressful events early in life are also known to play an important role (Liang et al., 2018). Traditionally has MDD been treated with medication and psychotherapy (Roth & Fonagy, 2006). Studies have shown that not all people with MDD get symptom improvements by these treatments (Jenkins, Nguyen, Polglaze, & Bertrand, 2016). Therefore there is a great need for new treatments and a better understanding of the causes of MDD (Liang et al., 2018). There is now a newly emerging field of research, which indicates that the *gut microbiota* plays a crucial role in disturbing normal brain functioning in MDD (Jenkins et al., 2016). People with MDD are known for having a dysregulated *hypothalamus, pituitary gland and adrenal gland axis* (HPA), regulating the stress response (Bailey & Coe, 1999). This dysregulation can affect neurotransmitters such as serotonin, which play an important role in MDD (Dinan, Stanton, & Cryan, 2013). In this new field

of research, studies indicates that there is bidirectional communication between the gut and the brain, which is called the *gut-brain axis* (Desbonnet, Garrett, Clarke, Bienenstock, & Dinan, 2008). These studies investigate if the gut-microbiota can affect our brain and if our brain can affect our gut microbiota via this bidirectional communication. Studies indicate that this gut-brain axis communicates via different pathways like the HPA axis, the immune system and the neuroendocrine system (Cryan & Dinan, 2012). Studies discuss that gut bacteria can produce many neurotransmitters, such as serotonin (Dinan et al., 2013). At the end of the nineteenth century theories about a gut-brain connection were popular among many scientists (Moore, Mathias, & Valeur, 2018). There was much acceptance that diet and the gut were able to affect mental health. Theories and treatment of these patients were seen as quackery in the early twentieth century and the interest and treatment ceased for a long time (Moore et al., 2018). Just recently, in the last 10-15 years, new theories about gut-brain connections have begun to emerge and it has been a trendy scientific topic again. How our gut microbiota affects the risk of MDD has developed into an interdisciplinary and complex field of research (Dinan et al., 2013). Researchers from several different disciplines are involved in research, like bacteriologists, biologists, neuroscientists and physicians. New disciplines like neuro gastroenterology are defined as neurology of the gastrointestinal tract, liver, gallbladder, and pancreas (Fang et al., 2008). Much of the research has been done in animal studies, but in the last couple of years, experimental studies on humans have begun to be published (Guida et al., 2018). There is much hope from many researchers to find new treatments for MDD, but so far, the results from different studies have been ambiguous. Huang, Wang, and Hu (2016) showed in a systematic review, including five studies, a significant effect of *Probiotics* on depression. However did Ng, Peters, Ho, Lim, and Yeo (2018) show in a systematic review including ten studies a non-significant effect of Probiotics on depression. Researchers in both articles discuss these ambiguous results because different studies use different designs and look at how different pathways or probiotics affect the gut-brain axis. This research area is relatively new and complex and there is a need for many more studies, particularly on humans. The thesis aims to investigate if there is a connection between gut-microbiota disruption and MDD and

if gut-microbiota restoration can be a potential effective future treatment for MDD. This thesis is a literature review in cognitive neuroscience and the following databases were used to find relevant articles: PsycINFO, PsycArtickles, MEDLINE, ScienceDirect, Academic Search Elite, Web of Science and Google Scholar. Specific search words such as Probiotics in MDD, *Fecal Microbiota Transplantation* in MDD, *Prebiotics* in MDD, Diet in MDD, inflammation in MDD, brain dysfunction in MDD and treatments in MDD where used in the different databases. The majority of articles were obtained using the search word microbiota-gut-brain axis, which yielded 14800 articles on Google Scholar and 1038 articles on Web of Science. In a refined search on the Web of Science, only articles related to neuroscience were involved, which yielded 300 articles. In this refined search were the first two articles published in 2011 compared to 2019 when 106 articles were published. This increase demonstrates that there is a strong development and increased interest in the research area. The thesis searched particularly for articles that focused on MDD, double-blind experimental studies on humans in the microbiota-gut-brain axis. The majority of the articles the thesis found focused on the physiological part of the research area, such as bacteria and gut function, and not so much on the mental aspects such as MDD. There were more studies concerning animals and the connection between MDD and the gut-brain axis compared to human studies. Therefore these articles were few and harder to find. A majority of the articles concerning humans and MDD in the gut-brain axis were found in review articles, which often were very positive and biased toward the gut-brain axis. The thesis is divided into three main sections. These three are Major Depression, Gut-Brain Axis and Gut-Brain Treatments and Major Depression. Section one, Major Depression, will begin with a brief analysis of the most common symptoms in major depression. This will be followed by an investigation of brain dysfunctions, such as which brain areas and functions are affected in people with MDD. The section will end with common traditional treatments in MDD. Section two, the Gut-Brain Axis will try to give an overview of the background research of the bidirectional communication between the gut and the brain and how the research has developed into today's hot research area. This will be followed by a brief overview of the connection between gut bacteria and the brain and which different pathways these bacteria use for

communication with the brain. There will be some focus on animal studies since much of the research so far on the gut-brain axis has been pre-clinical animal studies and most of the latest research is based on these animal studies. In contrast to animal studies, the section ends with studies concerning inflammation in humans, which play an important role in MDD. Section three, Gut-Brain Treatments and Major Depression will focus on human studies and potential future treatments for MDD. There will be a focus on four different methods: Fecal Microbiota Transplantation, healthy diet, Prebiotics and Probiotics. The thesis concludes with a discussion and a conclusion.

Major Depression

People diagnosed with MDD often displays specific brain change and dysfunction (Rot et al., 2009). This section explores the foundation of MDD, which, kind of brain change is characteristic for MDD, which symptoms are most common and what is needed to be diagnosed with MDD. The section will end with a summary of conventional traditional treatments for MDD, such as medication and psychotherapy, and the limitations of these treatments.

Symptoms

MDD is defined by the diagnostic and statistical manual of mental disorder (DSM-5, the fifth edition) and several criteria need to be fulfilled to be diagnosed with MDD. Some of these criteria are that a person should have displayed symptoms like a change of moods, low energy and a highly reduced interest in activities that are usually enjoyable each day for at least two weeks (Richards & O'Hara, 2014). Some people with MDD experience depression in periods that can be separated by years, while others suffer from symptoms almost daily throughout life. The symptoms in MDD are often severe and affect the whole life, like work, school, sleeping and eating habits, and general health. Some of these severe symptoms are long-lasting negative thoughts and rumination about suicide, death, worthlessness, inappropriate guilt or regret, helplessness, hopelessness, and self-hatred (Richards & O'Hara, 2014). Patients with MDD symptoms are often assessed by a suitably trained general practitioner or psychiatrist or psychologist. These professionals record the person's current circumstances, psychological, and social factors that may be impacting the

individual's mood. The assessment includes a mental state examination, which investigates the person's current mood and thought content, particularly the presence of hopelessness, pessimism, self-harm or suicide and absence of positive thoughts. These professionals may use rating scales such as the *Hamilton Rating Scale for Depression* or the *Beck Depression Inventory*, which this thesis will discuss in more detail. The score on a rating scale alone is not enough to diagnose depression to the satisfaction of the DSM, but it provides an indication of the severity of symptoms for some time so a person who scores above a certain point can be more thoroughly evaluated for an MDD diagnosis (Richards & O'Hara, 2014).

Brain Dysfunction and Major Depression

In addition to the more typical symptoms that professionals base the MDD diagnosis on, is MDD often characterized by brain alterations (Rot et al., 2009). Brain areas like the prefrontal cortex, hippocampus and amygdala play a crucial role in regulating emotion, stress response, self-control, motivation, and cognitive reaction. Studies have shown that many people with MDD display decreased activity in the prefrontal cortex and hippocampus and increased activity in the amygdala. Studies have shown that MDD can affect the brain at a molecular and cellular level (Rot et al., 2009).

Neurotransmitters play a crucial role in the brain and behavior and researchers claim that MDD is inseparable from neurotransmitter imbalance (Liang et al., 2018). Research indicates that several factors can affect neurotransmitter systems like serotonin, which is important in MDD (Rot et al., 2009). One crucial factor that has been shown to affect serotonin at a molecular level is the *Brain-Derived Neurotrophic Factor* (BDNF). BDNF is important for new neuronal growth (neurogenesis) and survival and crucial for allowing changes in the synapses (synaptic plasticity) throughout life. The raphe nuclei in the brain stem stimulate BDNF genes involved in serotonin function, such as serotonin transporters and the serotonin precursor molecule tryptophan. In turn, serotonin released from the raphe nuclei stimulates BDNF. During brain development, this cyclic process promotes outgrowth, synapse formation, and serotonin neurons (Rot et al., 2009). In a study

demonstrated Matrisciano et al. (2009) significantly lower BDNF levels in participants with MDD compared to healthy controls. The study tested 21 participants with MDD for BDNF levels by blood samples. MDD symptoms were assessed with the Hamilton Rating Scales for Depression (HRSD). The participants were divided into three groups and treated with three different antidepressant drugs, sertraline, escitalopram, or venlafaxine. BDNF levels were measured before the experiment, after five weeks and six months of treatment. Sertraline increased BDNF levels after five weeks and six months of treatment. Venlafaxine increased BDNF levels only after six months. Escitalopram did not affect BDNF levels at all. A significant association was found between the increase in BDNF levels and decreased HRSD scores after six months of treatment (Matrisciano et al., 2009). In contrast to BDNF, explain theories such as the neural circuit hypothesis, brain dysfunction in MDD resulting from distortion in communication between specific neural structures. The ventral tegmental area and the dorsal raphe nucleus are discussed to be involved in this distorted communication (Liang et al., 2018). In accordance with the neural circuit hypothesis did Arnone, McIntosh, Ebmeier, Munafò, and Anderson (2012), in a meta-analysis including 101 articles, demonstrate structural differences in several brain regions in people with MDD. The study compared different studies concerning *magnetic resonance imaging* (MRI) in people with MDD compared to controls. Regions that showed volume reductions were the frontal cortex, orbitofrontal cortex, hippocampus, and right anterior cingulate cortex (ACC). *Functional magnetic resonance imaging* (fMRI) studies on brain activity to sad and happy stimuli have repeatedly shown reduced activities in these regions in people with MDD compared to controls (Arnone et al., 2012). Additionally, did Lee et al. (2007) demonstrate in an fMRI study, lower neural activity in the right hippocampus to negative affective pictures in participants with MDD compared to healthy controls. The severity of MDD correlated with the activity of the left amygdala to negative pictures. The article discussed decreased activation in the right hippocampus in MDD participants to negative emotional stimuli. This finding may be explained by the fact that the hippocampus is a core part of the limbic system and has deep connections to the prefrontal cortex, amygdala and basal ganglia, which can comprise the neural circuit for mood regulation in MDD (Lee et al., 2007). Memory

impairment and HPA axis hyperactivity are often displayed in people with MDD. This HPA axis hyperactivity is discussed to negatively affect hippocampus memory function in people with MDD (Arnone et al., 2012). fMRI studies indicate that individuals with recurrent MDD episodes have relatively small hippocampi, even during improvements (Rot et al., 2009).

The pituitary gland is a part of the HPA axis and was the only structure that showed a volume increase in Arnone et al. (2012). They discuss that the enlargement of the pituitary gland can be due to stress or that hypothalamic released hormones activate the pituitary gland and lead to hyperactivity of the HPA axis. In accordance with Arnone et al. (2012), MacMaster et al. (2006) demonstrate an MRI study that increased pituitary gland volume in boys between 8-17 years old with MDD compared with control subjects. The participants were never treated with antidepressant medication. In contrast to MacMaster et al. (2006) reported Lorenzetti et al. (2009) in an MRI study no pituitary gland enlargement in adult participants with MDD. The study discussed that one possible explanation for the result could be that 21 of the 27 participants were on stable medication six months before and during the experiment. Studies have consistently demonstrated that antidepressant medication directly suppresses HPA axis activity (Lorenzetti et al., 2009).

In contrast to confirming alterations in other areas often associated with MDD did Arnone et al. (2012) not find any significant structural difference in the amygdala when they compared the different articles. Other studies have shown functional abnormalities like metabolic differences and increased activity in the amygdala to sad and happy stimuli in people with MDD compared to controls (Liang et al., 2018). Amygdala is known to play a vital role in regulating emotions (Liang et al., 2018). Studies have shown normalization of amygdala activity and metabolism in people with MDD after six months of treatment with sertraline (Arnone et al., 2012). In contrast to Arnone et al. (2012), Van Eijndhoven et al. (2009) demonstrate in an MRI study the enlargement of the amygdala in 20 people with ongoing MDD episodes compared to healthy controls. When the same participants were tested again after recovering from the depressive episode, they showed no significant difference compared to controls. The amygdala enlargement correlated positively with the severity of the depressive episode. The study discusses that this state related increase in

amygdala volume can, in part, explain why different studies have shown different results (Van Eijndhoven et al., 2009).

The various studies in this section point in the direction that there is a specific alteration in specific brain areas in people with MDD (Arnone et al., 2012). These studies also display that different theories explain MDD from a different perspective, such as the BDNF theories and the neural circuit hypothesis. The findings of brain dysfunction in the hippocampus, amygdala and the pituitary gland can be caused by hyperactivity in the HPA axis is important because people often display HPA axis disruption with MDD (Rot et al., 2009). The thesis will investigate and discuss why the HPA axis is so important for MDD further on.

Treatments

The different symptoms and brain dysfunction in MDD have traditionally been treated with common treatments such as psychotherapy and medication (Roth & Fonagy, 2006). Psychotherapy can be given to individuals, groups, or families by mental health professionals. Cognitive-behavioral therapy (CBT) teaches people with MDD to change counter-productive behaviors and challenge enduring self-defeating ways of thinking. CBT is the psychotherapy that has most research evidence for the treatment of MDD. Research began in the 1990s to indicate that CBT can perform as well as or better than antidepressants in people with moderate to severe MDD (Roth & Fonagy, 2006).

Medication is one of the most common treatments for MDD. Since neurotransmitter imbalance playing a large role in MDD and for the rest of the brain and behavior aims most medication treatments to restore this imbalance (Liang et al., 2018). Monoaminergic neurotransmitters like serotonin, norepinephrine, and dopamine are crucial for moods such as happiness. Change of moods that leads to MDD is linked to insufficient levels of these neurotransmitters. Recovering these neurotransmitter levels will have antidepressive effects, but the problem is that drugs such as selective serotonin reuptake inhibitors (SSRIs) work slowly (Liang et al., 2018). Around 48 % of people with moderate to severe MDD have an effect of SSRI treatment compared with 30 % for placebo (Jenkins et al., 2016). Studies concerning antidepressants like

(SSRI) have gilded some conflicting results and have shown to be less effective for people with acute, mild to moderate MDD and more effective for people with chronic or severe MDD (Kirsch et al., 2008). Studies have shown that this higher effectiveness for chronic or severe MDD compared to mild or moderate MDD can, in part be explained by decreased responsiveness to placebo for chronic or severe MDD, rather than to increased effectiveness to medication for chronic or severe MDD (Kirsch et al., 2008). Because only around 50 % of people with MDD get symptom improvement from these traditional treatments, there is a great need for new treatments and a better understanding of what causes MDD.

Gut-Brain Axis

As mentioned above in the previous section, there is a great need to better understand the causes of MDD. Recent research has begun to support the idea that there is bidirectional communication between the gut microbiota and the brain, which may affect the risk of MDD (Dinan et al., 2013). A complex system enables this communication with many different communication pathways. For example, can long term stress cause alteration in the gut-bacteria via the HPA axis, modification of gut-bacteria can, in turn, affect neurotransmitter systems such as serotonin, which can affect the brain and increase the risk of MDD (Lach, Schellekens, Dinan, & Cryan, 2018). This complex system enables the gut microbiota and the brain to communicate in both directions. The idea of a gut-brain connection has a long history. It has recently been a significant breakthrough in understanding the gut microbiota and its importance for both physiological and psychological health (Lach et al., 2018). This section will investigate the foundation of this bidirectional communication between the gut microbiota and the brain, how this can affect the risk of MDD and how this research has developed into the hot research area of today.

Background History of the Gut-Brain Axis

For a long time has well-known scientists such as Claude Bernard, William James, Ivan Pavlov, and Walter Cannon described the bidirectional communication between the gut and the brain and stated the importance of maintaining its homeostasis (Cryan & Dinan, 2012). In the late

nineteenth century, there was a great interest among many scientists on how our brain connected to the stomach and how this connection affected behavior and mental health (Moore et al., 2018). Theories about gut-brain relationships become a less popular research topic in the twentieth century because of popular treatments like herbal medicine and gut purges. The question was also associated with religious diet gurus like John Harvey Kellogg. All these different factors led to the opinion that gut-brain connections were seen as quackery for many years (Moore et al., 2018). Researchers in the late nineteenth century had a holistic view of the human body compared to researchers in the twentieth century. (Moore et al., 2018). In the twentieth century, most of this holistic view disappeared when new forms of anatomical, physiological, and surgical methods discovered new insights into the body's anatomy. These new insights tended to see each body part as isolated from each other. In today's modern view of the microbiota-gut-brain axis, it is the holistic view from the late nineteenth-century back (Moore et al., 2018).

Gut-Bacteria and Brain

In contrast to the research in the late nineteenth century, there have in the past 15 years been significant advances in the understanding of the gut-microbiota composition in the genetic level (Cryan & Dinan, 2012). This genetic knowledge has been crucial for understanding gut-microbiota and its interactions with the rest of the body, including the gut-brain axis. The estimated number of different gut-bacteria species differs significantly. There is generally agreed upon that over 1000 different species exist in the adult gut microbiota. The colonization of gut bacteria happens postnatal and begins when vaginal delivery exposes the infant to complex gut microbiota. It takes about one year for the infant to show a complex gut-microbiota comparable to adult-like gut microbiota (Lach et al., 2018). Gut-bacteria can generate many types of neurotransmitters. For example, have studies shown that bacterias such as Lactobacillus spp produce Gamma-Aminobutyric acid (GABA), bacteria Escherichia spp can produce noradrenaline and Candida spp produces serotonin (Cryan & Dinan, 2012). Infection, disease, and antibiotics may temporarily alter the gut microbiota's natural composition, which can affect general health (Liang et al., 2018).

Gut-Microbiota and stress

It has long been known that stress via the HPA axis can influence the composition of our gut microbiota. Chronic stress disrupt the intestinal barrier, making it leaky and increasing bacterial cell wall components such as lipopolysaccharide. Human studies have shown gut-bacteria translocation in stress-related psychiatric disorders such as MDD (Liang et al., 2018). The HPA axis is a complex system with feedback interactions between three components: hypothalamus, pituitary gland and adrenal gland (Dinan et al., 2013). The hypothalamus regulates specific metabolic processes and synthesizes and secretes certain neurohormones. It also controls other functions like hunger, thirst, and sleep. The pituitary gland connects to the hypothalamus and stores and secretes hormones. The adrenal gland is a small conical organ on top of the kidneys that produce a variety of hormones (Dinan et al., 2013). HPA axis regulates and controls reactions to stress and regulates many other body processes, like digestion, the immune system, sexuality, mood, and emotions. For example, under conditions of stress, the HPA axis regulates cortisol secretion. Cortisol is known to affect immune cells in the gut and elsewhere in the body. Studies have shown that cortisol can alter gut function and change the gut microbiota composition (Dinan et al., 2013). Dysfunction of the HPA axis is one of the most important mechanisms behind MDD and causes an imbalance in hormones and neurotransmitters (Liang et al., 2018). Studies have supported the claim that there is a connection between MDD and HPA axis dysfunction by demonstrating an alteration in parts of the HPA axis, such as the pituitary gland in people with MDD (MacMaster et al., 2006). Other studies discuss that HPA axis hyperactivity can negatively affect hippocampus memory function in people with MDD (Arnone et al., 2012). Stress in early-life can result in long-term effects on the gut microbiota composition. Bailey and Coe (1999) demonstrated in a study on rhesus monkeys decreased levels of fecal bacteria when they were separated from their mother in between 6-9 months of age. They also showed an altered fecal microbiota composition when separated three hours from their mother per day compared to non-separated monkeys (Bailey & Coe, 1999). A longitudinal study concerning humans over six years confirmed Kiecolt-Glaser et al. (2003) that prolonged stress in humans can also alter the gut microbiota. The study displayed that caring for a

spouse with dementia or chronic work stress can alter the gut microbiota and increase pro-inflammatory cytokines, which are known to play a role in MDD. The caregivers also displayed higher depressive symptoms and loneliness compared to healthy controls. These symptoms were measured ones each of the six years with the Perceived Stress Scale. Studies in this section have demonstrated that stress via the HPA axis can affect the gut-microbiota both in animals and humans. The HPA axis can affect certain brain areas in people with MDD and therefore play an essential role in MDD (Arnone et al., 2012).

Pathways

The HPA axis is one of several routes where bidirectional communication between the gut microbiota and the brain occurs (Liang et al., 2018). The German scientist Paul Trendelenburg published in 1917 results from his investigation of the gut and its peripheral nervous system (Gershon, 1999). This system is called the *enteric nervous system* (ENS). It is unique because it is the only part of the peripheral nervous system that can operate without any input from the *central nervous system* (CNS). Because of this unique feature, ENS is called the second brain (Gershon, 1999). ENS is one of these pathways which influence and affect our brains in the gut-brain axis. More recently, researchers discovered that other mechanisms are also involved in this bidirectional communication (Cryan & Dinan, 2012). There are indirect and direct pathways that influence and affect CNS. For example, regulating immune activity and the production of pro-inflammatory cytokines can indirectly affect the brain. This process stimulates the HPA axis to produce a corticotropin-releasing hormone (CRH), adrenocorticotropin hormone (ACTH) and cortisol (Dinan et al., 2013). In contrast, the production of neurotransmitters, such as serotonin in the gut, directly impacts our brain because it may enter circulation and cross the blood-brain barrier (Jenkins et al., 2016). The production of serotonin is a complex process which, takes place in several stages. The first stage in the production is tryptophan, essential amino acid and a precursor molecule to many hormones and neurotransmitters such as serotonin. In the next step transforms tryptophan into 5-HT, which in turn converts into serotonin. 95% of the body's 5-HT is located within the gastrointestinal tract, primarily synthesized by enterochromaffin cells, and 5% in the CNS. Tryptophan also

transforms into kynurenine which, is a part of the tryptophan metabolic pathway and accounts for over 95% of the available peripheral tryptophan in mammals. Kynurenine dysregulation is discussed to be involved in many disorders of both the brain and the gastrointestinal tract (Jenkins et al., 2016). Desbonnet et al. (2008) showed in an animal study that Probiotic administration, *Bifidobacteria infantis* bacteria, can increase peripheral tryptophan levels, and alter dopamine and serotonin levels in the frontal cortex and the limbic system. Results from the study showed reduced 5-HT concentration in the frontal cortex. The study associated a reduction in 5-HT with the role serotonin activity has in the prefrontal cortex and the regulation of anxious and emotional states (Desbonnet et al., 2008).

All these facts demonstrate that tryptophan is essential in the gut-brain axis. In accordance with Desbonnet et al. (2008) have some of the most promising studies about how the gut-brain axis affects the risk of MDD been targeting tryptophan metabolism.

Animal Studies

Much of the research on the gut-brain axis has been pre-clinical studies in animals as already displayed above. Some of the most critical research has been done in germ-free animals that lack gut microbiota (Guida et al., 2018). Germ-free animals are bred in a sterile environment to prevent postnatal colonization of their gastrointestinal tract. This method takes advantage of the fact that the prenatal environment in the uterine is sterile and that bacteria colonization of the gastrointestinal tract occurs postnatally in healthy rodents and humans. Germ-free mice allow direct comparison with control mice that have normally colonized guts (Guida et al., 2018). In an early groundbreaking study, Sudo et al. (2004) displayed that gut microbiota plays a role in developing the mice's HPA axis. They used germ-free mice in the study, which showed increased adrenocorticotropic hormone and corticosterone release compared with control mice when they were exposed to mild restraint stress. The stress response could be partially reversed by fecal matter transplantation from control mice and was fully reversed by ingested bacteria: *Bifidobacteri um infantis*. The study showed the earlier colonization of gut bacteria, and the more significant was the reversal effects (Sudo et al., 2004). Studies like this have demonstrated that gut microbiota is

essential for influencing the development of an appropriate stress response later in life. The article discusses a critical window in early life during which colonization of gut-bacteria must occur to ensure the healthy development of the HPA axis (Sudo et al., 2004). Mice are often used in the gut-brain axis research field. Researchers often interpret the mice's behavior to assess if they display behavior similar to human MDD. Guida et al. (2018) demonstrated in an experimental study on mice that antibiotic-induced gut-microbiota disruption leads to depressive-like response and chronic gut inflammation. Depressive-like behavior in the mice was measured with immobility in the tail suspension test (TST) and the force swimming test (FST). In the TST were the mice individually suspended by the tail 50 cm from the floor in six minutes. The mice's immobility was recorded in seconds and monitored by the researchers, defined as when the mice hung passively and utterly motionless with no escape-oriented behavior. In the FST were the mice placed in a cylinder with water and were forced to swim for six minutes. Immobility was measured in seconds and defined as when the mice were not struggling to make any movement and floated to keep the head over the water (Guida et al., 2018).

In another study concerning rats, Ong et al. (2018) demonstrate that diet can structurally change the white matter in the rats' brains. The study also showed that dietary manipulation influenced gut-microbiota composition, brain function, and behavior in the rats. After three weeks on a specific diet were the rats' brains analyzed with *diffusion tensor imaging* (DTI), which used an MRI system. The study displayed increase in the left frontal neocortex and decrease in the neocortex, the corpus callosum, the forebrain and the right external and left internal capsules. The study aimed to increase the general understanding of the connection between brain change and gut microbiota, but do not explain the specific brain change in detail. The study discusses that the observed structural changes may be explained by how gut-microbiota populations influence the activity of genes involved in neuronal myelination. The study demonstrates how new and complex the research field is and the need for more studies that can explain the often very complex results of the studies. There are many promising results from all these animal studies, but it has been harder to

demonstrate the same effects in humans. For example, there is much less support for that diet can affect the human brain compared to what Ong et al. (2018) demonstrated in the rats.

Inflammation and Major Depression

In contrast to all animal studies in the field, some specific and vital aspects of the gut-brain axis concerning MDD have been investigated in humans. One of these aspects is how chronic inflammation affects MDD. Many people with MDD show increased inflammation (Mass, Kubera, & Leunis, 2008). The cytokine hypothesis claims that MDD results from increased pro-inflammatory cytokines and the reduction of anti-inflammatory cytokines. The excessive pro-inflammatory cytokines are discussed to affect the HPA axis, increase the blood-brain barrier's permeability, reduce the synthesis of 5-HT and disturb the glutamate systems (Liang et al., 2018). A study including 51 people with MDD did Mass et al. (2008) demonstrate higher levels of pro-inflammatory cytokines than controls. Blood was taken from the participants for the determination of pro-inflammatory cytokines. They found a correlation between MDD and inflammatory diseases. Findings such as this have given support for the theory that activation of the *inflammatory response system* plays a role in MDD. The study discusses that traditional antidepressant treatments combined with anti-inflammatory methods may lead to better treatments for people with MDD (Mass et al., 2008). In line with Mass et al. (2008) did Chinese researchers Jiang et al. (2015) found in an experimental study that people with MDD had altered fecal microbiota composition compared to controls. In the study were 46 participants diagnosed with MDD and 30 participants were matched healthy controls. The MDD group showed decreased levels of bacteria faecalibacterium ,which is known to have anti-inflammatory effects. The study discusses that this decrease may explain the chronic low-grade inflammatory response, which is often displayed in people with MDD (Jiang et al., 2015). The research on inflammation in MDD is critical because studies have investigated humans and not only animals. Another critical factor is that people with MDD often display increased inflammation combined with altered gut microbiota. These factors support the idea of there being a connection between inflammation and MDD.

Gut-Brain Treatments and Major Depression

The research on the gut-brain axis has, as mentioned before, mostly focused on animal studies. Recently, there has been a great interest in proving results from animal studies in corresponding human studies. There is now a well-known link between gut function and mental health disorders, such as MDD (Lach et al., 2018). Depression is often accompanied by changes in the colon function, altering the gut microbiota's composition and stability. Stress, antibiotics and poor diets can all induce gut-brain axis dysfunction and increase the risk of several diseases such as MDD (Lach et al., 2018).

This section will focus on human studies and gut-brain treatments for MDD. There will be a comparison between four different gut-brain treatments to investigate and display how far the research on humans has come compared to, for example animal studies. These four treatments are Fecal Microbiota Transplantation (FMT), Healthy Diet, Prebiotics and Probiotics. The section will also investigate if gut-brain treatments on humans can be a future effective and complementary treatment for MDD.

Fecal Microbiota Transplantation

Fecal microbiota transplantation is the process of transplanting feces from a healthy donor to a receiver's gut to restore the gut-microbiota (Kelly et al., 2016). FMT has in animal studies shown to reduce symptoms similar to human MDD (Cryan & Dinan, 2012). Few human studies have investigated if FMT can be an effective treatment for MDD. However, did Kelly et al. (2016) demonstrate in a study published in the journal of *Psychiatric Research,* where rats received FMT from people with MDD, their behavior was similar to human MDD. 34 people with MDD matched with 33 healthy people as controls were involved in the study. The MDD group demonstrated higher pro-inflammatory bacteria and increased kynurenine to tryptophan ratio compared to the control group. Rats that received the depression FMT showed increased kynurenine to tryptophan ratio. The study concluded that MDD is associated with a dysregulated tryptophan metabolism, as indicated

by increased kynurenine to tryptophan ratio. This tryptophan dysregulation is discussed to affect neurotransmitter systems such as serotonin (Kelly et al., 2016).

The article is significant because of the experiment's design, which is one of the first to investigate how human FMT can affect the risk of MDD. The most important results from the study are the increase in pro-inflammatory bacteria and kynurenine to tryptophan ratio in the MDD group, which also were transferred to the mice. Measurement of the increase in pro-inflammatory bacteria and increased kynurenine to tryptophan ratio may, in the future, lead to an objective measurement of the risk and severity of MDD. The article has several limitations, humans and rats have different microbiota, and FMT from humans to humans may not have the same effect. The study did not test if FMT from healthy participants would bring elevation in mice with behavior similar to MDD and gut-microbiota disruption. Therefore it is difficult to conclude how well this method will work as a treatment for MDD based on the result (Kelly et al., 2016).

From the very few studies on FMT in humans concerning MDD, it is unlikely that this method will become a treatment for MDD. There will probably be more studies conducted on FMT in humans with MDD because of the indications of its effects on tryptophan metabolism. A better understanding of FMTs effects from human to human is probably of more interest for scientists than to make it a treatment for MDD in this early stage of research.

Healthy Diet

Several studies have investigated if healthy diets can affect the risk of MDD. In contrast to FMT have studies on healthy diets often focused on prevention instead of symptom treatments. Diet is one of the critical factors that can substantially affect the gut microbiota composition (Lach et al., 2018). The detailed dietary analysis combined with DNA sequencing has concluded that long-term nutritional patterns largely determine the gut microbiota's main profile. However, psychological stress or short-term dietary changes can induce specific bacteria changes in the gut-microbiota (Selhub, Logan, & Bested, 2014). Many of these studies concerning diet have problems controlling potential confounding variables such as habitual diets and lifestyle behaviors. Even with these

limitations, much of the data from these studies on humans can help assess the role of different dietary patterns on our gut-microbiota composition and function (Oriach, Robertson, Stanton, Cryan, & Dinan, 2016). Traditional dietary practices, like the Mediterranean and Japanese (include more fruits and vegetables, fish and seafood, cereals with limited processing, fiber, modest amounts of dairy and lean meats) have shown to have 25% to 30% lower risk of MDD compared to a modern Western diet (high in refined carbohydrates, saturated fatty acids, sugar, and food additives) (Selhub et al., 2014). Jacka et al. (2010) showed in a study published in the *American Journal of Psychiatry* concerning women that traditional diets such as the Mediterranean reduced the risk of MDD with 35% compared to the western diet. In the study, where 1,046 women between 20 and 93 years old were randomly selected. The participants were evaluated by a diet quality score for dietary patterns. They were also tested for psychological symptoms like MDD by the 12-item General Health Questionnaire (GHQ-12 score) and a structured clinical interview. Diets like the Mediterranean were associated with lower odds for MDD. Analyses of the study demonstrated that higher diet quality scores were correlated with lower mean GHQ-12 scores.

The result of the study is significant because it indicates that diet can affect the risk of MDD. The study's most considerable strength is a large number of participants and a wide range of ages. These two factors give more credibility to the claim of diet causes the lower mean GHQ-12 scores and not confounding factors such as age or results from individual participants. The experiment uses questionnaires that make it more affordable to test many participants, compared to Kelly et al. (2016), which uses a more expensive design and tested 67 participants. Jacka et al. (2010) recognize that unmeasured confounders can have been factors in the results, such as different lifestyle factors. It is known that lifestyle factors, such as physical activity, can affect the risk of MDD (Selhub et al., 2014). Therefore, is one possible confounding variable that people who eat a diet that gives higher diet quality scores are more physically active. Confounding variables like this may have affected the result of the study. Compared to Kelly et al. (2016) is Jacka et al. (2010) study design more likely to be affected by confounding variables. One reason is that the study only uses questionnaires and no objective measurements such as blood samples, compared to Kelly et al. (2016), which uses

a blood sample to measure the increase in pro-inflammatory bacteria and increased kynurenine to tryptophan ratio. Most questionnaires are only based on how the participants answer the question. This answer may be influenced by factors such as participants thinks there are expectations from the experimenter.

The article is relevant for diet as a future treatment of MDD because it indicates that diet may affect the risk of MDD. However, has the study several limitations, such as no objective measurement. The participants did not have MDD and low control over confounding variables. For a diet to be accepted as a standard complementary treatment of MDD needs more studies that investigate participants with MDD and use objective measurements such as blood samples over a long time. Already do doctors prescribe a healthy diet to patients with a wide range of symptoms (Selhub et al., 2014). Therefore it is likely that diet can be a standard complementary treatment of MDD in a relatively near future if more studies are conducted and confirm the result of Jacka et al. (2010).

Prebiotics

In contrast to FMT and a healthy diet, which are new methods to treat MDD, studies have tested more traditional treatments such as pills' ingestion. Prebiotics is defined as a substrate that selectively gives gut-microbiota health benefits (Bourassa, Alim, Bultman, & Ratan, 2016). Substrates described as Prebiotics are, for example, fructose-oligosaccharide, galactooligosaccharide, omega-3 fatty acid and mother's milk. Substrates that are not defined as Prebiotics are, for example, protein, fats, probiotics, antibiotics, vitamins, and non-fermentable substrates. Butyrate is a *short-chain fatty acid* produced by bacterial fermentation of fiber in the colon and considered a prebiotic. Animal studies have shown that butyrate and other Prebiotics can improve brain health, reducing depressive-like behavior and elevating levels of BDNF in specific brain regions such as the prefrontal cortex (Bourassa et al., 2016). There are far fewer studies concerning humans with MDD, which supports that Prebiotics improves MMD symptoms (Liang et al., 2018).

The researchers Kazemi, Noorbala, Azam, Eskandari, and Djafarian (2019) investigated in a double-blind *Randomized Controlled Trial* (RCT) study if ingested Prebiotics (galactooligosaccharide) could improve MDD symptoms. The study wanted to test if Prebiotics could stimulate the growth of health beneficial bacterial species in the gut microbiota. The study compared ingested Prebiotics to ingested Probiotics (Lactobacillus helveticus and Bifidobacterium longum) and ingested placebo in people with MDD. The participants were randomly assigned to one of the three groups. 27 in the Prebiotic group, 28 in the Probiotic group, and 26 in the placebo. The study used the 21 items questionnaire *Beck Depression Inventory* (BDI score) to evaluate the participants before the experiment and after eight weeks on their specific treatment. They also measured kynurenine to tryptophan ratio and tryptophan to branch chain amino acid ratios before and after the experiment. Results showed a significant decrease in the probiotic group's BDI score but not for the prebiotic or placebo groups. The probiotic group also showed a substantial reduction in the kynurenine to tryptophan ratio compared to the prebiotic and placebo groups. The study concluded that Prebiotics had no significant effect on the BDI score, kynurenine to tryptophan ratio, or tryptophan to branch-chain amino acid ratios (Kazemi et al., 2019). The article was published in the journal C*linical Nutrition*.

The experiment's most significant strength is using objective and subjective measurements, such as the BDI score from a questionnaire and blood samples. The study measured before and after the given substances. The study uses three groups, each where given different substances. The use of this design and measurement before and after gives more control over confounding variables. This design can be compared to Kelly et al. (2016) and Jacka et al. (2010), which only use one measurement type once. In contrast to Jacka et al. (2010) did Kazemi et al. (2019) test participants with MDD. Participants with MDD is more relevant in making the substance an effective treatment for MDD. Compared to Jacka et al. (2010), tested 1046 participants did Kazemi et al. (2019) only test 27 participants in the Prebiotic group. The relatively few participants may have affected the result.

Kazemi et al. (2019) is a well-done study with several levels to control confounding variables. Compared to Kelly et al. (2016) and Jacka et al. (2010), which both indicates that their method potentially can become an effective treatment for MDD, is the result more conclusive from Kazemi et al. (2019) that prebiotic galactooligosaccharide will not become an effective treatment for MDD. Other types of prebiotic may, in future studies, show more promising results. Still, it is not likely that Prebiotic will become a future treatment for MDD. One of the most convincing reasons for this is the other substance in the study Probiotics showed promising results, and scientists will probably focus on Probiotics over Prebiotics.

Probiotics

According to the research of ingestions of Prebiotics, studies have investigated if the intake of Probiotics can significantly affect MDD symptoms. Kazemi et al. (2019) displayed significant effects of ingested Probiotics for MDD symptoms and kynurenine to tryptophan ratio compared to Prebiotics and placebo. Additionally, have other studies also began to support the idea of Probiotics as an effective future treatment for MDD. Probiotics are defined as live bacteria with health benefits. Most human experimental studies on how gut-microbiota restoration affects MDD have been conducted on Probiotics. The word Probiotic comes from the Greek language and means *for life* (Dinan et al., 2013). The first one to describe a Probiotic was Elie Metchnikoff in 1908. He observed that people in a specific region in Bulgaria lived longer when they regularly drank a fermented milk product with Probiotic bacteria. Some Probiotics have beneficial health effects for people suffering from psychiatric illness and these types of Probiotics are called psychobiotics. Only specific types of Probiotics bacteria have these beneficial health effects and can, for example, help to produce neurotransmitters (Dinan et al., 2013).

In the past five years there has been a great increase in the interest of how Probiotics affect the risk of MDD. Promising results have already begun to emerge from these early studies. There is specific result such as, Probiotics directly affect the tryptophan metabolism, which, as mentioned earlier, is known to affect the risk of MDD, (Desbonnet et al., 2008). Studies have also

demonstrated positive results in different MDD symptoms rating scales. The field is still very new and often, with many limitations, not all studies support these early positive results.

Probiotic studies have investigated different types of Probiotics and used different methods in various studies. Therefore is it difficult to compare existing Probiotics studies concerning MDD. Researchers Huang et al. (2016) investigated in a meta-analysis, including five studies, if Probiotics may have potential preventive significant effects on depression. The meta-analysis compared the five studies and found that participants in the Probiotic group had significantly reduced risks of MDD compared to controls. There were differences in the measurement of MDD (different scales), Probiotics, and doses. One study, including participants over 65 years, showed no significant effects on depression (Huang et al., 2016). In contrast to Huang et al. (2016) did Ng et al. (2018) display no significant effects from Probiotics on MDD in a systematic review, including ten studies. Ng et al. (2018) used the same five studies, as Huang et al. (2016) and five additional studies. They were all double-blind experimental studies. Ng et al. (2018) explained their no significant results in part by differences in the Probiotic dosing and treatment durations, which may impair the comparability between the different studies. They also state that the use of various bacterial strains and strain combinations is also likely to impact trial results because specific bacteria are known to produce superior anti-depressant effects compared to others (Ng et al., 2018).

One of the many limitations in Probiotics studies is often a few participants. However, in contrast to this, did Slykerman et al. (2017) test 423 pregnant women from New Zealand in a double-blind RCT. The study investigated if the ingestion of Probiotic Lactobacillus rhamnosus HN001 could reduce the risk of MDD in pregnant women. The 423 pregnant women were recruited and randomized into a placebo or Probiotic group. They were tested with a modified version of the *Edinburgh Postnatal Depression Scale* (EPDS) to assess MDD symptoms. EPDS is a ten-item screening questionnaire widely used to determine maternal mood. They were tested with EPDS three times, 14-16 weeks of gestation, 6 and 12 months after birth. The women were given treatment from 14 to16 weeks of pregnancy and until six months after given birth. Women in the

Probiotic treatment group had significantly lower EPDS scores than the placebo group (Slykerman et al., 2017). The study was published in the journal *The Lancet*.

In a double-blind RCT study on participants with MDD did Rudzki et al. (2019) demonstrated improvements in cognitive performance after eight weeks of ingestion of Probiotic *LP299v*. Cognitive performance was assessed using five different attention and perceptivity tests, such as the *California Verbal Learning Test* (CVLT), which measures episodic verbal learning and memory and demonstrates sensitivity to a range of clinical conditions. The participants were also tested for their MDD symptoms using the *Hamilton Depression Rating Scale* (HAM-D 17) before and after the experiment. Biochemical parameters such as tryptophan, kynurenine, and kynurenic acid were measured. 30 participants were given an SSRI treatment that included the Probiotic *LP299v,* which were compared to a group with 30 participants that were given a standard SSRI treatment.The Probiotic group did not display any improvement in HAM-D 17 score. However, showed the Probiotic group significantly lower kynurenine levels compared to the SSRI group. The probiotic group displayed improvements in two of the five cognitive performance tests, which one was the CVLT test. The study discussed that the improvement in cognitive performance might be due to the decreased kynurenine levels. Too high levels of kynurenine are known to have neurotoxic and neurodegenerative effects on CNS, but in the right amount, kynurenine is important for immunomodulation and neuroprotection (Rudzki et al., 2019). The study was published in the journal *PsychoNeuroEndocrinology*.

One of Rudzki et al. (2019)'s most significant strengths is the design of an SSRI treatment in both groups. This design can be beneficial for Probiotic to become a complementary treatment for MDD. Considering SSRI today is one of the most common treatments for MDD. This combination of SSRI and probiotics would make it possible for people with MDD to continue with their SSRI treatment and still get the benefits of Probiotics. Rudzki et al. (2019) is in concordance with Kazemi et al. (2019) a well-done study with several levels to control confounding variables. For example, uses Rudzki et al. (2019) measurement before and after the experiment and subjective and objective measurement. The most critical finding of Rudzki et al. (2019) is, once again in concordance with

other studies, the alteration in tryptophan metabolism. In contrast to Kazemi et al. (2019), did Rudzki et al. (2019) Probiotics group not display any improvement in their MDD symptoms rating scale. The studies used different scales and different Probiotics, which, in part, can have affected the results. The Probiotic group showed only improvements in two of the five criteria for cognitive performance tests. Compared to Slykerman et al. (2017), tested 423 participants did Rudzki et al. (2019) tested 30 participants. These relatively few participants could potentially be a factor in the result. Rudzki et al. (2019) are an excellent example of how new Probiotic research concerning MDD is and its limitations. However, several limitations are Rudzki et al. (2019), one of the first to demonstrate connections between the ingestion of probiotics in people with MDD and improvements in cognitive performance, combined with gut-microbiota restoration.

Studies in this section indicate that gut-microbiota restoration may be useful as a treatment for MDD. Comparing the different methods shows that most research has been done in Probiotics and the most promising results are also from these studies. Despite these promising results, there are still many limitations in Probiotic research and many more studies are needed to improve the comparability between the different studies and CBT and SSRI studies.

Discussion

This thesis aimed to investigate if there is a connection between gut-microbiota disruption and MDD and if gut-microbiota restoration can be a potential effective future treatment for MDD. The thesis found specific main findings, such as people with MDD often display alteration in their gut-microbiota composition combined with chronic low-grade inflammation (Mass et al., 2008). Additionally, found the thesis that chronic stress can disrupt the intestinal barrier, making it leaky and increasing bacterial cell wall components such as lipopolysaccharide. Stress is one of the factors that can cause low-grade inflammation (Bailey & Coe, 1999). Furthermore, display studies in the thesis that alteration in the gut-microbiota and low-grade inflammation can affect crucial pathways in the gut-brain axis, such as the HPA axis. Studies indicate that HPA axis disruption can cause some specific brain change often displayed in people with MDD. One of the most important

findings was that tryptophan metabolism was shown in different studies to affect several gut-brain axis pathways, such as neurotransmitter systems, inflammation and the HPA axis.

Much of the research in the gut-brain axis is done in animal studies. Therefore, the studies concerning inflammation are critical because studies have investigated people with MDD and not only animals. The cytokine hypothesis claims that MDD results from increased levels of pro-inflammatory cytokines and the reduction of anti-inflammatory cytokines. The excessive pro-inflammatory cytokines are discussed to affect the HPA axis, increase the blood-brain barrier's permeability and reduce the synthesis of 5-HT (Mass et al., 2008). Studies in the thesis indicate that the tryptophan metabolism is crucial for balance in all the above systems and therefore, a potential target in future treatments aiming to restore this balance. All these factors are essential because it supports a holistic view of what causes MDD. This view explains MDD as a cyclic process in which dysfunction in one system, such as the gut-microbiota, can lead to a knock-on effect in other systems, leading to specific brain change.

The second question investigated if gut-microbiota restoration can be an effective future treatment for MDD. Compared to traditional treatments such as SSRI and CBT have promising complementary treatments such as probiotics still much to prove before becoming an effective treatment for MDD. One of the problems with these new treatments is that few human studies and much research are based on animal studies. Kazemi et al. (2019) demonstrate in their prebiotic study that animal studies' results can not always be replicated in human studies. Review studies such as Ng et al. (2018) and Huang et al. (2016) display how difficult it is to compare different Probiotics studies' effectiveness. The different studies often use different designs both between themself and compared to SSRI and CBT studies. This difference in design makes it difficult to conclude how effective new treatments targeting the gut-brain axis would be. Probiotics studies such as Kazemi et al. (2019) and Rudzki et al. (2019) test the effectiveness of MDD over eight weeks. This time is relatively short, considering MDD known to be long-lasting and sometimes life long. CBT and SSRI treatments have been shown to be effective over a long time (Roth & Fonagy, 2006).

The research area is very complex and still emerging. Research on the gut-brain axis in MDD is much like a puzzle where there are very complex studies from different research fields, on particular aspects of the research area, often in animal studies. The results of these different studies are then pieced together in review articles, which are interpreted in different ways. The thesis found that most support for how the gut-brain axis affects MDD is in review articles. These review articles claim some time that RCT studies have used certain methods to support specific results. For example, Liang et al. (2018) claim that four methods (Prebiotics, probiotics, FMT and healthy diet) are useful for gut-microbiota restoration. When following the articles' references, it is hard to support this claim for more than maybe probiotics. Liang et al. (2018) is an excellent example of how new and speculative the research area sometime is (Liang et al., 2018).

Because of the many limitations from studies on the gut-brain axis concerning MDD, it is not likely that treatments such as Probiotics will become a new treatment for MDD on its own soon. In contrast to this, many limitations indicate studies such as Rudzki et al. (2019) that probiotics combined with SSRI may have a significant effect as a complementary treatment for MDD. If more studies can confirm, results such as those from Rudzki et al. (2019), Probiotics and SSRI combinations be useful for people who now have no or little effects from SSRI or CBT treatments today. The research on the gut-brain axis is still in a very early stage and it will probably take a relatively long time before this research turn into a treatment for MDD. On the contrary, this research already began to better the understanding of the mechanisms behind MDD. Results may also contribute to an additional objective method to measure the risk of MDD by measuring tryptophan metabolism and inflammatory cytokines.

Future research should try to set a standard, for example, to test the effects of probiotics in MDD and do follow-up studies over long periods. It would be interesting to see studies that combined Probiotics and fMRI or MRI with seeing if there is a connection with probiotics and brain change in participants with MDD.

Conclusion

This literature review concludes that there is support for a connection between the disruption of our gut-microbiota and MDD. The most important finding for this connection is the low-grade inflammation and disruption of gut-microbiota that several studies have displayed in people with MDD. Inflammation can cause HPA axis dysfunction, leading to the specific brain dysfunction often displayed in areas such as the hippocampus, amygdala and pituitary gland, which are influenced by the HPA axis. Furthermore, studies show that tryptophan metabolism is involved in inflammation and HPA axis dysfunction and is therefore used as a target in studies for new treatments for MDD. The thesis found that the research field of gut-brain treatments for MDD is very new and still emerging. Therefore, few studies have many limitations to support that gut-brain treatment can be effective for MDD. Gut-brain treatments such as Probiotics are compared with traditional treatments such as SSRI and CBT, not likely to soon become a treatment for MDD. It is more likely that the research on the gut-brain axis concerning MDD will be used to better the understanding of the causes of MDD and develop new measurements for MDD.

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