Review

Mendelian randomisation: a potential tool to resolve the age-long gut microbiome–depression conundrum

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Introduction

Depression affects about 350 million people and is a leading cause of disability and mortality worldwide.[1] Antidepressants work in about 50–75% of patients and may have undesirable side effects, constituting a major public health challenge.[2,3] This inefficacy of treatment may be due to the current lack of understanding of the pathogenesis of depression. The leading hypotheses for the cause of depression are: neurotransmitter imbalance,[4] and dysfunctions of brain neuroplasticity,[5] the hypothalamus–pituitary–adrenal (HPA) axis[6] or the immune system.[7] However, these hypotheses are not universally accepted.[8]

The gut microbiome is believed to influence the functions of both the gut and the brain by modulating the host's neural, immune, and endocrine pathways. Also, there is a consensus of an alteration in the abundance and distribution of gut microbes in depressed subjects.[9–11] This suggests that the development of therapeutics that perturb the gut microbiome could be the solution to the current challenge of treating depression. Hence, a role of the gut microbiome in the pathogenesis of depression is now a major focus of research and public interest, and this is enabled by advances in sequencing technology. This review aims to discuss the existing evidence for a causal relationship between the gut microbiome in depression and proposes Mendelian randomisation (MR) as a new paradigm to investigate this relationship.

The gut microbiome–depression hypotheses

Hypothesis 1: a change in the gut microbiome causes depression
Rodent studies of faecal microbiome transplantation, germ-free mice, and vagotomy indicate that the gut microbiome influences brain physiology and, thus, mental health, directly through activation of the vagus nerve, and indirectly through the production of microbial metabolites that ultimately enter the brain.[12–21]

Hypothesis 2: depression causes a change in the gut microbiome
Studies that used olfactory bulbectomy and stress models to induce depression-like behaviours in rodents have reported that depression influences the HPA axis and immune responses, resulting in a change in the gut microbiome.[22–25] Mice exposed to chronic social defeat, used as a stress model, demonstrated transiently elevated levels of IL-10^{+} T regulatory cells, which preceded reduced diversity and reduced microbial richness in the mice.[25] Mice that underwent olfactory bulbectomy have been reported to exhibit increased HPA axis activity, significantly greater colonic motility, and altered microbiome composition. It is believed that activation of the HPA axis increases colonic motility, leading to changes in the gut microbiome.[23]

Limitations of the current evidence of the gut microbiome–depression relationship

The hypothesis that a change in the gut microbiome causes depression and not vice versa may be significant in the development of effective therapeutics for depression. However, the evidence for the gut microbiome–depression hypotheses mentioned above has come from animal studies. Unfortunately, translating findings from animal models into effective therapeutics in humans frequently fails due to inter-species differences in biochemistry, behaviour and physiology.[26,27]

Human studies have shown correlations between specific gut bacteria, their metabolites and depression,[15,28–31] but these have all been observational studies that do not infer causality, and can be affected by unmeasured confounding factors.[32,33] A randomised controlled trial (RCT) is the gold standard design to infer causation between two variables,[34] and RCTs involving faecal mass transplantation may provide an answer as to whether there is a causal role of the gut microbiome in depression in humans. However, RCTs can be expensive, time-consuming and impractical.[35] Hence, MR is proposed as a novel tool to detect a causal effect of the gut microbiome in depression in humans.

MR as a tool to investigate causality

MR is a method that uses genetic variants as instrumental variables to investigate the causal relationship between modifiable risk factors and health outcomes in observational data.[36] MR is comparable to RCTs because genetic alleles are randomly assorted and fixed at conception, making the MR method less likely to be affected by confounding or reverse causation than conventional observational studies.[37] Various studies have shown that it is possible to detect variants in the human genome that influence the composition of the gut microbiome.[38–40] Using these genetic variants, the MR approach can be used to identify whether any gut bacterial species has a causal effect on depression.[41] Some authors have reported that environmental factors play a bigger role than host genetics in the host's microbiome composition.[42] However, MR could still be used to investigate the gut microbiome, as demonstrated by recent studies investigating a causal relationship between the gut microbiome and metabolic diseases.[41,43]

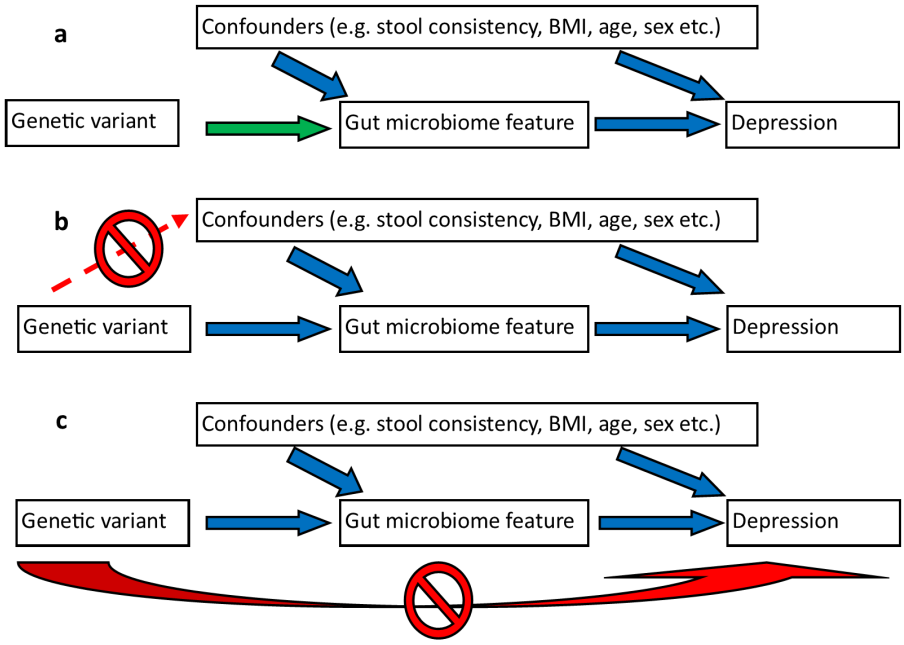


Figure 1. An illustration of the MR method and its assumptions for investigating a causal relationship between the gut microbiome and depression. (a) Assumption 1: the genetic variant must be reproducible and directly associated with the gut microbiome feature. **(b)** Assumption 2: the genetic variant must not be associated with confounders. **(c)** Assumption 3: the genetic variant should only be associated with depression through the gut microbiome feature. Figure adapted with permission from Sekula et al.[35]

A proposed MR study to investigate the gut microbiome–depression hypothesis

As a proposed MR study, genome-wide genetic data, gut metagenomic sequencing, measurements of faecal microbiome-dependent metabolites, and depression status could be collected from cohorts such as the Belgian Flemish Gut Flora Project (FGFP) and the Dutch Lifelines DEEP (LLD) cohorts.[28] Once the microbiome features (metabolites, pathways, and unique taxa) associated with depression are established, data from genome-wide association studies (GWAS) from the LLD and FGFP projects could be used to identify the independent genetic variants that are associated with each depression-related microbiome feature.

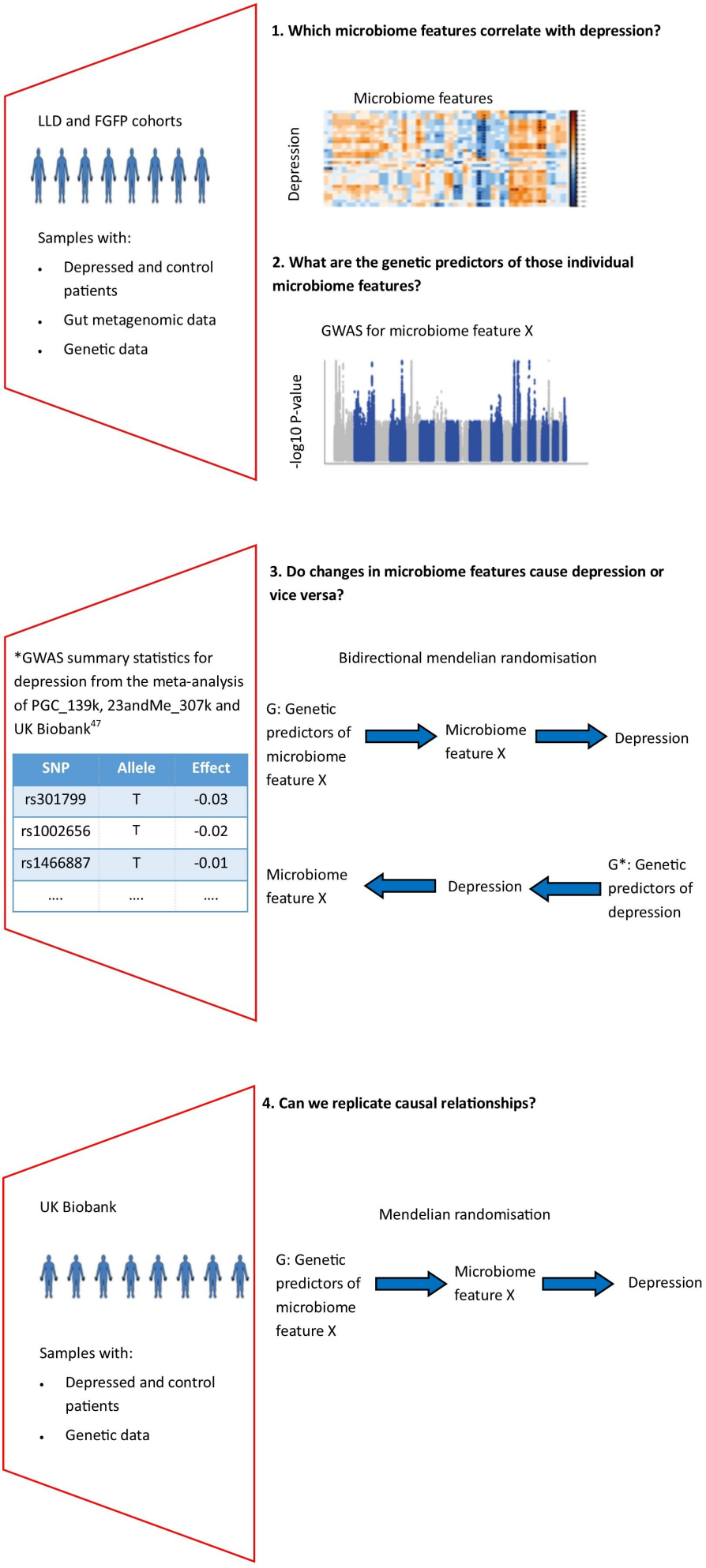
The MR approach must satisfy three assumptions **(Figure 1)**: firstly, that the genetic variant selected as the instrumental variable is strongly associated with the gut microbiome feature; secondly, that the genetic variant is not associated with any unmeasured confounders of the gut microbiome feature and depression relationship; and thirdly, that the genetic variant is associated with depression only through gut microbiome features, not through other pathways.[32,37] However, it is difficult to ascertain that none of the MR assumptions are violated. Thus, any findings of a causal relationship in the proposed study cohorts (LLD and FGFP) should be validated in an independent cohort, such as the UK Biobank.[41] Previous articles have provided details on how to conduct MR and assess the plausibility of MR assumptions.[32,35–37]

Although the MR approach as described above could provide inference of a causal relationship between the gut microbiome and depression, it does not distinguish the directionality of a causal association. Bidirectional MR is an approach that could be used to determine the directionality of a causal association by investigating the hypotheses that depression causes a change in the host's gut microbiome and vice versa.[32,36] Three recent GWAS have reported reproducible genetic variants for depression.[44–46] These studies culminated in a genome-wide meta-analysis of depression which reported 102 associated variants that were replicated in an independent sample.[47] The variants had an equivalent direction of allelic effect across the three studies that contributed to the meta-analysis, suggesting a robust association between the genetic variants and depression.[47] Hence, if MR assumptions are met, MR could be used to investigate the hypothesis that depression causes a change in the gut microbiome. **Figure 2** summarises a proposed MR study that could be used to investigate a causal role for the gut microbiome in depression.

Conclusion

Currently, there is no consensus on the pathogenesis of depression. Advances in sequencing technology have enabled large observational studies, which implicate the gut microbiome in the pathogenesis of depression. However, conventional observational studies do not infer causality and are affected by confounding factors. MR analysis of observational data reduces the likelihood of data analysis being affected by confounding factors and reverse causation. If MR analysis reports that the imbalance of specific gut bacteria and their metabolites causes depression, this could lead to the development of effective therapeutics targeting the gut microbiome, helping to overcome the major challenges faced in treating depression.

Figure 2. Schematic representation of a proposed MR study to test for a causal relationship between the gut microbiome and depression. Microbiome features refer to metabolites, pathways, and unique taxa. Figure adapted by permission from Springer Nature, from Sanna et al.[41]



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