Dysbiosis is not an answer

Dysbiosis is not an answer

Scott W. Olesen and Eric J. Alm

Dysbiosis, an imbalance in the microbiota, has been a major organizing concept in microbiome science and medicine. Here, we discuss how the balance concept, a holdover from prescientific thought, is irrelevant to—and may even distract from—useful research about the microbiome.

Throughout most of Western medical history, it was believed that the digestion of foods produced humors: mostly invisible liquids that were transformed into the materials that make up the body. Humoral theory dictated that diseases were caused by imbalances in these humors. A doctor's role was to help the body correct this imbalance, either by altering a patient's diet—thus changing the intake of humors—or by expelling excess humors, for example, by bloodletting[1].

Now, with modern DNA sequencing technology, we know our digestive systems are actually filled with mostly invisible microorganisms that respond to our diet. We have found that altering diet, ingesting probiotics, and wholesale replacement of the microbial community can improve our health. A common explanation for the effectiveness of these therapies is that they ameliorate "dysbiosis", an imbalance in the microbiota[2-4].

The word "imbalance" is crucial, because it implies a dysfunction of some complex set of processes, that is, that dysbiosis causes disease. This is not implausible; there is accumulating evidence of how imbalances in the microbial ecology of the gut or in the host immune system could cause disease. However, at the moment, the concept of balance is supported by evidence from only a few model systems and model interactions.

In terms of microbial ecology, dysbiosis can be conceptualized as a disruption in the many potential ecological links between microorganisms: competition, inhibition, commensalism, and perhaps symbiosis. In theory, a shift away from the "healthy" ratios of species in the microbiota could lead to a disrupted ecosystem that harms the host. This explanation, though theoretically sound and intellectually engrossing, ignores the wide gap between what we actually know about the microbiome and how we suspect it may be. In general, the signal of microbe-microbe interactions in the human gut is not strong. Only a small fraction of taxa in the gut are correlated[5], and there are only a few examples of known ecological relationships among microbiota constituents (e.g., ref. 6). There are few or no examples of instances in which breakdowns in ecological relationships between gut microorganisms are proven to affect host health. We may eventually discover a vast network of strong ecological interactions, but we may also discover that such a network does *not* exist. So far the data have not provided unequivocal support for either scenario.

Dysbiosis can also be tied to the immune system's need to balance tolerance, which prevents attacks against the host's own tissues and its healthy commensal microorganisms, against inflammation, which protects against infection. For example, Lee and Mazmazian[7] explain how an imbalance between the abundance of pro-inflammatory segmented filamentous bacteria (SFB), which causes the generation of Th17 cells, and the abundance of anti-inflammatory *B. fragilis*, which causes the generation of Treg cells, can lead to disease. This is a compelling argument, and it may be that many microbial species beyond *B. fragilis* and SFB are "balanced" in this way. However, only a small number of model systems have been studied, and it is premature to assert that balance between *any* particular microbial species is important to host health. Notably, the relevance of SFB to the human microbiome is still unclear[8,9].

Given that the possible mechanisms by which dysbiosis could cause disease are still under investigation and that the relevance of most microbiota compositions to disease remains speculative, we believe researchers may be using the word "dysbiosis" to refer to the result of disease, rather than the cause. Indeed, dysbiosis has such varying definitions in the literature that the term could apply to either cause or effect. The most common definition of dysbiosis is an imbalance between beneficial microorganisms like *Lactobacillus* or *Bifidobacterium* and harmful microorganisms like *Escherichia coli*[2-4]. However, the range of definitions is broad enough to capture any difference in microbial composition: dysbiosis has been defined as a change in the abundance or diversity of some groups of microorganisms[10], a change in community composition caused by lifestyle[7], a microbiota composition in which harmful organisms outweigh beneficial ones[11], a bloom of a single pathobiont[12], and as a reduction in bifidobacteria[13].

This ambiguity in definition means that any measured difference in microbial composition, whether the cause or effect of disease, can be called dysbiosis. It is a "mechanism-free" cause of disease to which we can retreat when plausible mechanistic explanations are discounted. For example, although there has been suspicion that a single, unknown pathogenic species causes inflammatory bowel disease (IBD), an autoimmune disorder characterized by chronic inflammation of the parts of the digestive system[2], such a microorganism has not been found. Now most discussion about the role of the microorganisms in IBD focuses on dysbiosis.

The slipperiness of the dysbiosis concept means that a study can claim a "positive" result—that dysbiosis was discovered—without actually accomplishing anything useful. For example, if you intend to design a diagnostic using microbiome data, then profiling the microbiome of healthy and ill people is not sensible because a diagnostic would not be needed to determine health status. A questionnaire is usually sufficient for this. Microbiome data can still add value to diagnostics however. For example, it is easy to determine that someone *might* have IBD based on their symptoms. The more difficult challenge is to determine if someone who appears to have IBD is suffering from IBD or from a disease with related symptoms such as irritable bowel syndrome (IBS). Thus, designing a diagnostic requires comparing people who have IBD and people who have diseases that might be mistaken for IBD.

A microbiome diagnostic must also add value beyond other diagnostics. For example, fecal calprotectin, a biochemical assay on patient stool, can help diagnose IBD. If microbiome data has no explanatory power beyond fecal calprotectin measurements, then a microbiome-based diagnostic is essentially an expensive and complicated way to estimate fecal calprotectin.

Like IBD, obesity is likely related to the gut microbiome. Microbiome diagnostics for obesity make no sense when we can just use a weighing scale. The more important possibility, of course, is that the microbiome *causes* obesity. Microbiome surveys comparing healthy and ill subjects are very valuable for addressing the causality of a disease, but only insofar as those surveys are connected with experiments that can verify causality. For example, if an organism is depleted in the microbiome of ill subjects, that organism could be re-introduced as a probiotic[14] or in a gnotobiotic animal to test its effect on disease state, host immunity, or gut function. Even if it remains unclear whether the depletion of a beneficial microorganism interferes with host health, and even if the mechanism by which the addition of a microorganism affects health remains unclear, a simple experiment can show that the addition of the microorganism improves host health. Causality without mechanism is still important: a therapeutic requires causality, not necessarily mechanism.

Developing diagnostics and addressing causality are difficult problems (indeed, even the causative role of the microbiome in obesity remains debated)[15]. Studies designed to do either of these things can have a high probability of failure. However, negative results about diagnostics or therapeutics are more useful than positive results about dysbiosis. We already have two thousand years of tradition supporting the idea that balance is related to health, and the fact that healthy and ill people have different microbiomes is no longer a novel or useful observation. We need to show that differences in the microbiota can be used to predict or ameliorate disease, not just show that differences exist.

References

1. Magner, L. N. *A history of medicine*. (CRC Press, 2005).

2. Tamboli, C. P., Neut, C., Desreumaux, P. & Colombel, J. F. Dysbiosis in inflammatory bowel disease. *Gut* **53,** 1–4 (2004).

3. Bäumler, A. J. & Sperandio, V. Interactions between the microbiota and pathogenic bacteria in the gut. *Nature* **535,** 85–93 (2016).

4. Mazmanian, S. K., Round, J. L. & Kasper, D. L. A microbial symbiosis factor prevents intestinal inflammatory disease. *Nature* **453,** 620–625 (2008).

5. Friedman, J. & Alm, E. J. Inferring correlation networks from genomic survey data. *PLoS Comput Biol* **8,** e1002687 (2012).

6. Fischbach, M. A. & Sonnenburg, J. L. Eating for two: How metabolism establishes interspecies interactions in the gut. *Cell Host Microbe* **10,** 336–347 (2011).

7. Lee, Y. K. & Mazmanian, S. K. Has the microbiota played a critical role in the evolution of the adaptive immune system? *Science* **330,** 1768–1773 (2010).

8. Sczesnak, A. *et al.* The genome of Th17 cell-inducing segmented filamentous bacteria reveals extensive auxotrophy and adaptations to the intestinal environment. *Cell Host Microbe* **10,** 260–272 (2011).

9. Caselli, M., Tosini, D., Gafà, R., Gasbarrini, A. & Lanza, G. Segmented filamentous bacteria-like organisms in histological slides of ileo-cecal valves in patients with ulcerative colitis. *Am J Gastroenterol* **108,** 860–861 (2013).

10. Henao-Mejia, J. *et al.* Inflammasome-mediated dysbiosis regulates progression of nAFLD and obesity. *Nature* **482,** 179–185 (2012).

11. Roberfroid, M. *et al.* Prebiotic effects: Metabolic and health benefits. *Br J Nutr* **104,** S1–S63 (2010).

12. Devkota, S. *et al.* Dietary-fat-induced taurocholic acid promotes pathobiont expansion and colitis in Il10-/- mice. *Nature* **487,** 104–108 (2012).

13. Hopkins, M. J., Sharp, R. & Macfarlane, G. T. Age and disease related changes in intestinal bacterial populations assessed by cell culture, 16S rRNA abundance, and community cellular fatty acid profiles. *Gut* **48,** 198–205 (2001).

14. Sokol, H. *et al. Faecalibacterium prausnitzii* is an anti-inflammatory commensal bacterium identified by gut microbiota analysis of Crohn disease patients. *Proc Natl Acad Sci USA* **105,** 16731–16736 (2008).

15. Harley, I. T. & Karp, C. L. Obesity and the gut microbiome: Striving for causality. *Mol Metab* **1,** 21–31 (2012).