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Gut Health Explained

Course Guidebook

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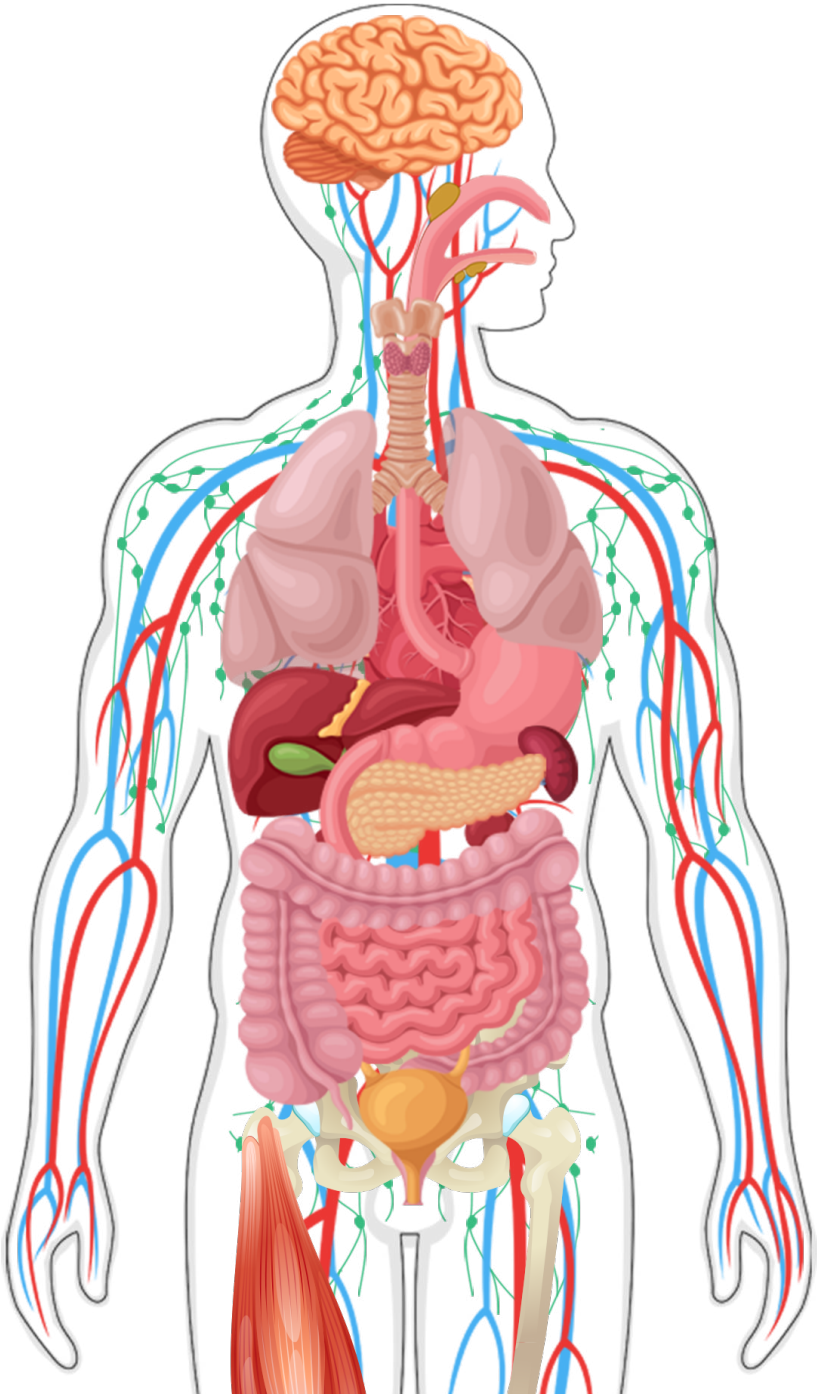


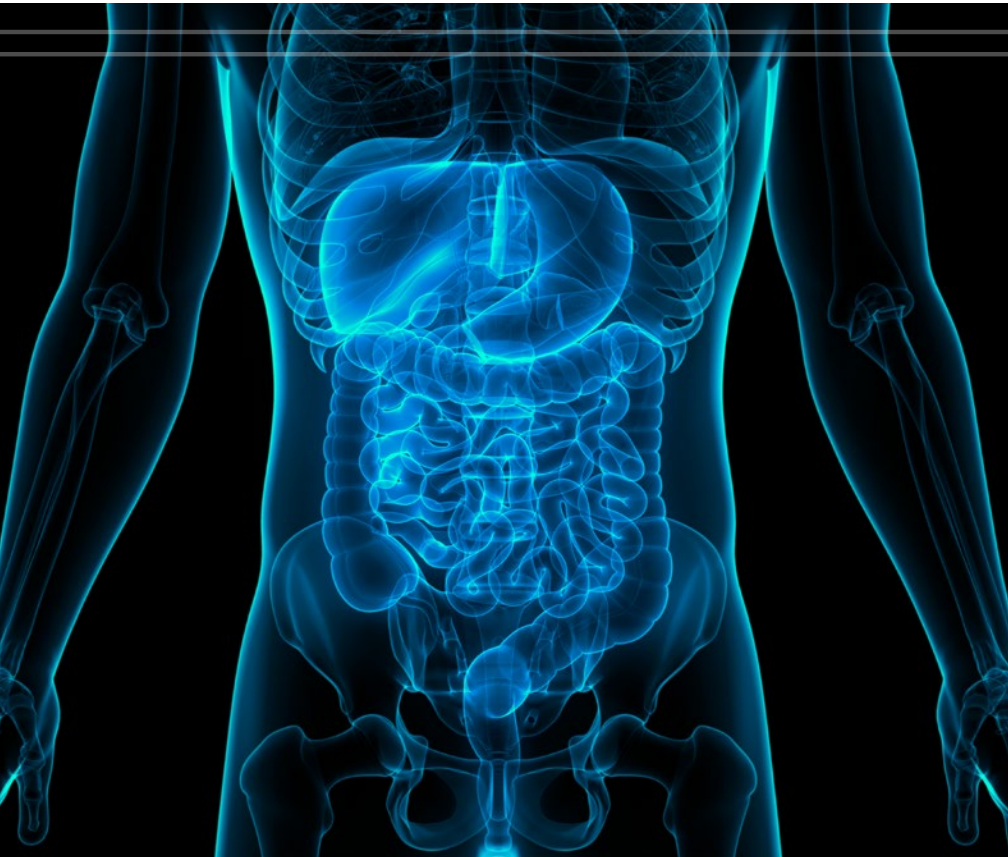
What Is Gut Health?

1

Inside your body right now, a community of microscopic organisms is living, reproducing, changing, and communicating with the rest of your body. This population of microbes that resides along the length of your digestive tract is called the gut microbiome. Within the last 20 years, researchers have found that, from digestion and metabolism to immune defense and even brain activity, these microbes play integral roles in human development, health, and disease from birth until death. As you'll learn throughout this course, the field of gut microbiome science is rife with opportunity, excitement, and hope for new ways to predict, diagnose, and even cure diseases—but it's also fertile soil for pseudoscience, exaggeration, extrapolation, and unscrupulous marketing.

Gut Health

What is gut health? This is one of the most important questions in microbiome science. Many researchers believe it may be impossible to define a healthy gut microbiome given the dizzying number of factors that influence the microbial community. This makes "healthy" too subjective to be meaningful. However, you can still talk about gut health in a meaningful way because, over the last decade, researchers have collected more evidence to support the idea that the gut microbiome plays an important role in both health and disease. Simultaneously, it's important to clear up some of the poorly defined ideas that have clouded this conversation and often led to exaggeration, misunderstanding, and the dissemination of flat-out falsehoods.



Gut microbiome publications are plagued by the use of poorly defined terms to explain their findings, which could threaten the progress of gut microbiome science. For example, when researchers describe the gut microbiome using terms like *balanced* or *imbalanced* and *eubiosis* or *dysbiosis*, they're perpetuating old and somewhat unscientific beliefs about how the human body works. These kinds of broad, catchall explanations for how the gut microbiome works have a tendency to gain popularity.

Take the word *imbalance*. It's a poorly defined term that nonetheless feels like it could explain all kinds of ailments. And it's a brilliant marketing tool: "If only there were a product that could fix the imbalance of my gut microbiome, then I could be healthier." Without a way to define a healthy gut microbiome, you have no way to determine what types of changes might be unhealthy in comparison or—importantly—why they're unhealthy. So, when influencers and popular media use the term *gut health*, they're building on the ambiguity of these definitions. They also tend to assume that changes in the microbiome can cause specific health outcomes (for example, introduce a microbe or prevent a disease) even though the evidence doesn't support this assumption. Keeping this in mind, a more satisfying answer has been developed, which you can call the three *D*s of gut health.

The Three *D*s

The first *D* stands for disease. Here, this refers to the presence or absence of disease in the digestive tract. About 40% of people globally are suffering from a gastrointestinal (GI) disease.

The second of the three *D*s is digestion. This encompasses both the subjective experience of digestion, like nausea, bloating, gas, and stool quality, and the objective ability of your GI tract to effectively break down and absorb nutrients. For example, you might feel some gas and bloating after eating certain foods (subjective experience), but your intestinal tract is functioning normally (objective ability).

The third *D* is diversity, or the variety and proportions of microbes that inhabit the digestive tract. Diversity is often used to describe the richness of microbial species in the gut—that is, the number of different types of microbes—as well as the evenness, or the proportions, of microbes and how they're distributed. Diversity can also measure the variety of genes these microbes have. The number and proportions of the microbes, or taxonomic diversity, tells you only one part of the story. You need to know the functional diversity, or the variety of genes, to know what those microbes are capable of doing.

Functional and Organic Diseases

How can researchers evaluate—or even establish—the complete well-being of an entire organ system populated by trillions of living organisms? There isn't a clear answer to that question yet, but it might be easiest to start by determining the presence or absence of a GI disease and how it affects digestion. GI diseases can be organized generally into two categories: functional and organic.

A functional disease only affects the way the GI tract works, without any damage to the tissues. Irritable bowel syndrome (IBS) is a common example. Although the intestinal tract looks healthy, it doesn't function properly, so people with IBS will still experience some uncomfortable symptoms like abdominal pain and irregular bowel habits. Meanwhile, inflammatory bowel disease (IBD) is an organic disease because it affects both the tissues and the function of the GI tract. IBD can cause ulcers to form in the intestinal lining, which impair nutrient absorption and can put a person's life at risk if an infection forms.

Some diseases, like infectious diarrhea, can be cured, but others, like IBS or IBD, are incurable and need to be managed. GI diseases can be managed in a number of ways. Lifestyle changes like exercising regularly and getting adequate sleep can help manage some symptoms of GI diseases, while in other cases, prescription medications are required.

"Balanced" and "Imbalanced"

The digestion aspect of gut health is obviously linked to the disease aspect, and both digestion and disease are related to microbiome diversity in ways researchers are just beginning to understand. Unlike digestion and disease, a "healthy" level of diversity can't be defined yet. The idea that a healthy microbiome is a balanced one is based on ancient beliefs that have since been debunked.

In ancient Greece, doctors thought that illness was caused by an imbalance of the four humors in the body: blood, phlegm, black bile, and yellow bile. They believed that digestion produced these humors and that it was the physician's job to correct imbalances. Today, when it comes to the gut microbiome, *balance* is still a buzzword.

Without any real parameters to evaluate the healthiness of someone's microbiome, many researchers and media outlets use subjective terms like *balanced* or *imbalanced*, which imply health or dysfunction, respectively. These ideas have led to the assumption that a healthy person will also have a healthy gut microbiome, so whatever the composition is, it must be balanced, or in a state of eubiosis. A person with a disease, however, must have an

imbalanced microbiome, or dysbiosis. In other words, a healthy person has the "right" amount of microbial diversity, while an unhealthy person has the "wrong" amount.

To assert that someone's microbiome is actually imbalanced, you need some sort of reference for what you'd expect to see in a balanced community. But just because one healthy person's microbiome looks a certain way, you can't assume that all microbiomes should look that way. Plenty of healthy people have vastly different microbiomes. What researchers have actually observed is simply a difference in microbial communities, and they don't know enough yet to say that it's a "bad" difference.

Benefits of Microbial Diversity

You may have seen recent gut health tests claiming to provide information about your level of dysbiosis, with reference ranges to indicate whether microbes are higher or lower than they "should" be. Unfortunately, far from being based on clinically validated data, these reference ranges are based on the cohorts, or the people who were chosen by the test's development team. So, you could be labeled as having dysbiosis because your microbiome differed from the microbiome of a healthy person, without any real scientific evidence to support the notion that you're the one with dysbiosis and not the healthy person used to develop the reference ranges.

However, even if researchers don't know what microbes or proportions lead to better health, they can still get an inkling, from experimental data, about how microbes interact with one another in ways that could affect their human hosts. Fecal transplants in rodents, for example, have revealed a relationship between body weight and certain microbes that cohabitate the gut.

Researchers Buck S. Samuel and Jeffrey I. Gordon, along with colleagues, wanted to know the digestive impact of a particular archaeon commonly found in our colon, *Methanobrevibacter smithii*. Using fecal transplant, they gave rodents another microbe that forages the polysaccharides in our diet, *Bacteroides thetaiotaomicron*, which they introduced with or without the microbe of interest, *M. smithii*. They found that animals who were co-colonized with both of these microbes exhibited significant increases in

weight gain and fatty tissue—compared to animals colonized with just *B. thetaiotaomicron* alone or those co-colonized with a microbe that was not *M. smithii*. Although *M. smithii* might seem insignificant based on its extremely low abundance relative to other microbes, it makes the process of microbial fermentation more efficient by recycling the end product, hydrogen, to create methane.

However, while studies like these are helpful for identifying certain community dynamics or cell signaling pathways, it's still unclear exactly how changes in gut microbiome diversity translate to changes in the health or disease of the host. This study and others like it confirm that both taxonomic and functional diversity need to be considered to understand how the microbiome might affect your health. Researchers need to look at both the variety of microbes and what they're actually doing—how they interact with each other and the host. The hope is that future studies will be able to establish clinical markers for disease as it relates to the gut microbiome.

Researchers have also begun to identify relationships between microbial diversity and healthy lifestyle habits like exercising and eating a prudent diet. While the mechanisms are still unclear, there's enough consistent evidence to notice some patterns. For example, several observational studies have found that long-term dietary patterns have a more significant and lasting impact on the gut microbiome than short-term interventions. These long-term dietary patterns also happen to modify your risk of certain diseases. A Mediterranean diet could reduce your risk of colorectal cancer by about 20%, and it's also associated with a more diverse microbiome compared to a standard American diet.

Exercise plays a role, too. If you're physically fit and exercise regularly, you'll probably have a more diverse microbiome than a sedentary person, with specific enrichment of microbes that produce beneficial short-chain fatty acids (SCFAs). Some elite athletes have unusually high levels of microbes that specialize in converting metabolic leftovers into useful energy sources for their human hosts, for example. It's still unclear whether these microbes contribute directly to exercise performance or health, but some research suggests that dietary patterns can dictate whether an active person's microbiome will be more diverse than that of a sedentary person.

So, the take-home message is simply to try eating a prudent diet while exercising regularly to maximize the potential benefits of either habit. Researchers are not yet able to quantify how much or what kind of diversity we need to optimize health. But the appropriate amount—whatever that may be for a person—creates a resilient microbial ecosystem that fulfills all of its functions, like immune defense and energy harvesting, and bounces back from perturbations, like antibiotics or infections.

READING

Durk, R. P., E. Castillo, L. Márquez-Magaña, G. J. Grosicki, N. D. Bolter, C. M. Lee, and J. R. Bagley. "Gut Microbiota Composition Is Related to Cardiorespiratory Fitness in Healthy Young Adults." *International Journal of Sport Nutrition and Exercise Metabolism* 29, no. 3 (2019): 249–253. https://doi.org/10.1123/ijsnem.2018-0024.

Garcia-Larsen, V., V. Morton, T. Norat, A. Moreira, J. F. Potts, T. Reeves, and I. Bakolis. "Dietary Patterns Derived from Principal Component Analysis (PCA) and Risk of Colorectal Cancer: A Systematic Review and Meta-Analysis." *European Journal of Clinical Nutrition* 73, no. 3 (2019): 366–386. https://doi.org/10.1038/s41430-018-0234-7.

Hooks, K. B., M. A. O'Malley. "Dysbiosis and Its Discontents." *mBio* 8, no. 5 (2017). https://doi.org/10.1128/mBio.01492-17.

Jang, L. G., G. Choi, S. W. Kim, B. Y. Kim, S. Lee, and H. Park. "The Combination of Sport and Sport-Specific Diet Is Associated with Characteristics of Gut Microbiota: An Observational Study." *Journal of the International Society of Sports Nutrition* 16, no. 1 (2019): 21. https://doi.org/10.1186/s12970-019-0290-y.

Loughman, A., and H. M. Staudacher. "Treating the Individual with Diet: Is Gut Microbiome Testing the Answer?" *The Lancet Gastroenterology & Hepatology* 5, no. 5 (2020): 437. https://doi.org/10.1016/S2468-1253(20)30023-6.

McBurney, M. I., C. Davis, C. M. Fraser, B. O. Schneeman, C. Huttenhower, K. Verbeke, J. Walter, and M. E. Latulippe. "Establishing What Constitutes a Healthy Human Gut Microbiome: State of the Science, Regulatory Considerations, and Future Directions." *Journal of Nutrition* 149, no. 11 (2019): 1882–1895. https://doi.org/10.1093/jn/nxz154.

Olesen, S. W., and E. J. Alm. "Dysbiosis Is Not an Answer." *Nature Microbiology* 1 (2016): 1–2. https://doi.org/10.1038/nmicrobiol.2016.228.

2

Introducing the Gut Microbiome

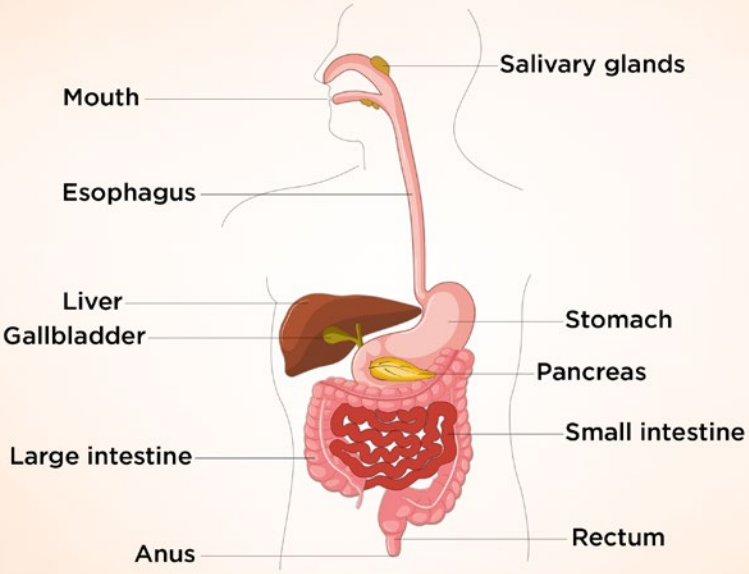
Your cardiovascular, immune, nervous, and endocrine systems are all influenced by your gut microbes. The microbiota can produce useful compounds that nourish intestinal cells, support metabolic health, protect against certain types of cancer, modify the immune system, and even influence brain activity. Gut microbes can also harvest energy from the diet by converting indigestible fibers into fatty acids that your cells can utilize. In this lecture, you'll get to know your gut microbes a bit better. You'll start by exploring the digestive tract to see how this landscape shapes the microbial community. Then, you'll learn how you acquired your personal set of microbes and the many factors that create a microbiome as unique as your fingerprints.

The Digestive Tract

Gut is a term used to describe the GI or digestive tract, which includes the mouth, esophagus, stomach, small and large intestines, rectum, and anus. The liver, gallbladder, pancreas, and salivary glands are considered accessory organs. The variability of the pH, anatomy, oxygen, and nutrient availability along the GI tract provides a variety of microhabitats where microbes compete for access to resources. Because microbes interact with each other while responding to environmental changes, the growth of one group may inhibit or promote the proliferation of another. This leads to the distinct biogeography, or geographical distribution, of microbes throughout the intestine.

The upper GI tract consists of the mouth, esophagus, stomach, and the first part of the small intestine (or the duodenum). Mastication, or chewing, breaks your meal down into smaller pieces that mix with the solution of water and digestive enzymes in your saliva. After passing through the esophagus, you come to the stomach—an expansive reservoir that can temporarily stretch enough to accommodate anywhere from 50 milliliters up to 2 liters. As it churns, food is mixed with the 1 to 3 liters of gastric juices it produces each day. It maintains an impressively acidic pH of about 1 to 2, which serves two functions: breaking down food and providing an initial line of defense against potential pathogens. Between the extreme acidity and the high oxygen levels, many pathogenic bacteria won't survive transit through the stomach.

The lower GI tract includes the small and large intestines, which are long, fleshy tubes. Your abdominal cavity is packed with 22 feet of small intestine and 5 feet of large intestine. The entire lower GI tract is surrounded by layers of smooth muscle called the muscularis externa, which contract to mix food with digestive enzymes and push it along, like a conveyor belt in a factory. The intestines are the main site of digestion and absorption, so they're lined with specialized cells that produce digestive enzymes and absorb nutrients. The cells of the small intestine are covered with a thin layer of protective mucus, but in the large intestine, they are covered by two thick layers of mucus. The uppermost layer serves as a shelter and nutrient source for certain microbes, while others prefer to exist in the lumen, the cavity of the intestine itself.

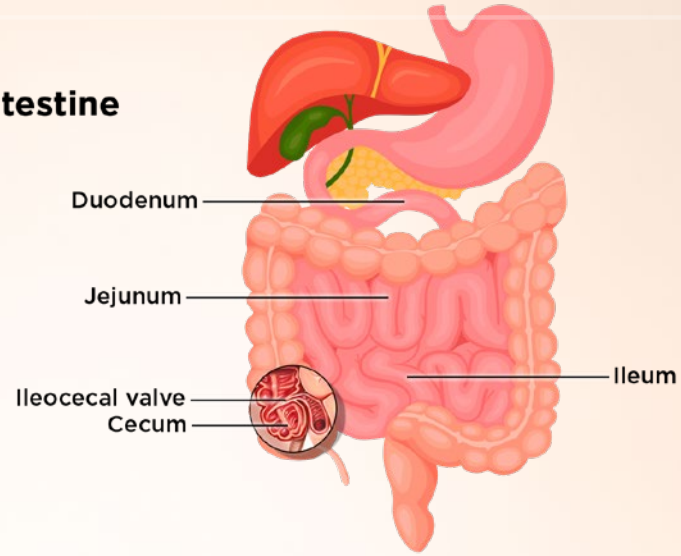


The gut contains an estimated 70% of your immune tissue and cells. Beneath the intestinal cells lies the lamina propria, a supportive connective tissue rich in immune cells. The proximity of the microbiota to the lamina propria and its multitude of immune cells and receptors provides opportunities for bidirectional communication between the immune system and the microbes throughout your life span.

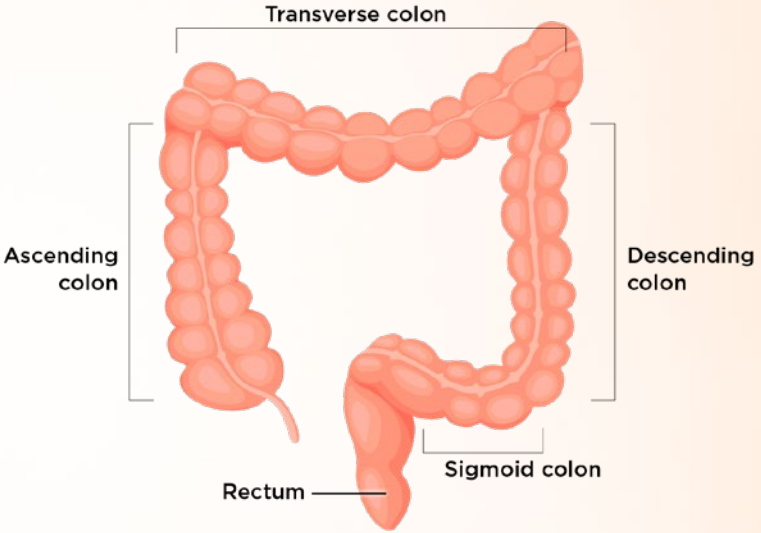
The Small and Large Intestines

The small intestine is divided into three sections: The first segment, the duodenum, is connected to the stomach; the middle section is called the jejunum; and the third segment is called the ileum. It joins the large intestine at an area called the cecum, and a sphincter known as the ileocecal valve maintains a barrier between the small and large intestines to prevent backflow of digestive contents and bacteria. It also controls the flow of digestive contents into the large intestine in response to pressure from food reaching the latter end of the ileum.

Small Intestine



Large Intestine



The internal structure of the small intestine is specialized to maximize its surface area. This internal lining is bunched into finger-like projections called villi, which are covered with tiny hair-like projections called microvilli. These are coated with digestive enzymes. The extensive folding of the small intestine increases its absorptive capacity by 600 times what it would be as a straight tube, and it's so adaptive that up to 50% of it can be lost or removed before its functional capacity is affected.

Because it's connected to the stomach, the duodenum is also fairly acidic and high in oxygen. Here, bacterial numbers and diversity are only slightly greater than what you would find in the stomach. However, numbers of bacteria in the jejunum and ileum are two to four times greater than those in the stomach or duodenum, and they're also much more diverse. The cecum is a large pouch where digested food can pool and provide a rich source of nutrients for bacteria, making it a major site for bacterial growth.

Through the cecum, you pass into the large intestine, which plays an important role in both fluid and electrolyte balance as it absorbs these from feces. The large intestine is folded into three portions: the ascending, transverse, and descending colon—the latter of which ends in an S-shaped structure called the sigmoid colon. The sigmoid colon terminates at the rectum, which controls the exit of feces via the external sphincter.

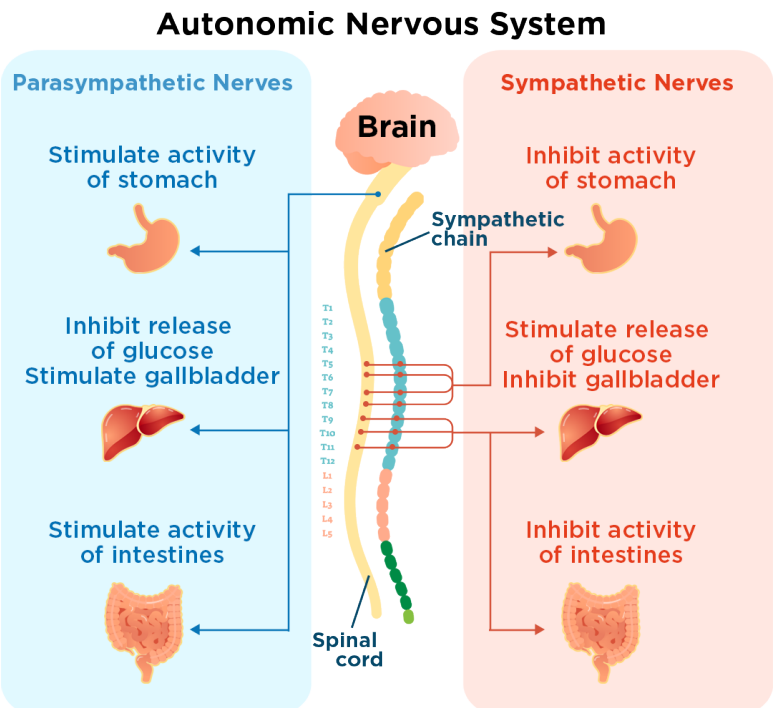
The large intestine forms stool by holding digestive contents in place while absorbing water, electrolytes, and small amounts of vitamins and minerals. Stool is usually about 75% water. The remaining 25% is composed primarily of dead bacteria and indigestible fiber, along with some undigested fats, minerals, and other dead cells. The large intestine contracts to move stool toward the rectum, where defecation can take place.

The large intestine houses most of the gut microbiome. It's filled with nearly 2 kilograms—or about 4 pounds—of microbes. There are approximately 10 times more bacteria in the large intestine than in the stomach and duodenum and up to 2.5 times more than in the jejunum and ileum. The microbial population here is so diverse because the environment is low in acid and oxygen but rich in dietary fibers indigestible to humans. The large intestine is also the primary site of bacterial fermentation, which is a process bacteria use to convert that fiber to usable energy. Fermentation produces gases (such

as methane), SCFAs (such as butyrate), and even some neurotransmitters. Some bacteria also produce small amounts of vitamins such as biotin and vitamin K.

The Second Brain

The gut has its own specialized nervous system, which has earned it the title of the "second brain." Our autonomic nervous system, which regulates automatic processes like perspiration, still controls the digestive tract for the most part, but the enteric nervous system of the gut can act independently. Chemical messengers called neurotransmitters are produced by cells in the digestive tract itself as well as nerves from the two branches of the autonomic nervous system.



The sympathetic branch controls your fight-or-flight response, while the parasympathetic branch regulates digestion and helps you return to a calmer state once you've escaped a threat. Most sympathetic signaling is sent along splanchnic nerves in the abdomen.

The vagus nerve, which carries most of the parasympathetic signals, runs straight from the brain to the gut. These signals control the wavelike contractions that move food along, the secretion of digestive enzymes, and the contraction and relaxation of sphincter muscles that control passage of digestive contents throughout—and out of—the digestive tract.

What's in Your Gut?

All three main domains of life can be found in the human gut: Eukarya (which includes plants, animals, and fungi), Archaea, and Bacteria. As far as researchers know, the human gut microbiome can be divided into 12 bacterial phyla. The phylum level of organization is general, like comparing vertebrates to invertebrates. Ninety percent of the bacteria in the digestive tract come from four groups: Bacteroidetes, Firmicutes, Actinobacteria, and Proteobacteria. These large groups contain numerous genera, which you can organize into more closely related groups of species.

A species of bacteria is defined by a high degree of structural and functional similarity. However, it isn't the same definition that is used to describe a species of lizards or birds. At this point, *species* is the best word available, so it is used to differentiate between members of a genus, just like you differentiate between dogs and jackals. In some cases, species can be further divided into strains. This is important to keep in mind because the activities and potential health effects of bacteria are strain specific. Differentiating strains is similar to differentiating between dogs and wolves. Within a single species, some strains are sold as probiotic supplements, while others can cause GI diseases. As researchers discover and categorize new organisms, the taxonomy changes.

The gut is remarkably diverse. In microbiome science, if the number of each species in the gut is relatively proportional, the microbiome is more diverse. If one species is disproportionately abundant in comparison to the others, the microbiome is less diverse. However, not all researchers use both metrics,

and it becomes more difficult to determine the diversity of a microbiome with many species in disproportionate amounts or an even distribution of a limited number of species. So, the meaning of diversity still varies from one publication to the next.

Bacterial Exposure

Bacterial exposure in infancy has a significant and lasting effect on the gut microbiome. Infants' digestive tracts are primarily colonized during the birth process. The bacterial colonies rapidly multiply and diversify for 2 to 3 years before reaching the more stable adult composition. Delivery mode, gestation time, and diet all play a role in shaping your gut microbiome.

A baby born vaginally after a full-term pregnancy will have a gut microbiome similar to that of the mother within about 3 days. This is probably due to exposure to colonic bacteria, such as *Bifidobacteria,* during birth. This is likely why the dietary pattern of the mother significantly influences the gut microbiome of her infant in the case of a vaginal birth. A baby born via cesarean section, however, develops a less-diverse gut microbiome that resembles the environment of the delivery room and the mother's skin microbiome. These differences appear to persist up to about 6 months of age, and their impact on health and disease later in life is still unclear.

Breast milk delivers human milk oligosaccharides and bacteria, so babies who breastfeed have more *Bifidobacteria* and *lactobacilli* in their microbiomes compared to those who are formula-fed. The period of time before weaning plays an important role in the development of immunotolerance to gut microbes. Some evidence points to a connection between low microbial diversity early in life and the development of eczema, asthma, and allergies later in life, but the cause is still unknown. The infant microbiome becomes more diverse once complex carbohydrates are introduced during or after weaning.

Finally, current theories suggest that early antibiotic use could lead to lasting changes in the microbiome, such as reduced diversity and the growth of antibiotic-resistant bacteria. The effects of short-term broad-spectrum antibiotic use on the microbiome and metabolic function vary from person to person, and they could last months or years.

It's estimated that about 60% of our bacterial inhabitants remain unchanged throughout adulthood until old age, when the microbiome starts to lose diversity. Researchers estimate that somewhere between 20% and 60% of our bacterial diversity can be influenced by diet and exercise, with our genes accounting for only 12%. It also appears that roughly one-third of the biome is shared among humans, but there's so much individual variability that no two microbiomes are the same.

READING

Cossart, P. *The New Microbiology*. Washington DC: ASM Press, 2018. https://www.perlego.com/book/1353534/the-new-microbiology-pdf.

Cullen, C. M., K. K. Aneja, S. Beyhan, C. E. Cho, S. Woloszynek, M. Convertino, S. J. McCoy, et al. "Emerging Priorities for Microbiome Research." *Frontiers in Microbiology* 11 (February 2020). https://doi.org/10.3389/fmicb.2020.00136.

Douglas, A. *Fundamentals of Microbiome Science*. Princeton, NJ: Princeton University Press, 2021.

Flint, H. J. *Why Gut Microbes Matter*. Cham, Switzerland: Springer International Publishing, 2020. https://doi.org/10.1007/978-3-030-43246-1.

Haller, D., ed. *The Gut Microbiome in Health and Disease*. Cham, Switzerland: Springer International Publishing, 2018. https://doi.org/10.1007/978-3-319-90545-7.

3

How Researchers Study the Gut Microbiome

In 2011, a group of researchers made a “keystone discovery” in gut microbiome research: People could be organized into at least three groups called enterotypes based on how their gut microbes were distributed. However, new methods eventually revealed flaws in the theory, and the three proposed types of microbiomes might be less distinct than initially thought. The seismic changes around the idea of enterotypes reflect how gut microbiome science is a way of becoming less wrong over time. In this lecture, you’ll explore how researchers study the gut microbiome as well as a few questions you can ask to spot bad research. You’ll also see the strengths and limitations of common methods and how those methods are evolving to give researchers a more accurate and useful picture of human gut health.

The Gut Microbiome Research Process

Like any other scientific field, gut microbiome science begins and ends with the scientific method. This is a process of making educated guesses, or hypotheses, based on your observations and then testing the accuracy of your guesses with experiments to collect more observations in a specific scenario. The gold standard for hypothesis testing is a randomized, placebo-controlled trial (RCT).

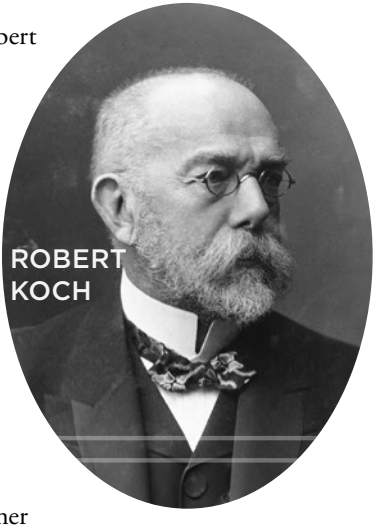
In an RCT, an intervention—like a specific probiotic—is compared to a placebo, or an inert substance that shouldn't have an effect. Ideally, the experiment is also blinded, which means that neither the participants nor the researchers know who's getting what. This reduces the chances of bias or a placebo effect. By comparing the intervention's effects to the placebo's effects, researchers can be more certain that the intervention had some effect and that the results weren't due to chance.

Researchers are only able to get closer to correctly explaining a phenomenon by ruling out other potential answers that are more likely to be wrong. In some cases, they aren't even trying to determine whether one thing causes another. Instead, they might be looking for correlations, or relationships, between two things. A study that looks for correlations is called an observational study. Importantly, observational studies can help researchers form hypotheses that can be tested with RCTs.

Many studies linking dietary patterns to the gut microbiome are performed as observational studies. Researchers collect survey data and stool samples from a large population and then compare something like their vegetable intake to the diversity of their stool sample to draw a correlation. They may go on to develop a hypothesis about certain vegetables supporting microbiome diversity, which they could then test with an RCT. This is a key take-home message: If someone says that a specific study proves something, they might be more interested in making a persuasive statement than an accurate one. At the moment, no causative relationships have been established between the gut microbiome and any health or disease outcome.

The History of Microbiome Research

The field of microbiology emerged as a distinct science in the mid-1800s, when Louis Pasteur used microscopy to demonstrate that microorganisms were responsible for fermentation and food spoilage. At the time, the leading hypothesis was that disease and fermentation happened spontaneously. However, Pasteur's new germ theory suggested that disease and spoilage were caused by tiny organisms called germs. This was a huge step toward the most accurate explanation researchers have so far, which is that microorganisms cause both fermentation and disease.



In 1878, a German microbiologist named Robert Koch developed four criteria for establishing a cause-and-effect relationship between a microbe and a disease: First, the microbe should only be found in people with the disease. Second, the microbe has to be cultured, or grown, in a petri dish without the presence of other organisms. Third, the microbe then needs to be introduced to a healthy person or animal, and that recipient needs to develop the disease in question. Finally, the microbe needs to be found in the recipient, without having mutated in some way so as to make it a new strain. Koch's postulates have been modified slightly to include microbial communities rather than single microbes, but they still serve as criteria to establish a causal relationship between gut microbes and health outcomes.

Today, when researchers collect a stool sample, they can preserve the genetic material in that sample and then either amplify certain genes of interest to identify specific microbes or look at all of the genetic material and try to piece the profile of the microbiota together like a puzzle. In short, gene sequencing allows researchers to see what microbes actually populate a sample. Such techniques allow researchers to describe microbiomes in a few different ways. They can calculate the alpha-diversity—or richness and evenness—of a single sample based on the number and proportions of observable microbes, or

they can measure the microbial products, like SCFAs, in the sample. These measurements give researchers an idea of what the microbes were doing when the sample was collected.

These days, a fecal sample can also be transplanted from a donor host—human or animal—into a recipient. When this is done for research purposes, it's considered an intervention. This would be an example of an RCT. The recipient's physiology or behavior can be studied to determine whether the microbiota in the donor's sample caused a change in the recipient. This procedure is called a fecal microbiota transplantation (FMT), and it's uniquely capable of bringing researchers as close as possible to understanding a causal relationship between microbes and disease.

Researchers now have many options for collecting, processing, and analyzing microbiome samples. However, the more methods there are, the harder it is to standardize procedures and replicate results between research groups. There are also some known issues with common research methods. A sample can be easily contaminated by the local environment, and sample storage and processing can also affect the results. Moreover, microbes differ in their ability to tolerate oxygen and withstand the processes used to extract their genetic material, which means that certain species might be over- or underrepresented based on the extraction methods. This makes it difficult to form consensus statements about the gut microbiome and sometimes leads to premature conclusions that later turn out to be incorrect.

Determining Fact versus Fiction

Dr. William Hanage, a Harvard University epidemiology professor, recommends asking a few key questions as you read the latest microbiome research while keeping the abovementioned limitations in mind.

First, did the researchers detect changes that actually matter? For example, a 2021 study compared fermented foods to high-fiber foods and found that people who ate the fermented foods had lower inflammatory markers than those who ate the high-fiber foods. Some media outlets claimed that the fermented foods were anti-inflammatory or that high-fiber foods cause inflammation. However, the data really revealed that both groups had normal, healthy levels of

inflammatory markers before and after the study. So, compared to each other, the participants might have had lower or higher inflammatory markers, but they were never experiencing objectively high or low levels of inflammation. So, did the change in inflammatory markers matter?

Second, did the study indicate causation or just correlation? Remember that researchers haven't established any cause-and-effect relationships between the gut microbiome and any aspect of health and disease. And microbiomes are so complex that they might never achieve a complete understanding of cause and effect between the microbiome and human health. Establishing causation is further complicated by the fact that researchers don't know the genomes of a great number of microbes, so they can't identify all microbes in a sample. This makes it difficult to suggest causal relationships between the sample microbiome and health or disease. The ability to differentiate between specific strains is also limited with certain techniques, meaning experiments sometimes lack the resolution necessary to identify a strain of interest and link it to any outcome.

Third, did the researchers identify an actual mechanism, and does it make sense? A mechanism is a specific phenomenon that could explain an outcome, like a metabolic process that a microbe uses to produce a SCFA. Many researchers will use the idea of dysbiosis as the "mechanism" to explain the results of their study. But in the words of Drs. Scott Olesen and Eric Alm, biological engineering researchers out of the Massachusetts Institute of Technology, dysbiosis is "a 'mechanism-free' cause of disease to which we can retreat when plausible mechanistic explanations are discounted."

Fourth, Hanage also encourages readers to question how well an experiment reflects reality. For example, stool samples can be useful, but researchers should be careful about the conclusions they draw from them. A human stool sample closely represents the microbes that live in the lumen of the large

intestine. But that population is significantly different from the community that you'd find in the mucosal layer of the large intestine or any part of the small intestine. Despite this important distinction, many publications use the term *gut microbiome* when they've actually only studied the stool microbiome. So, when you consider whether an experiment reflects reality, you're really asking two questions: Are the terms precisely and accurately defined? And do the results really apply to the human gut microbiome?

Finally, you should ask whether the results could be explained by anything else. So many factors shape the gut microbiome—from geography to gender and prescriptions to pets. Researchers call these confounding factors because they distort the relationship between variables in a study. For example, when the abundance of a microbe differs after an intervention, it's tempting to conclude that the difference is due to the intervention itself. But it could be the result of something that's almost impossible to account for, like whether the participant had a childhood pet. The microbiota themselves can also be confounding factors because they interact with one another in ways researchers don't understand—or haven't discovered yet.

Genome Sequencing Advancement

Recent technological advancements in sample analysis have made genome sequencing faster and more affordable. Whole genome sequencing, for example, breaks the genome into small pieces, sequences each one, and then puts them all together to get a complete genome sequence. It has only been in use since 2006, but its accuracy allows researchers to update genomic reference libraries as various labs discover new microbes. This is making the view of the microbiome more complete and identification more reliable.

Gut scientists are also borrowing new statistical methods from other sciences that are better suited to studying complex ecosystems. Mendelian randomization, for example, is a method borrowed from epidemiology. It looks at the differences in health outcomes between people who have a genetic variation—or "mutation"—and those who don't to discern whether a specific risk factor is truly causing a certain health outcome. It has the ability to rule out other potential causes because the genetic variation isn't affected by any

confounding factors (such as age, sex, or lifestyle) that could be present in traditional observational studies. Researchers can use this method to compare the impact of a person's genes against the impact of certain gut microbes to estimate how causal the microbes might be.

The integration of these techniques, which analyze whole, complex systems rather than singular microbes, has started to reshape how researchers think about causation. Some are even questioning whether it's appropriate to apply the same set of criteria historically used to establish a cause-and-effect relationship between a single microbe and a disease to an entire microbiome and a disease. As a result, the relevance of Koch's postulates has come under fire. Perhaps one of the most important changes is that researchers are naming these limitations and calling for more collaboration to standardize best practices from sample collection to the reporting of results. Eventually, this could lead to better translation from research to real life.

READING

Allaband, C., D. McDonald, Y. Vázquez-Baeza, J. J. Minich, A. Tripathi, D. A. Brenner, R. Loomba, et al. "Microbiome 101: Studying, Analyzing, and Interpreting Gut Microbiome Data for Clinicians." *Clinical Gastroenterology and Hepatology* 17, no. 2 (2019): 218–230. https://doi.org/10.1016/j.cgh.2018.09.017.

Cani, P. D., E. Moens de Hase, and M. van Hul. "Gut Microbiota and Host Metabolism: From Proof of Concept to Therapeutic Intervention." *Microorganisms* 9, no. 6 (2021). https://doi.org/10.3390/microorganisms9061302.

Cullen, C. M., K. K. Aneja, S. Beyhan, C. E. Cho, S. Woloszynek, M. Convertino, S. J. McCoy, et al. "Emerging Priorities for Microbiome Research." *Frontiers in Microbiology* 11 (February 2020). https://doi.org/10.3389/fmicb.2020.00136.

Hanage, W. "Microbiology: Microbiome Science Needs a Healthy Dose of Skepticism." *Nature* 512 (2014): 247–248. https://doi.org/10.1038/512247a.

Olesen, S. W., and E. J. Alm. "Dysbiosis Is Not an Answer." *Nature Microbiology* 1 (2016): 1–2. https://doi.org/10.1038/nmicrobiol.2016.228.

Walter, J., A. M. Armet, B. B. Finlay, and F. Shanahan. "Establishing or Exaggerating Causality for the Gut Microbiome: Lessons from Human Microbiota–Associated Rodents." *Cell* 180, no. 2 (2020): 221–232. https://doi.org/10.1016/j.cell.2019.12.025.

4 How to Spot Gut Health Pseudoscience

The scientific method is a systematic approach used by scientists to investigate and understand natural phenomena. It involves several steps, including formulating a hypothesis, designing and conducting experiments, analyzing data, and drawing conclusions based on the evidence gathered. It relies on empirical evidence, rigorous experimentation, and critical analysis. Pseudoscience, however, refers to beliefs, theories, or practices that are presented as scientific but lack the characteristics of genuine scientific inquiry, such as empirical evidence. Pseudoscientific claims often lack consensus within the scientific community and are frequently based on anecdotal evidence or subjective experience. In this lecture, you'll examine several pseudoscientific gut health claims and learn how to separate the science from the science fiction.

Identifying Pseudoscience

One of the most obvious indicators of pseudoscience is that it relies heavily on anecdotal evidence, personal testimonials, appeals to authority, or subjective experiences to support its claims. In addition, pseudoscientific claims tend to go against the consensus of the scientific community. Consensus here means that multiple rigorous investigations have consistently replicated the same results, which gives researchers a degree of confidence that these results are reliable. Pseudoscience also often lacks rigorous peer review and is not published in reputable scientific journals. Peer review involves independent experts critically evaluating and providing feedback on the research before it is published.

Pseudoscientific claims are often formulated in such a way that they cannot be tested or disproven. Promoting unfalsifiable claims prevents scientific progress, which relies on the ability to adapt and revise theories based on new evidence. Peddlers of pseudoscience also tend to selectively choose data that support their claims while disregarding or downplaying contradictory evidence. In genuine scientific research, all relevant data are considered, including both supporting and conflicting results. Finally, pseudoscientific claims may be driven by financial motivations or conflicts of interest.

"Leaky Gut"

One of the most common examples of pseudoscience is leaky gut. Essentially, *leaky gut* refers to increased intestinal permeability, which means that substances can pass between the cells of the intestinal wall more easily than they normally would. The permeability of the intestinal lining does, in fact, vary from person to person, and increased permeability is associated with some health conditions. However, it's important to note that leaky gut is not a recognized medical condition that can be diagnosed or treated specifically.

Under normal conditions, the intestinal wall is semipermeable, allowing certain substances to pass through in a controlled manner. However, certain factors, such as GI disease, obesity, or a high-fat diet, may increase intestinal permeability by affecting the proteins that hold the intestinal cells tightly together. As these proteins loosen their hold, substances can pass between the intestinal cells and encounter immune cells, which can trigger chronic inflammation. Researchers can measure altered intestinal permeability in humans, mostly by quantifying a specific protein or using a dual-sugar test. However, there is currently no gold standard test. Also, altered intestinal permeability is not, in itself, a disease, but it can be associated with certain disease states, such as advanced intestinal disease.

Currently, tests for intestinal permeability are not diagnostic, and no causative links have been established between altered intestinal permeability and any disease. Additionally, there is no known treatment for this change in permeability. It seems that preventing altered intestinal permeability is the best course of action, and this may involve making healthy lifestyle choices. A Westernized dietary pattern, a body mass index over 25, and intense endurance exercise (especially under heat stress) are all associated with an increased likelihood of this issue.

Unfortunately, some online "gut health gurus" claim to diagnose and treat leaky gut based on vague symptoms, such as brain fog, fatigue, and GI pain. However, there is no clear link between these symptoms and increased intestinal permeability. So, in this case, a scientific reality is stretched into the realm of pseudoscience.

Selling Solutions

Another pattern is to create a problem to sell a solution. Many influencers attract attention with fear-based marketing, like claiming that certain substances, such as artificial sweeteners, will destroy the gut microbiome. The Food and Drug Administration (FDA) has approved several artificial sweeteners, including acesulfame K, aspartame, saccharin, and sucralose, for use as sugar substitutes. Their impact on the gut microbiome is a newer area of research. Some studies have exposed isolated cells to artificial sweeteners, and the effects have ranged from increased hormone production to inflammation. However, most rodent models and human data don't show any of these findings. So, based on the highest-quality data available, artificial sweeteners are safe for both humans and their gut bacteria.

You may have also heard of comprehensive stool analysis tests that claim to detect microbes in your gut that could be contributing to your illness or other vague symptoms. These tests often come with recommendations for a specific diet to fix any issues. However, there are several red flags associated with these tests. For example, distributors establish their own reference ranges for "normal" levels of organisms, despite the lack of a scientific consensus.

No current technology can identify all of the microbes present in the fecal microbiome. Furthermore, no nutritional recommendations will cause targeted changes to the microbiome. These stool tests also falsely equate certain microbes with disease; however, some microbes are essential inhabitants that likely play an important role in the development of our immune systems. Without the ability to determine "ideal" relative abundances of microbes or identify species at the strain level, these clinical applications are limited. In conclusion, gut health protocols that claim to diagnose dysbiosis or reset the gut are lacking in empirical evidence and physiological probability.

What about tests that claim to identify food sensitivities? Unfortunately, they're not valid. To understand why, it's helpful to know a little bit about the immune system and the different types of antibodies. Antibodies are produced in response to foreign substances entering the body, including allergenic foods: Allergies are immune responses, so they'll prompt the production of antibodies. Immunoglobin E antibodies are involved in allergic

responses, while immunoglobulin G (IgG) antibodies help the immune system neutralize pathogens. An IgG antibody response is actually a sign of a healthy immune system recognizing an antigen as familiar and harmless.

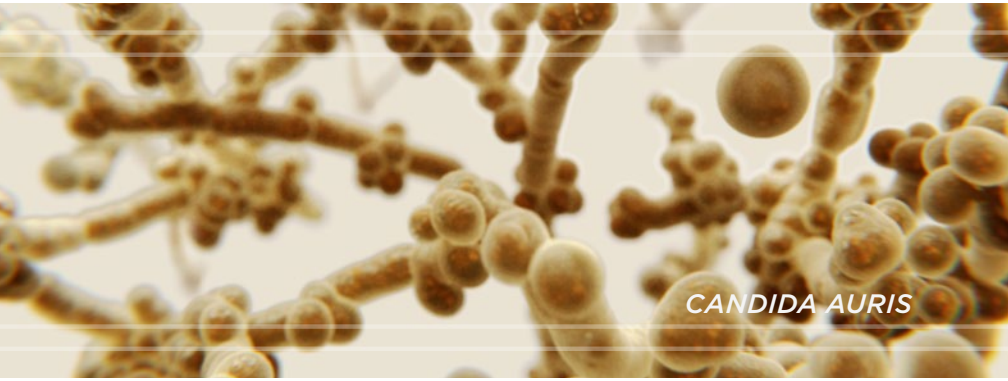
The idea behind food sensitivity testing is to look for certain antibodies that supposedly appear when a specific food is presented. Food sensitivity tests only measure one antibody, though: the IgG antibody. That is, these tests will list foods that you've eaten in the past—foods that your immune system recognizes as harmless—but removing those foods may lead to nutritional deficiencies and psychological harm.

Detoxes and Cleanses

Cleanses and detoxes are marketed as a way to get rid of "toxins" in your body, lose weight, and improve gut health. But the term *toxins* is used pretty loosely in the detox diet industry. Pollutants, man-made chemicals, processed food ingredients, and heavy metals are often grouped together under this umbrella term, which is poorly defined to begin with. The truth is, there's no evidence that any commercial detox diet will remove any of these substances from your body. Despite this, many practitioners still prescribe and publish "clinical detox" procedures, which is concerning.

Your body already has ways to remove waste products from your system through enzymatic pathways and excretory systems in the liver, kidneys, GI tract, skin, and lungs. The best way to support your body's detoxification system is to eat a healthy diet that's high in fruits, vegetables, whole grains, beans, poultry, and fish. The risks of detox diets and cleanses are serious, including nutrient deficiencies, electrolyte imbalances, diarrhea, dehydration, disordered eating, and even death. Some diets involve long-term fasting, exclusion of solid foods, laxative use, unregulated supplements, extended sauna use, and strict dietary rules.

You may have also heard that you should protect your gut from yeasts. Yeasts are eukaryotic microorganisms classified as fungi. They can be found in both the oral and vaginal cavities, and they can also be detected in adult stool samples. The most abundant group of yeasts found in the human body is *Candida,* followed by *Saccharomyces* and *Malassezia*. Though they make up a small portion of the gut microbiome, emerging research suggests that they're essential members of the community. They've even earned their own title: the mycobiome.



Some yeasts can cause infections, particularly *Candida*, but the evidence does not support claims that *Candida* causes GI distress or impaired gut barrier function or that it leads to nebulous symptoms, such as fatigue. However, *Candida* can delay the healing time of ulcers in IBD and may overpopulate the GI tracts of newborns, making them susceptible to a systemic infection. To diagnose intestinal *Candida* infection, endoscopy and histology are required, while systemic fungemia requires a blood test.

Studies suggest that a large proportion of the yeasts found in stool samples actually come from our diet or saliva. So, making dietary modifications to avoid yeast-containing food and brushing and flossing frequently can help reduce the amount of certain yeasts in stool, but that may not reflect what's actually going on in your gut. Nevertheless, unsupported hysteria around *Candida* has given rise to plenty of pseudoscientific claims that advocate for ineffective dietary changes.

Contrary to popular belief, eating refined carbohydrates is not associated with elevated *Candida albicans* in humans. Also, adding sugar to the diet only raises fecal *Candida albicans* in individuals who already had high levels of oral yeast. Moreover, fecal counts don't indicate actual intestinal colonization, and oral yeasts are likely ingested with saliva without colonizing the intestines of healthy individuals. It's also important to know that there are no interventions to cleanse the gut of yeasts, and even antifungal drugs can't eliminate them entirely. However, antifungal drugs are effective treatments when fungal infections are present.

Supplements

Lately, many fitness personalities have been talking about taking collagen supplements to improve gut health. Collagen is a protein that's important for the structure of our bodies, but there's no evidence yet that it affects gut function or the gut microbiome. It is broken down in our bodies just like other proteins, so it's unlikely to have a direct effect on the lamina propria in the gut, which is one of the proposed mechanisms by which some suggest it could "heal" the gut.

You might also encounter enzyme supplements that supposedly aid in digestion. To evaluate how well digestive enzymes work, you need to understand the differences in pH along our digestive tract. The pH levels in our saliva, stomach, small intestine, and large intestine are all different. Enzymes are active at specific pH ranges, and if they're outside of that range for too long, they'll lose their shape and function, a process called denaturation. Our bodies produce different types of enzymes to break down the macronutrients we consume, like carbohydrates, proteins, and fats. If we lack certain enzymes, as in the case of lactose intolerance, we might experience digestive issues like gas, bloating, or diarrhea.

Enzyme supplements claim to alleviate these issues, but there isn't much evidence to support their effectiveness across the board. Some manufacturers try to protect the enzymes from denaturation by encapsulating them in a coating, but even prescription-strength enzymes can struggle to survive the

acidic environment of the stomach. In fact, well-designed RCTs have shown that only about half the participants notice any improvements after using digestive enzymes.

And while powdered produce blends are often marketed as a way to improve digestion and cardiovascular health, as well as providing the equivalent of multiple servings of fruits and vegetables, the evidence here is also pretty mixed. Most of the research has been done on specific brands of these powders, and the studies have often been carried out by physicians who are also selling the products—which makes the results suspect, to say the least. Some studies have shown that they can reduce markers of protein, lipid, and DNA oxidation or damage to these molecules. But the evidence is still limited, and these products are sold as supplements, not as food. This means that their blends are often proprietary, and testing for purity and efficacy isn't required. Plus, some vitamin and herbal supplements can interact with prescription medications.

READING

Chen, Chung-Yu, Cheng-Hao Tien, Yue-Hwa Chen, D. Garrido, A. Farzi, H. Herzog, Hsien-Yu Fan, and Yang-Ching Chen. "Effect of Sucralose Intake on Human and Mouse/Rat Gut Microbiota Composition: A Systematic Review and Meta-Analysis." *Food Reviews International* (2023). https://doi.org/10.1080/87559129.2023.2212045.

Flores R., J. Shi, B. Fuhrman, X. Xu, T. D. Veenstra, M. H. Gail, P. Gajer, et al. "Fecal Microbial Determinants of Fecal and Systemic Estrogens and Estrogen Metabolites: A Cross-Sectional Study." *Journal of Translational Medicine* 10, no. 1 (2012): 1–11. https://doi.org/10.1186/1479-5876-10-253.

Gingras, B. A., and J. A. Maggiore. "Performance of a New Molecular Assay for the Detection of Gastrointestinal Pathogens." *Access Microbiology* 2, no. 10 (2020). https://doi.org/10.1099/acmi.0.000160.

Lacour, M., T. Zunder, R. Huber, A. Sander, F. Daschner, and U. Frank. "The Pathogenetic Significance of Intestinal *Candida* Colonization: A Systematic Review from an Interdisciplinary and Environmental Medical Point of View." *International Journal of Hygiene and Environmental Health* 205, no. 4 (2002): 257–268. https://doi.org/10.1078/1438-4639-00159.

Leech, B., E. McIntyre, A. Steel, and D. Sibbritt. "Risk Factors Associated with Intestinal Permeability in an Adult Population: A Systematic Review." *International Journal of Clinical Practice* 73, no. 10 (2019): e13385. https://doi.org/10.1111/ijcp.13385.

McFarland, L. V., C. T. Evans, and E. J. C. Goldstein. "Strain-Specificity and Disease-Specificity of Probiotic Efficacy: A Systematic Review and Meta-Analysis." *Frontiers in Medicine* 5 (2018): 124. https://doi.org/10.3389/fmed.2018.00124.

Ramezani Ahmadi, A., E. Rayyani, M. Bahreini, and A. Mansoori. "The Effect of Glutamine Supplementation on Athletic Performance, Body Composition, and Immune Function: A Systematic Review and a Meta-Analysis of Clinical Trials." *Clinical Nutrition* 38, no. 3 (2019): 1076–1091. https://doi.org/10.1016/j.clnu.2018.05.001.

Stapel, S. O., R. Asero, B. K. Ballmer-Weber, E. F. Knol, S. Strobel, S. Vieths, and J. Kleine-Tebbe. "Testing for IgG4 against Foods Is Not Recommended as a Diagnostic Tool: EAACI Task Force Report." *Allergy* 63, no. 7 (2008): 793–796. https://doi.org/10.1111/j.1398-9995.2008.01705.x.

Tepler, A., G. Hoffman, S. Jindal, N. Narula, and S. C. Shah. "Intake of Artificial Sweeteners among Adults Is Associated with Reduced Odds of Gastrointestinal Luminal Cancers: A Meta-Analysis of Cohort and Case-Control Studies. *Nutrition Research* 93 (2021): 87–98. https://doi.org/10.1016/j.nutres.2021.07.007.

Tomicić, S., G. Norrman, K. Fälth-Magnusson, M. C. Jenmalm, I. Devenney, and M. F. Böttcher. "High Levels of IgG4 Antibodies to Foods during Infancy Are Associated with Tolerance to Corresponding Foods Later in Life." *Pediatric Allergy and Immunology* 20, no. 1 (2009): 35–41. https://doi.org/10.1111/j.1399-3038.2008.00738.x.

5 The Microbiome and Gastrointestinal Disease

In 1984, Dr. Barry Marshall drank a microbial cocktail to intentionally infect himself with *Helicobacter pylori*. After years of failed attempts in animal studies, he hoped to fulfill Koch's postulates and prove that *H. pylori* was the cause of peptic ulcers. He didn't develop any peptic ulcers, but he did experience nausea, bad breath, and gastritis. This allowed him to establish a causal relationship between *H. pylori* and gastritis but not with peptic ulcers. However, Dr. Marshall and his collaborator Dr. J. Robin Warren were still later awarded the Nobel Prize for Physiology or Medicine for their discovery that *H. pylori* could colonize the stomach and its relationship with gastritis and peptic ulcers. Dr. Marshall's experiment demonstrates how complicated the relationship can be between the microbiome and GI disease—which is what you will learn about in this lecture.

The Difficulty of Determining Pathogenicity

Your microbiome is under constant surveillance by your intestinal cells, which have the difficult task of defending against pathogens while remaining tolerant of the other microbes. Some of the native, or commensal, microbes can slip past these defenses, only to become pathogenic under specific circumstances. Whether a microbe becomes pathogenic is likely determined by the host's genetics, intestinal immune activity, and the overall composition of their gut microbiome. In other words, one beneficial microbe for you could be a pathogen for someone else. It's challenging to make sense of the relationship between microbes and disease because, in many cases, studies can only identify microbes down to the genus or species level—and determining pathogenicity requires analysis at the subspecies or strain level.

However, several studies have identified differences in the gut microbiota of people with a disease versus healthy individuals, and some interesting patterns have emerged. Many diseases are associated with lower levels of both taxonomic and functional diversity. That is, people with certain diseases often present with fewer distinct species of microbes, and there's less genetic variation among the species that do populate their gut. This lower level of diversity is often due to a lack of species richness compared to what's seen in healthy individuals. In some diseases, a sparser number of microbial species coincides with higher levels of potential pathogens or lower levels of beneficial microbes.

IBS is often characterized this way, with lower microbial diversity in the intestine. But IBS is also often associated with a higher level of intestinal permeability. That means the mucosal lining of the intestine is more likely to let molecules (which could be pathogens) cross the intestinal barrier. Increased intestinal permeability could allow microbe-associated inflammatory substances to enter circulation and trigger a mild but chronic inflammatory response. Some IBS symptoms, like abdominal pain, are linked to alterations of the gut-brain axis: a bidirectional communication system between the gut microbiome and the central nervous system, which regulates the activity of the GI tract. So, it wouldn't be accurate to say that lower microbial diversity causes IBS.

Researchers are faced with similar uncertainties in the case of small intestinal bacterial overgrowth (SIBO), which is a condition characterized by an abnormally high concentration of bacteria in the small intestine, especially the types that should be found in the large intestine. Symptoms often include bloating and gas, and in diagnostic tests, people with SIBO will exhale different levels and ratios of gasses compared to healthy people. These gasses are only produced by bacteria, which is how they can be used to estimate a potential overgrowth of bacteria in the small intestine.

However, research on SIBO suggests that the presence of the microbes may not play as large of a role in these symptoms as the activity of the microbes does. In other words, you might have a higher-than-average number of microbes in your small intestine, but that doesn't necessarily mean you'll have symptoms of SIBO. This microbial overabundance could be caused by several factors. So, once again, you're left to question the utility of analyzing the taxonomic diversity without evaluating changes in the function of the microbiome.

Fecal Microbiota Transplantation

IBS and SIBO both teach the same lesson: It can't be said for certain that lower microbial diversity or the abundance of certain microbes are a potential cause or consequence of GI disease activity. But if you want to find out, some study designs could provide clarity. For example, discordant twin studies use identical twins with different health statuses to discern the role of genetics versus other factors in causing disease. Identical twins share more genes than any other relatives share with one another; if one twin has a genetic disease, the other will likely have it as well. This allows researchers to attribute differences in their health to other factors, like their microbiota or the environment.

Unfortunately, these twin studies have some shortcomings. For example, it's not possible to control for some variables, like long-term dietary intake and exposure to environmental factors. To do so, researchers have used FMT to closely mimic a variety of different diseases in rodent models. Rodents who

receive an FMT from people with IBD will develop IBD-like symptoms, which points to the possible role of the microbiome in initiating the disease process.

Studies like these have led to a few theories about the role of the microbiome in GI diseases like IBD or IBS. Both IBD and IBS are associated with lower gut microbiome diversity compared to people without a GI disease. In both cases, the ratios of beneficial to potentially pathogenic microbes are skewed. For example, compared to healthy controls, fecal samples from people with IBD will often reveal a lower abundance of anti-inflammatory species, such as *Faecalibacterium prausnitzii*, and higher levels of pro-inflammatory species of *Enterobacteriaceae*. This skewed ratio suggests that the elevated immune activity could actually be a response to unusually high levels of opportunistic pathogens.

FMT can also be used as an effective treatment for certain GI diseases, with the most prolific effects in treatment-resistant *Clostridium difficile* infections *C. difficile* is a commensal microbe, so it usually exists in harmony with other members of the microbiome. But certain conditions—like long-term antibiotic use—can disrupt the microbiome, allowing *C. difficile* to overcome the competition and start producing toxins that cause severe diarrhea. When strong antibiotics aren't enough to resolve a *C. difficile* infection, FMT from a healthy donor is incredibly effective. FMT has also shown promise for reducing the symptoms of IBS, and in both cases, the improvements in symptoms are associated with lasting changes to the recipient's gut microbiome.

These findings suggest that at least some symptoms of certain diseases are much more likely a consequence, rather than a cause, of gut microbiome characteristics. However, it's still unclear which microbes could be playing the greatest role. Other research indicates that while the microbiome might not directly cause any disease, there are some microbes that could predict or even raise someone's risk of developing a GI disease later in life. For example, high levels of *Fusobacterium nucleatum* have been observed with remarkable consistency in patients with colorectal cancer, and intervention studies have found that this microbe can promote tumor growth.

Dietary Management

When it comes to diseases with complex, poorly understood causes—like IBD or IBS—there are no clear means for prevention. But it is possible to reduce your risk of developing colorectal cancer or an infectious disease, and there are several ways to alleviate symptoms of incurable diseases. Research suggests that a prudent dietary pattern could reduce your risk of colorectal cancer by about 20%. One model diet is the Dietary Approaches to Stop Hypertension (DASH) diet. The DASH diet emphasizes fruits, vegetables, whole grains, poultry, low-fat dairy, beans, and nuts and recommends limiting saturated fats and processed foods that are often high in sodium.

If you're managing a chronic GI disease like IBS or IBD, you'll likely come across a lot of conflicting information about the foods you should exclude from your diet. Some situations do warrant the exclusion of specific foods, but generally, it's best to follow a prudent, plant-centric, inclusive dietary pattern that provides as many diverse foods as possible. That will be more nutritious for you, and it will also provide a wider variety of nutrient sources to your gut microbes.

That being said, there are some evidence-based diets that can help to manage GI diseases and their symptoms. The most obvious example would be a gluten-free diet for people with celiac disease. Celiac disease is an autoimmune condition that causes severe damage to the intestines in response to the wheat protein gluten. People with non-celiac gluten sensitivity might feel better when

they eliminate gluten, but this could actually be due to the coincidental removal of fructans, which are fermentable carbohydrates found in wheat products. That leads into the next reasonable elimination diet: the low-FODMAP diet.

FODMAP stands for fermentable oligosaccharides, disaccharides, monosaccharides, and polyols. FODMAPs are highly fermentable carbohydrates found in a number of different grains, fruits, and vegetables. The fructans in wheat products are one example. Microbes can potentially produce a lot of gas when they ferment these carbohydrates to produce energy, and some of these FODMAPs also pull a lot of water into the intestines. This can result in bloating, abdominal pain, and loose stool. People with IBS might experience more severe symptoms because of the altered physiology of their intestines. A low-FODMAP diet only limits FODMAPs for a short period of time before systematically reintroducing them to test a person's tolerance to each one.

There are many other restrictive dietary patterns that are only supported by anecdotes. The specific carbohydrate diet, for example, was developed in the 1920s as a treatment for celiac disease before medical professionals knew gluten was the culprit; they thought dietary carbohydrates were to blame for people's symptoms. In the 1990s, a biochemist repopularized the specific carbohydrate diet after her child was diagnosed with celiac disease, but she modified it to create specific phases during which you could only eat certain foods. Unfortunately, she made a number of erroneous claims about the diet as a treatment for other conditions, including autism spectrum disorder, none of which are supported by any evidence.

The specific carbohydrate diet has been studied to some extent—in Crohn's disease rather than celiac disease—but so far, the research has shown that it doesn't perform any better than other prudent, whole-food-based dietary patterns. It could also increase the risk of nutrient deficiencies in populations who are already at increased risk because their disease has damaged their intestinal lining.

READING

Avuthu, N., and C. Guda. "Meta-Analysis of Altered Gut Microbiota Reveals Microbial and Metabolic Biomarkers for Colorectal Cancer." *Microbiology Spectrum* 10, no. 4 (2022). https://doi.org/10.1128/spectrum.00013-22.

Biesiekierski, J. R., S. L. Peters, E. D. Newnham, O. Rosella, J. G. Muir, and P. R. Gibson. "No Effects of Gluten in Patients with Self-Reported Non-celiac Gluten Sensitivity after Dietary Reduction of Fermentable, Poorly Absorbed, Short-Chain Carbohydrates." *Gastroenterology* 145, no. 2 (2013): 320–328. https://doi.org/10.1053/j.gastro.2013.04.051.

Elli, L., C. Tomba, F. Branchi, L. Roncoroni, V. Lombardo, M. T. Bardella, F. Ferretti, et al. "Evidence for the Presence of Non-celiac Gluten Sensitivity in Patients with Functional Gastrointestinal Symptoms: Results from a Multicenter Randomized Double-Blind Placebo-Controlled Gluten Challenge." *Nutrients* 8, no. 2 (2016): 84. https://doi.org/10.3390/nu8020084.

Illescas, O., M. Rodríguez-Sosa, and M. Gariboldi. "Mediterranean Diet to Prevent the Development of Colon Diseases: A Meta-Analysis of Gut Microbiota Studies." *Nutrients* 13, no. 7 (2021): 2234. https://doi.org/10.3390/nu13072234.

Limketkai, B. N., Z. Iheozor-Ejiofor, T. Gjuladin-Hellon, A. Parian, L. E. Matarese, K. Bracewell, J. K. MacDonald, et al. "Dietary Interventions for Induction and Maintenance of Remission in Inflammatory Bowel Disease." *Cochrane Database of Systematic Reviews* 2 (2019). https://doi.org/10.1002/14651858.CD012839.pub2.

McFarland, L. V., and S. Goh. "Are Probiotics and Prebiotics Effective in the Prevention of Travellers' Diarrhea: A Systematic Review and Meta-Analysis." *Travel Medicine and Infectious Disease* 27 (2019): 11–19. https://doi.org/10.1016/j.tmaid.2018.09.007.

Parra-Soto, S., D. Ahumada, F. Petermann-Rocha, J. Boonpoor, J. L. Gallegos, J. Anderson, L. Sharp, et al. "Association of Meat, Vegetarian, Pescatarian and Fish-Poultry Diets with Risk of 19 Cancer Sites and All Cancer: Findings from the UK Biobank Prospective Cohort Study and Meta-Analysis." *BMC Medicine* 20, no. 1 (2022): 79. https://doi.org/10.1186/s12916-022-02257-9.

Saffouri, G. B., R. R. Shields-Cutler, J. Chen, Y. Yang, H. R. Lekatz, V. L. Hale, J. M. Cho, et al. "Small Intestinal Microbial Dysbiosis Underlies Symptoms Associated with Functional Gastrointestinal Disorders." *Nature Communications* 10 (2019): 1–11. https://doi.org/10.1038/s41467-019-09964-7.

van Lanen, A.-S., A. de Bree, and A. Greyling. "Efficacy of a Low-FODMAP Diet in Adult Irritable Bowel Syndrome: A Systematic Review and Meta-Analysis." *European Journal of Nutrition* 60 (2021): 3505–3522. https://doi.org/10.1007/s00394-020-02473-0.

Vonaesch, P., M. Anderson, and P. J. Sansonetti. "Pathogens, Microbiome and the Host: Emergence of the Ecological Koch's Postulates." *FEMS Microbiology Reviews* 42, no. 3 (2018): 273–292. https://doi.org/10.1093/femsre/fuy003.

6

The Microbiome and Immunity

During the 1918 influenza pandemic, enemas were given to patients suffering from severe pneumonia as a way to help them rehydrate and potentially remove toxins and waste from the body. At the time, the use of enemas was a common medical practice and was thought to have therapeutic benefits. It turns out that enemas did not, in fact, combat influenza. But even wrong turns in science can sometimes lead to productive outcomes. Of the many enemas doctors administered to patients with influenza, some contained fecal matter from healthy individuals—and this led to a surprising discovery: Patients who received such enemas recovered more quickly from pneumonia than those not receiving the treatment. This observation eventually led to the theory that there might be a connection between the gut microbiome and the immune system. In this lecture, you'll explore that connection.

Innate and Adaptive Immunity

Immunity refers to an organism's ability to resist infection and disease by recognizing and eliminating foreign substances, such as viruses, bacteria, and other pathogens. The GI immune system, also known as the gut-associated lymphoid tissue, is the defense system of your gut. It's a complex network of immune cells and tissues that play a critical role in maintaining the health and function of the GI tract.

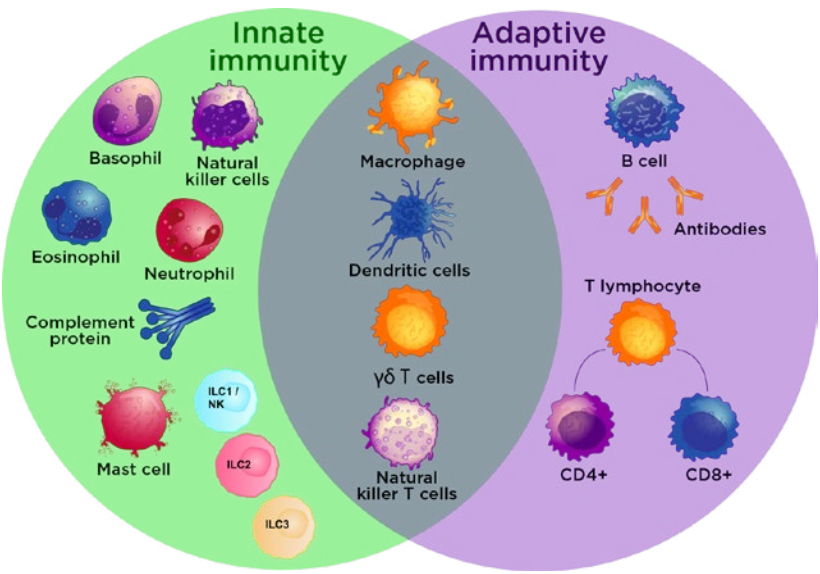
The GI immune system consists of several specialized structures, including Peyer's patches, mesenteric lymph nodes, and isolated lymphoid follicles, which are scattered throughout the intestinal mucosa. These structures contain a variety of immune cells that work together to detect and respond to pathogens and other foreign substances, initiating immune responses to clear the threat. The GI immune system is able to protect against the invasion of pathogens and harmful substances while also maintaining tolerance to harmless food and commensal bacteria. It does this by employing two different but synergistic arms: the innate immune system and the adaptive immune system. While both systems work together to maintain gut homeostasis and protect against pathogens, they differ in how they respond to an infection.

The innate immune system is your first line of defense. You were born with it. It provides rapid, nonspecific defense against all invading pathogens alike. It uses physical and chemical barriers, like your skin and stomach acid, as well as specialized immune cells, like macrophages, dendritic cells, and innate lymphoid cells. These cells have pattern-recognition receptors that allow them to recognize conserved molecular patterns on the surface of pathogens. When immune cells "recognize" these patterns, they trigger an immediate response to eliminate the pathogen. Innate immune cells also secrete chemical messengers called cytokines and chemokines that recruit other immune cells to the site of infection and activate adaptive immune responses.

The adaptive immune system provides specific, long-lasting defense against a particular pathogen. The adaptive immune response starts with antigen-presenting cells, such as dendritic cells and macrophages. These cells identify potential pathogens as foreign, recognizing specific substances, called

antigens, on their surfaces. Once they've identified a foreign pathogen, they literally catch and digest it. The antigen-presenting cells then go around showing the antigen's structure to T cells, which are white blood cells that recognize the antigen and kick-start the immune response if necessary. The T cells can then activate B cells, another type of white blood cell. B cells produce antibodies, which are specialized proteins that can bind to the antigens and neutralize the pathogen.

After the infection is cleared, a special, long-lived type of B cell called a memory B cell remains. These B cells allow the adaptive immune system to generate immunological memory, which helps the body respond more quickly and effectively to subsequent infections by the same pathogen while maintaining immune tolerance to the body's own cells. If the body is exposed to the same pathogen again, memory B cells will produce large quantities of antibodies specific to the antigens of the previously encountered pathogen. This allows the immune system to quickly and efficiently neutralize the pathogen before it can cause significant harm.



Importantly, these antibodies are also used to identify the body's own cells and harmless antigens to prevent an over-reactive immune response. Of course, this system can malfunction. Allergies are one common example, when the immune system misidentifies harmless foreign substances in the environment as harmful and attacks them. Similarly, autoimmune diseases occur when the immune system attacks the body's own healthy cells.

The Gut Immune System

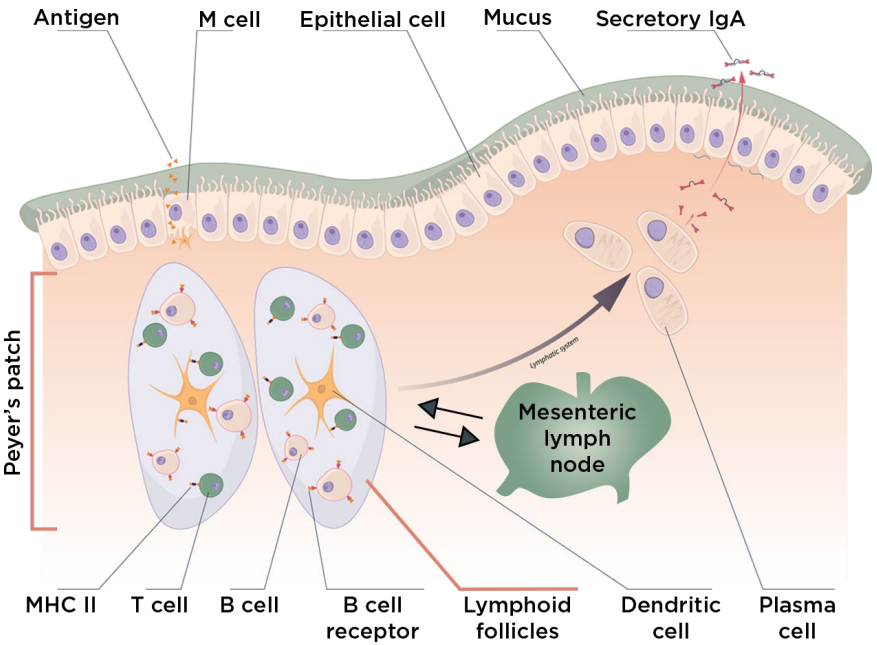
The gut immune system is intimately connected to the rest of the body's immune system through a complex network of cells, cytokines, and signaling pathways. The gut is a major site of interaction between the host and the environment, with the gut microbiota and dietary antigens constantly stimulating the immune system. As a result, the gut immune system has a significant impact on the development and function of the entire immune system, and dysregulation of gut immune responses can have far-reaching consequences.

The gut immune system is constantly communicating with the rest of the body, and one of the key pathways is through the lymphatic system. This system produces and stores white blood cells. The lymphatic vessels in the gut drain into the mesenteric lymph nodes. These lymph nodes serve as a hub for immune activation and cell trafficking, which is the regulated movement of cells to different areas of the body via the bloodstream or lymphatic system. From the mesenteric lymph nodes, immune cells can travel to other lymphoid organs, such as the spleen and thymus, where they can influence immune responses throughout the body.

The gut immune system also communicates with the rest of the body's immune system through the production of cytokines and other signaling molecules. These send signals that directly affect what other cells do. However, dysregulation of these communication systems can lead to immune-mediated diseases, such as IBD, in which the dysregulation of innate immune responses can lead to chronic inflammation and tissue damage.

Human studies have shed some light on how the early gut microbiome—during the first few years of life—shapes the immune system. For example, research has shown that the gut microbiome is essential for the maturation of immune cells, particularly T cells, which are crucial for adaptive immune responses. In a study of infants, the presence of specific bacterial species in the gut was associated with an increase in the number of T cells and their maturation markers, suggesting a role for the gut microbiome in T cell maturation.

The gut microbiome also plays a role in regulating innate immune responses. Studies have shown that specific bacterial strains can activate innate immune cells, such as dendritic cells, macrophages, and natural killer cells, promoting immune activation and inflammation. This early exposure to microbial stimuli helps to shape the innate immune response and provides protection against later infections.



In a mature immune system, regulatory T cells (or Tregs) play a critical role in immune tolerance, preventing harmful immune responses to harmless antigens. Studies have shown that the gut microbiome is essential for the development and maturation of Tregs and that specific bacterial strains can induce the generation of Tregs. This early exposure to microbial stimuli helps the immune system learn to distinguish between pathogenic antigens—like a bacterial infection—and beneficial microbes. Early microbial exposure also helps the immune system learn to recognize and tolerate the body's own cells.

The gut microbiome also influences the production of antibodies, which play a crucial role in adaptive immune responses. Studies have shown that infants with a diverse gut microbiome have higher levels of antibody-producing B cells, suggesting that the gut microbiome supports the development of adaptive immune responses. Microbes can also help regulate the immune system with SCFAs. These are produced when gut bacteria ferment dietary fiber; they have immune-modulating properties and can regulate the balance between pro- and anti-inflammatory responses.

However, the relationship between the microbiome and the immune system isn't always perfectly functional or even positive. In molecular mimicry, microbial antigens that are similar to human antigens can sometimes trigger an autoimmune response. For example, consider multiple sclerosis, a disease in which the immune system attacks the nervous system. There is evidence that gut bacteria can produce antigens that resemble myelin, a component of nerve fibers. These microbial antigens can then trigger immune responses that cross-react with myelin, leading to autoimmune damage.

Immune System "Boosting"

Can you "boost" your immune system by way of your gut microbes? Some influencers recommend letting your children play in the dirt, washing your hands less, or taking certain supplements. Many of these recommendations are based on the "old friends" hypothesis, which suggests that modern lifestyles have sheltered us from exposure to certain microorganisms that were once present in our natural environment. These microbes—the "old friends"—include types of commensal bacteria as well as helminths, a kind

of parasitic worm. Supposedly, immune deficiencies arise and become widespread when we're not exposed to these microorganisms throughout our lives.

Most of the evidence comes from epidemiological studies of people who live in rural or farming communities, where it's more common to be exposed to a wide range of microorganisms. These people tend to have a lower incidence of allergies, asthma, and autoimmune diseases compared to those who live in urban areas. In addition, animal studies have shown that exposure to some types of microorganisms, such as helminths and certain bacteria, can reduce inflammation and improve immune function. Clinical trials have introduced helminths into patients with autoimmune disorders, such as multiple sclerosis and IBD—and these little parasites might have helped. Symptoms sometimes did improve.

However, not all of the available evidence supports the "old friends" hypothesis. There is an association between rural living and lower incidence of chronic inflammatory diseases, but it may be confounded by other factors, such as diet and lifestyle. Exposure to certain microorganisms may increase the risk of infectious diseases and other health problems. And while some animal and clinical studies have shown that exposure to certain microorganisms can improve immune function, the specific mechanisms are still unclear.

Plenty of products claim to have immune-boosting properties, but boosting the immune system is a myth. First of all, what is meant by "boosting" the immune system? Are you just amplifying the body's immune response? If so, be careful. A hyperactive immune system can lead to autoimmune diseases, allergies, and chronic inflammation. Instead of focusing on "boosting" the immune system, it is more important to maintain a healthy lifestyle, including a balanced diet, regular exercise, adequate sleep, and stress reduction.

Diet is one of the most important lifestyle factors that can influence the gut microbiome and the immune system. A diet high in fiber and plant-based foods can promote the growth of beneficial bacteria in the gut. In turn, the beneficial bacteria can fend off pathogens and help regulate your normal immune function by interacting with your immune cells. Bacteria also create immune-regulating SCFAs as a byproduct when they ferment fiber.

READING

Belkaid, Y., and T. W. Hand. "Role of the Microbiota in Immunity and Inflammation." *Cell* 157, no. 1 (2014): 121–141. https://doi.org/10.1016/j.cell.2014.03.011.

Berer, K., and G. Krishnamoorthy. "Microbial View of Central Nervous System Autoimmunity." *FEBS Letters* 588, no. 22 (2014): 4207–4213. https://doi.org/10.1016/j.febslet.2014.04.007.

Borody, T. J., E. F. Warren, S. Leis, R. Surace, O. Ashman, and S. Siarakas. "Bacteriotherapy Using Fecal Flora: Toying with Human Motions." *Journal of Clinical Gastroenterology* 38, no. 6 (2004): 475–483. https://doi.org/10.1097/01.mcg.0000128988.13808.dc.

Frank, D. N., A. L. St. Amand, R. A. Feldman, E. C. Boedeker, N. Harpaz, and N. R. Pace. "Molecular-Phylogenetic Characterization of Microbial Community Imbalances in Human Inflammatory Bowel Diseases." *Proceedings of the National Academy of Science*s 104, no. 34 (2007): 13780–13785. https://doi.org/10.1073/pnas.0706625104.

Gensollen, T., and R. S. Blumberg. "Correlation between Early-Life Regulation of the Immune System by Microbiota and Allergy Development." *Journal of Allergy and Clinical Immunology* 139, no. 4 (2017): 1084–1091. https://doi.org/10.1016/j.jaci.2017.02.011.

Mowat, A. M., and W. W. Agace. "Regional Specialization within the Intestinal Immune System." *Nature Reviews Immunology* 14, no. 10 (2014): 667–685. https://doi.org/10.1038/nri3738.

Neurath, M. F. "Targeting Immune Cell Circuits and Trafficking in Inflammatory Bowel Disease." *Nature Immunology* 20, no. 8 (2019): 970–979. https://doi.org/10.1038/s41590-019-0415-0.

Rook, G. A. "Regulation of the Immune System by Biodiversity from the Natural Environment: An Ecosystem Service Essential to Health." *Proceedings of the National Academy of Sciences* 110, no. 46 (2013): 18360–18367. https://doi.org/10.1073/pnas.1313731110.

Vangay, P., A. J. Johnson, T. L. Ward, G. A. Al-Ghalith, R. R. Shields-Cutler, B. M. Hillmann, S. K. Lucas, et al. "US Immigration Westernizes the Human Gut Microbiome." *Cell* 175, no. 4, (2018): 962–972. https://doi.org/10.1016/j.cell.2018.10.029.

7

The Microbiome and Metabolic Health

Early research suggested that a person's gut microbes could predispose them to developing obesity. For a short time, researchers used the ratio of two bacterial phyla, Firmicutes and Bacteroidetes, as a biomarker for obesity and other metabolic diseases; a lower F/B ratio was assumed to predict obesity. The F/B ratio was based on a groundbreaking study that used FMT from rodents or humans with obesity to germ-free rodent recipients, which rapidly developed obesity after the transplant. These studies strongly suggested that the microbiome played a causal role in weight gain. But this ratio was quickly revealed to be unreliable, in part because a phylum contains hundreds or thousands of microbial species that all behave differently. Improved study methods and designs have since shown that the trillions of microorganisms living inside your gut play a crucial role in weight regulation and metabolism, as you'll learn in this lecture.

Metabolic Health

Metabolic health refers to the body's ability to regulate blood glucose levels, maintain healthy lipid profiles, and effectively utilize energy through a series of complex chemical reactions that are often referred to as metabolism. A metabolically healthy person will have metabolic flexibility, or the capacity to switch between different fuel sources—like fats or carbohydrates—for energy. For example, during high-intensity exercise, your muscles rely mostly on glucose as an energy source because it can be broken down rapidly. While you're sleeping, however, your body can rely more on slowly metabolizing its stored fats for energy and reserve the glucose for more urgent energy needs.

Metabolic disease refers to a group of disorders caused by abnormalities in these chemical reactions. Type 2 diabetes, for example, occurs when the body's cells become resistant to the hormone insulin or when the pancreas can't produce enough insulin to keep up with the incoming glucose. When this happens, glucose can't be transported from the blood into the cells, which leads to elevated blood sugar. If left untreated, type 2 diabetes can lead to other health problems, like nerve or kidney damage.

So, what role does the microbiome play in metabolic health? One interesting finding has to do with energy balance. This is the difference between the energy your body absorbs from food and the energy it expends in the operations of day-to-day life: moving, pumping blood, thinking, and so on. Put simply, energy balance is a function of energy in versus energy out. Some relatively recent studies have lead researchers to believe that the microbiome factors into this metabolic equation.

Short-Chain Fatty Acids

A large portion of your gut microbes ferment carbohydrates to harness energy for themselves. In the process, they produce SCFAs, such as butyrate, acetate, and propionate. The SCFAs produced by this process can then be metabolized by the human body as a form of caloric energy. This is called energy harvesting. Gut microbes convert one source of energy that isn't accessible

to humans (like the calories stored in dietary fiber) into a source that can be used by the human host (like butyrate.) This essentially makes more calories available from the diet, which can impact energy balance.

Research in this area is still new, however, and it's unclear whether the conversion of fiber to SCFAs always results in higher energy absorption, as some of those SCFAs can be excreted in feces. In other words, microbes might make more energy available through SCFAs, but the body may or may not absorb that energy. Also, there isn't a clear correlation between fecal or circulating SCFA content and body weight. So, even if your microbiome is particularly active when it comes to energy harvesting, it doesn't mean that you'll be predisposed to weight gain or have difficulty losing weight.

The body also uses these SCFAs in varying ways. SCFAs can be absorbed like dietary fats, but they can also bind to receptors on intestinal cells. Unlike larger fatty acids, they can circulate freely in the bloodstream and cross the blood-brain barrier. This allows them to bind to receptors on a variety of cells, including muscle and fat cells, where they act more like chemical messengers than absorbable nutrients.

Although the mechanisms aren't completely understood and much of the research is still limited to rodent models, there are a few ways SCFAs have been shown to regulate metabolism, hunger, and appetite. First, they can affect how quickly food travels through the digestive tract. This so-called transit time can impact energy balance because if food passes more quickly through your digestive system, you will tend to absorb fewer nutrients. SCFAs can also modify the release of hunger and satiety hormones, and they influence the production of thyroid hormones, which regulate metabolism. They can even act on areas of the brain, such as the hypothalamus, to reduce appetite or food reward, which is the pleasure we experience when eating something tasty.

Researchers are still a long way from implicating any specific microbes or a specific "blend" of SCFAs necessary to assert control over hunger or appetite. But one recent study has shed some light on how much energy the gut microbiome might harvest based on someone's diet. Researchers placed a group of healthy adults into a metabolic chamber where they could tightly control and measure their energy balance, including the energy lost in their

feces. Each participant ate either a Westernized diet or a minimally processed, fiber-rich diet for 6 days. Then, they took a 2-week break. When they returned, participants switched to the other diet and followed that for 6 more days. Each diet provided enough calories for weight maintenance, and all of the macronutrient content was matched, except fiber (which was higher in the minimally processed diet).

Researchers found that the participants' microbes harvested more energy from the minimally processed diet. In other words, the microbes produced more SCFAs on the minimally processed diet compared to the Westernized diet. This made sense considering that the gut microbes had access to more dietary fiber. However, more energy was lost in feces when the participants followed the minimally processed diet. Although they produced and absorbed more SCFAs, they were in a slight caloric deficit of about 116 calories per day, on average (range: 60 to 172 calories). While following the Westernized diet, however, they absorbed nearly all of the available energy. Also, participants lost more weight after following the minimally processed diet compared to the Westernized diet (0.6 kilograms versus roughly 130 grams, respectively).

Many energy-harvesting microbes are considered beneficial (or neutral), and SCFAs provide several different benefits. For instance, it's believed that they have anti-inflammatory effects, support the immune system, and promote heart health. So, you shouldn't think of energy harvesting as a harmful process. It's simply another factor to consider if you're seeing some unexpected results based on your expected energy balance.

The Microbiome and Weight

While the gut microbiome doesn't predict weight loss (or weight gain), there is some evidence that it could play a role in the common phenomenon of weight regain. Much of this evidence has come from studying changes in the microbiomes of people who have lost weight by following a weight loss diet. These studies often reveal that dietary changes don't totally reshape the gut microbiome. A person's gut microbiome is relatively stable through adulthood.

One study investigated changes in the microbiome immediately after a period of weight loss. Researchers theorized that the temporary gut microbiome of the post-weight loss phase might help the participants maintain their weight loss. They used an autologous fecal transplant, treating participants with their own post-weight-loss fecal samples for 6 months after completing their weight loss intervention. But this only helped to maintain the new, lower body weight in the participants whose microbiomes changed significantly during weight loss.

There's more evidence that a microbiome's baseline composition *doesn't* affect your ability to maintain weight loss. Rather, weight loss might depend more on the microbiome's day-to-day variability and its responsiveness to dietary changes. In one study, researchers gave participants one of two diets to follow: one low fat and the other low carbohydrate. The weight loss results varied depending on how responsive or variable each participant's microbiome was. A low-fat diet led to more weight loss in participants whose microbiomes varied a lot day to day while they were following the diet. Those on a low-carbohydrate diet, though, tended to lose more weight if their microbiome responded strongly to the new diet, changing a lot initially but then remaining stable day to day. In other words, weight loss maintenance reflected both how dynamic or stable the microbiome was and the diet that the participants were on. But no one's baseline microbiome stood out as particularly predictive of weight loss maintenance.

When it comes to metabolism, your gut microbiome can affect more than just weight regulation. Certain gut microbes also release an endotoxin called lipopolysaccharide (LPS). Sometimes, elevated levels of LPS enter the bloodstream, causing a condition called metabolic endotoxemia. When endotoxins enter the bloodstream, they can activate the immune system, triggering an inflammatory response. This inflammation has been linked to various health problems, including insulin resistance, type 2 diabetes, obesity, cardiovascular disease, and nonalcoholic fatty liver disease. In addition, excessive exposure to endotoxins can overwhelm the liver's detoxification pathways, leading to liver damage and dysfunction.

Metabolic endotoxemia has been linked to a high-fat, high-sugar diet and increased intestinal permeability, sometimes referred to as "leaky gut." A more permeable intestine can allow more LPS to enter the bloodstream,

which raises the risk for metabolic endotoxemia. And intestinal permeability is associated with several of the health problems mentioned previously. However, scientists still don't know whether the intestinal permeability is a cause or consequence of the disease or if there is perhaps some feedback loop perpetuating each one. In any case, you'll get the greatest benefits from eating a minimally processed, plant-forward diet and limiting alcohol intake.

READING

Brüssow, H. "Problems with the Concept of Gut Microbiota Dysbiosis." *Microbial Biotechnology* 13, no. 2 (2020): 423–434. https://doi.org/https://doi.org/10.1111/1751-7915.13479.

Cani, P. D., E. Moens de Hase, and M. van Hul. (2021). "Gut Microbiota and Host Metabolism: From Proof of Concept to Therapeutic Intervention." *Microorganisms* 9, no. 6 (2021). https://doi.org/10.3390/microorganisms9061302.

Corbin, K. D., E. A. Carnero, B. Dirks, D. Igudesman, F. Yi, A. Marcus, et al. "Host-Diet-Gut Microbiome Interactions Influence Human Energy Balance: A Randomized Clinical Trial." *Nature Communications* 14 (2023): 3161. https://doi.org/10.1038/s41467-023-38778-x.

Fragiadakis, G. K., H. C. Wastyk, J. L. Robinson, E. D. Sonnenburg, J. L. Sonnenburg, and C. D. Gardner. "Long-Term Dietary Intervention Reveals Resilience of the Gut Microbiota Despite Changes in Diet and Weight." *American Journal of Clinical Nutrition* 111, no. 6 (2020): 1127–1136. https://doi.org/10.1093/ajcn/nqaa046.

Gong, J., Y. Shen, H. Zhang, M. Cao, M. Guo, J. He, B. Zhang, et al. "Gut Microbiota Characteristics of People with Obesity by Meta-Analysis of Existing Datasets." *Nutrients* 14, no. 14 (2022): 2993. http://dx.doi.org/10.3390/nu14142993.

Grembi, J. A., L. H. Nguyen, T. D. Haggerty, C. D. Gardner, S. P. Holmes, and J. Parsonnet. "Gut Microbiota Plasticity Is Correlated with Sustained Weight Loss on a Low-Carb or Low-Fat Dietary Intervention." *Scientific Reports* 10, no. 1 (2020): 1405. https://doi.org/10.1038/s41598-020-58000-y.

Lahtinen, P., A. Juuti, M. Luostarinen, L. Niskanen, T. Liukkonen, J. Tillonen, J. Kössi, et al. "Effectiveness of Fecal Microbiota Transplantation for Weight Loss in Patients with Obesity Undergoing Bariatric Surgery: A Randomized Clinical Trial." *JAMA Network Open* 5, no. 2 (2022): e2247226. https://doi.org/10.1001/jamanetworkopen.2022.47226.

Ojo, O., Q. Q. Feng, O. O. Ojo, and X. H. Wang. "The Role of Dietary Fibre in Modulating Gut Microbiota Dysbiosis in Patients with Type 2 Diabetes: A Systematic Review and Meta-Analysis of Randomised Controlled Trials." *Nutrients* 12, no. 11 (2020): 3239. https://doi.org/10.3390/nu12113239.

Perna, S., Z. Ilyas, A. Giacosa, C. Gasparri, G. Peroni, M. A. Faliva, C. Rigon, et al. "Is Probiotic Supplementation Useful for the Management of Body Weight and Other Anthropometric Measures in Adults Affected by Overweight and Obesity with Metabolic Related Diseases? A Systematic Review and Meta-Analysis." *Nutrients* 13, no. 2 (2021): 666. https://doi.org/10.3390/nu13020666.

Rinott, E., I. Youngster, A. Y. Meir, G. Tsaban, A. Kaplan, H. Zelicha, E. Rubin, et al. "Autologous Fecal Microbiota Transplantation Can Retain the Metabolic Achievements of Dietary Interventions." *European Journal of Internal Medicine* 92 (October 2021): 17–23. https://doi.org/10.1016/j.ejim.2021.03.038.

8

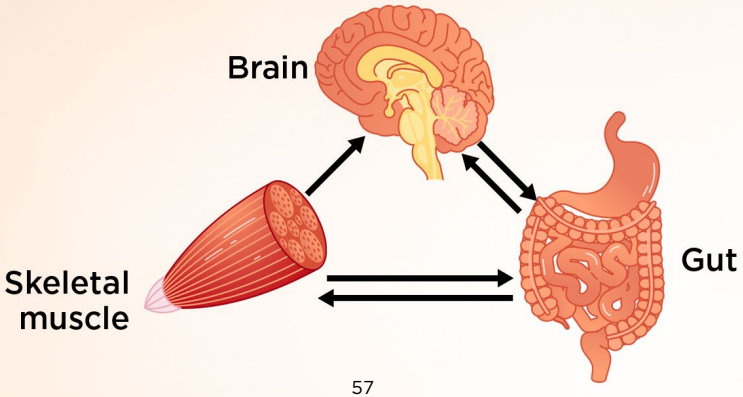
The Gut-Muscle Axis and Exercise

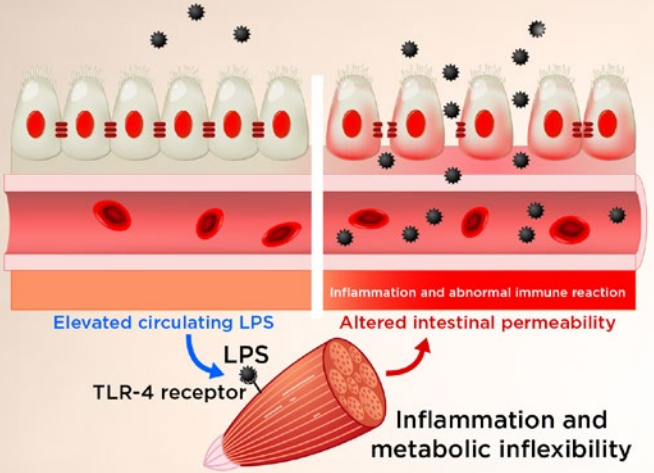
You might think that the benefits of exercise are all about building muscle, shedding fat, and boosting your cardiovascular health. But did you know that your gut bacteria might also be getting in on the action? Recent studies have shown that the trillions of microorganisms in your gut can play a crucial role in how your body responds to physical activity, influencing everything from energy levels to recovery time. In this lecture, you'll discover the surprising ways in which exercise and the gut microbiome are intertwined.

The Gut-Muscle Axis

Intense physical activity exerts mechanical and chemical stressors on the GI tract. Exercise directs blood flow to working muscles and increases activity in the sympathetic nervous system. In turn, these changes modulate the pH, oxygen levels, and nutrient availability in the gut. Because the microbiota play integral roles in nutrient assimilation, immune activity, and metabolic flexibility, they are an obvious target for research into exercise performance. The gut-muscle axis—the bidirectional communication between the gut microbiome and the skeletal muscle system—is a fairly recent discovery in microbiome science. It's through this pathway that the microbiome could influence muscle metabolism, age-related muscle loss, and even athletic performance.

Metabolic endotoxemia is one link between the microbiome and muscle metabolism. Remember that this refers to the chronic, low-grade inflammation linked to endotoxins from certain microbes, particularly an overabundance of LPS in the blood. When LPS binds to immune receptors on skeletal muscle cells, it starts an inflammatory cascade, which may lead to metabolic inflexibility and mitochondrial dysfunction. In other words, gut microbes can produce endotoxins that cause skeletal muscle to become both insulin resistant and worse at breaking down fats for energy, meaning that more glucose remains in circulation, and fats are stored in the muscle cell instead of fat cells. These abnormalities are linked to metabolic diseases like type 2 diabetes.





Human studies have also investigated the relationship between the gut microbiome and muscle function in older adults, who will begin to experience sarcopenia, or age-related muscle loss, around the age of 50. Interestingly, older adults also lose microbial diversity over time, but certain microbes, like *Akkermansia muciniphila*, have been linked to better muscle function and physical performance in this population. So, both muscle metabolism and age-related muscle loss and performance seem to have a relationship with the microbiome.

A recent systematic review examined the impact of exercise on the gut microbiome in various species, including rodents, dogs, horses, and humans. Despite the lack of standardization of diet, training methodology, or microbial analysis in these studies, some trends emerged. In the rodent studies, the effects of exercise were examined using diets ranging from 25% to 60% calories from fat in models of health, obesity, type 2 diabetes, cardiovascular disease, and metabolic syndrome, both with and without control groups.

In a forced swim test, mice with a complete microbiome had better endurance than those with a single beneficial strain, which in turn performed better than those with no bacteria. Forced treadmill running either had no effect on the microbiome or marginally increased or decreased the abundance of some strains.

However, the caveat here is the amount of intense exercise that the mouse does. Compared to a mouse's voluntary activity, the volume of forced treadmill running is extremely low. Voluntary wheel running studies have shown an increase in bacteria that produce the beneficial SCFA butyrate. Differing microbial responses to exercise may also depend on age, as older rodents exhibit less change in diversity after exercise compared to younger rodents.

Observational studies of people across the spectrum of physical activity—from recreational exercisers to competitive athletes—have shown that those with higher levels of fitness also have more diverse microbiomes enriched with beneficial butyrate-producing microbes. While human intervention studies are limited, the majority have focused on endurance exercise. These studies have observed changes from baseline, including variations in several taxa and gene expression after completion of a marathon. In longer-term studies, adding an hour of cycling, walking, boat racing, or concurrent endurance and resistance training resulted in heterogeneous results. This suggests that the type and intensity of exercise might also exert different effects on the gut microbiome, but no clear patterns have emerged.

Both cardio and resistance training offer benefits to gut health, albeit through different mechanisms, and the effects of cardio are probably more direct. Cardiovascular exercise has been shown to increase gut microbiome diversity, while resistance training can affect muscle mass, which in turn impacts metabolic pathways related to gut health. That being said, the majority of research on exercise and the gut microbiome has focused on aerobic activity. At the moment, researchers don't know what effect resistance training has on its own.

Bacteria and Exercise

When it comes to overall health, the American College of Sports Medicine recommends at least 150 minutes of moderate aerobic exercise and at least 2 days of resistance training per week. Intervention studies have also provided some evidence that the microbiome you have when you start a new exercise routine might influence how it changes and could even influence how much your muscles respond to the training.

Currently, a few groups of bacteria have been consistently linked with exercise, including species found within the *Prevotella*, *Akkermansia*, *Lactobacillus*, *Lachnospiraceae*, and *Ruminococcus* groups. These groups appear to be either enriched in physically active individuals, associated with cardiovascular fitness, or increased after an exercise intervention. However, evidence suggests that these changes are transient. Within a few weeks after completing the exercise intervention, the number of these species return to the baseline.

A key question is whether the microbiome influences exercise performance—or the other way around. Rodent intervention studies could suggest the directionality of this dynamic. Earlier, you saw that mice with a complete microbiome fared much better during forced endurance tests compared to those that had no gut bacteria or a single beneficial strain.

Food, Your Microbiome, and Exercise

Additionally, a relationship between diet, the microbiome, and exercise has emerged in recent years based on data examining the dietary patterns and microbial profiles of endurance and resistance-trained athletes. Most athletes consume a diet higher in carbohydrates and protein compared to sedentary people, so it is a challenge to determine the effect of diet or exercise alone. But some studies have shown that exercise and dietary fiber may have the greatest impact on the gut microbiome when they're provided simultaneously. It's possible that, without a proper diet, exercise may not affect the microbiome much at all.

For example, in one study, bodybuilders whose diets were fiber deficient had microbiomes that resembled those of sedentary participants, while those eating adequate fiber had more diverse microbiomes. Observational data have also illustrated an inverse relationship between dietary protein intake and overall diversity in endurance athletes—the more protein, the less diversity, although the lower diversity could also have been attributed to low fiber intake. These findings hint that a fiber-deprived microbiome may not have the capacity to respond to an exercise intervention because many of the microbes associated with cardiovascular fitness rely heavily on fermenting fiber to produce energy.

Soluble fiber dissolves in water to form a gel-like substance and helps lower cholesterol and stabilize blood sugar levels. It's also the preferred energy source for your gut microbes because most soluble fibers are highly fermentable. It's found in various foods, like oats, beans, and lentils. Insoluble fiber doesn't dissolve in water and isn't readily fermentable, but it adds bulk to your stool. It's found in a variety of fruits and vegetables. Both types of fiber are beneficial for gut health, but soluble fiber has a more direct impact on the gut microbiome by serving as a prebiotic. It slows transit time and allows stool to absorb more water, making it easier to pass. Insoluble fiber speeds up the passage of stool, supporting regularity. As for the best way to get fiber, whole foods are generally superior to supplements for their additional nutrients and better bioavailability.

Fiber and Physical Activity

There is some evidence that certain microbes could play a role in exercise performance. More recent studies have combined human studies with rodent fecal transplant models. They've identified microbes of interest in humans and then used FMTs to measure their effects on rodent exercise performance.

In one study, endurance runners were supplemented with alpha-cyclodextrin, which is a carbohydrate similar to resistant starch, one of the favorite foods of gut bacteria. The supplement was linked to improved performance and higher levels of *Bacteroides uniformis*. When researchers transplanted that microbe into mice, their exercise performance improved, too. The researchers believe that the microbe might play a role in glucose metabolism, which makes more fuel available during endurance exercise.

In a similar study, fecal samples were taken from elite runners after they completed a marathon. These runners were found to have higher-than-average levels of the genus *Veillonella* in their stool. Researchers transplanted a strain of *Veillonella* into mice and found that they were able to run longer. It seems that at least some strains in this genus can convert lactate—a product of energy production from glucose—to propionate, which reduces the amount of energy the body needs to clear lactate.

Probiotic supplements that introduce strains of beneficial bacteria haven't shown much promise for improving performance in any sport so far. However, in some cases, they could improve recovery and support an athlete's immune system to prevent them from taking unplanned breaks due to illness. Of course, it's not as simple as exercising your way to a resilient microbiome or shaving minutes off of your mile time with the right probiotic. Rodent models are useful for explaining mechanisms in isolation, but they can't accurately represent the complexity of the human gut microbiome. In addition, most of these studies were observational, so they examined correlation, not causation. And almost none controlled for diet, which often differs between athletes and sedentary people and has a significant impact on the gut microbiome. However, the findings do provide support for the following statement: Fiber and physical activity are important both for you and your gut microbiome.

READING

Bycura, D., A. C. Santos, A. Shiffer, S. Kyman, K. Winfree, J. Sutliffe, T. Pearson, et al. "Impact of Different Exercise Modalities on the Human Gut Microbiome." *Sports* 9, no. 2 (2021): 14. https://doi.org/10.3390/sports9020014.

Clauss, M., P. Gérard, A. Mosca, and M. Leclerc. "Interplay between Exercise and Gut Microbiome in the Context of Human Health and Performance." *Frontiers in Nutrition* 8 (2021). https://doi.org/10.3389/fnut.2021.637010.

Estaki, M., J. Pither, P. Baumeister, J. P. Little, S. K. Gill, S. Ghosh, Z. Ahmadi-Vand, et al. "Cardiorespiratory Fitness as a Predictor of Intestinal Microbial Diversity and Distinct Metagenomic Functions." *Microbiome* 4 (2016): 42. https://doi.org/10.1186/s40168-016-0189-7.

Jäger, R., A. E. Mohr, K. C. Carpenter, C. M. Kerksick, M. Purpura, A. Moussa, J. R. Townsend, et al. "International Society of Sports Nutrition Position Stand: Probiotics." *Journal of the International Society of Sports Nutrition* 16, no. 1 (2019): 62. https://doi.org/10.1186/s12970-019-0329-0.

Mitchell, C. M., B. M. Davy, M. W. Hulver, A. P. Neilson, B. J. Bennett, and K. P. Davy. "Does Exercise Alter Gut Microbial Composition? A Systematic Review." *Medicine & Science in Sports & Exercise* 51, no. 1 (2019): 160–167. https://doi.org/10.1249/MSS.0000000000001760.

Morita, H., C. Kano, C. Ishii, N. Kagata, T. Ishikawa, A. Hirayama, Y. Uchiyama, et al. "*Bacteroides uniformis* and Its Preferred Substrate, α-cyclodextrin, Enhance Endurance Exercise Performance in Mice and Human Males." *Science Advances* 9, no. 4 (2023): eadd2120. https://doi.org/10.1126/sciadv.add2120.

Rodriguez, J., A. M. Neyrinck, M. van Kerckhoven, M. A. Gianfrancesco, E. Renguet, L. Bertrand, P. D. Cani, et al. "Physical Activity Enhances the Improvement of Body Mass Index and Metabolism by Inulin: A Multicenter Randomized Placebo-Controlled Trial Performed in Obese Individuals." *BMC Medicine* 20 (2022): 110. https://doi.org/10.1186/s12916-022-02299-z.

Scheiman, J., J. M. Luber, T. A. Chavkin, T. MacDonald, A. Tung, L.-D. Pham, M. C. Wibowo, et al. "Meta-omics Analysis of Elite Athletes Identifies a Performance-Enhancing Microbe That Functions via Lactate Metabolism." *Nature Medicine* 25 (2019): 1104–1109. https://doi.org/10.1038/s41591-019-0485-4.

Son, J., L. G. Jang, B. Y. Kim, S. Lee, and H. Park. "The Effect of Athletes' Probiotic Intake May Depend On Protein and Dietary Fiber Intake." *Nutrients* 12, no. 10 (2020): 2947. https://doi.org/10.3390/nu12102947.

Torquati, L., T. Gajanand, E. R. Cox, C. R. G. Willis, J. Zaugg, S. E. Keating, and J. S. Coombes. "Effects of Exercise Intensity on Gut Microbiome Composition and Function in People with Type 2 Diabetes." *European Journal of Sport Science* 23, no. 4 (2023): 530–541. https://doi.org/10.1080/17461391.2022.2035436.

9

The Gut-Brain Axis and Mood

You might be wondering what the brain has to do with gut health. Have you ever felt butterflies in your stomach when you're nervous? Perhaps your stomach feels upset when you're stressed. Believe it or not, your gut and brain are closely connected, and this connection is known as the gut-brain axis, which involves a complex network of signals and pathways that facilitate two-way communication between the GI tract and the central nervous system. In this lecture, you'll explore the intricacies of this connection and its importance to bodily functions and mental health.

Your Nervous System

The nervous system is a complex system of specialized cells that transmits information to many parts of your body, allowing you to coordinate your actions and move. On a basic level, the nervous system is composed of two types of cells: neurons and glial cells. Neurons transmit messages across the body, and glial cells protect and maintain neurons. The "messages" that neurons send are both chemical and electrical, and they are sent to other neurons or cells using special junctions called synapses. The chemical signals that cross these synapses are neurotransmitters. Neurotransmitters, including serotonin and dopamine, aid in a host of biological functions, such as sleep, heart rate, movement, and even thinking.

Your nervous system can be divided into the central nervous system and the peripheral nervous system. Your brain and spinal cord make up your central nervous system. Your brain is like the central control hub for every bodily function—it processes incoming sensory information and sends out instructions to the peripheral nervous system, which covers all of the nervous cells outside of your brain and spinal cord. The main function of the peripheral nervous system is to connect the central nervous system with the rest of the body, like a relay station. It can also be broken down into two parts: the somatic nervous system and the autonomic nervous system.

The somatic nervous system connects your central nervous system to your skeletal muscles, and it's mostly under voluntary control. It also transmits tactile sensory information, like heat, texture, or pain, to your central nervous system. The autonomic nervous system, however, acts largely unconsciously and automatically. It regulates your internal organs to control things like heart rate, breathing, digestion, arousal, and the fight-or-flight response. It can also be separated further into the sympathetic and parasympathetic branches.

The sympathetic branch stimulates the fight-or-flight response, or a physiological reaction to danger. It primes the body to either fight or escape by releasing neurotransmitters that release glucose into the bloodstream and route more blood to the muscles. Some functions, like digestion, are paused when the sympathetic nervous system is active. In contrast, the parasympathetic nervous system is more active during periods of rest and

digestion. Both branches of the autonomic nervous system are always active, but one might be more active depending on what you're doing, like going for a run versus sitting down to eat.

When it comes to gut health, the enteric nervous system is even more fascinating. It is a group of nerves that runs along your digestive tract from your esophagus to your anus, and it's sometimes called the second brain because it can operate independently of your brain and spinal cord. It controls the movement of food through your digestive tract. When you eat, your stomach and intestines need to contract to move the food along. The enteric nervous system coordinates this process by sending signals to the muscles in your digestive tract. Moreover, nerves in your enteric nervous system sense the presence of food in your digestive tract and signal your body to produce the right blends of digestive juices and enzymes. This nervous system can even sense when something is wrong with your digestive tract. For example, if there is an infection, it can send signals to your immune system to help fight it off.

The Gut-Brain Axis

The enteric nervous system, the autonomic nervous system, the gut microbiota, and all of the associated neurotransmitters and hormones combine to form the gut-brain axis. This is a two-way communication highway that connects your digestive system with your brain. It uses various signaling pathways to communicate, including hormones, neurotransmitters, and immune cells. Much of this bidirectional communication travels along the vagus nerve, a major parasympathetic nerve that extends from the brainstem to the abdomen. The vagus nerve transmits signals between the gut and the brain, allowing the brain to regulate gut function and the gut to influence brain activity.

The gut-brain axis plays a potentially important role in many bodily functions. It aids in digestion, regulating the movement of food through the digestive tract and the secretion of digestive enzymes and hormones. And it plays a role in immune response, recruiting immune cells to fight infection and inflammation in the gut. The gut-brain axis also aids in maintaining the balance of the gut microbiome, and it could influence your mood and

emotions. Many neurotransmitters are produced in the gut, some of which can enter the brain, and researchers believe that this is one way the gut microbiota could play a role in mood disorders and mental health.

The co-occurrence of psychiatric and GI symptoms is quite common. About half of patients with a functional GI disorder experience psychiatric symptoms, and a similar percentage of psychiatric patients are diagnosed with IBS. But do gut bacteria play a role in mood disorders? The evidence is inconclusive. Certain types of bacteria produce substances that affect the nervous system, like neurotransmitters and neuromodulators. For example, some bacteria, such as Enterococcus, can produce serotonin. Serotonin derived from the gut plays a role in the movement of the digestive system and can affect how quickly the stomach empties and feelings of fullness. However, researchers have not seen this serotonin cross the blood-brain barrier and directly influence mood.

There's a bit more evidence that the gut-brain axis may play a role in GI distress. This has been researched in patients who suffer from various clinical GI conditions, including IBS and IBD. For example, studies have shown that alterations in the gut microbiome and abnormal enteric nervous system activity could contribute to IBS symptoms like abdominal pain and dysregulated GI motility. There's also some evidence that psychological stress is strongly associated with IBS symptoms.

Recent evidence also indicates that the microbiome could enhance or suppress hunger and appetite via the gut-brain axis and the endocannabinoid system. This system helps regulate intestinal permeability and eating behaviors. Endocannabinoids are lipid-based neurotransmitters produced by the body in response to various physiological signals. They help regulate a wide range of functions, including mood, pain perception, appetite, inflammation, and immune response. Theoretically, if the endocannabinoid system becomes overactive, it could increase the desire for pleasure-driven eating while reducing the appetite-suppressing effects of a hormone called leptin. Simultaneously, it could compromise the integrity of the intestinal barrier, which may contribute to metabolic endotoxemia.

A high-fat diet and obesity have been linked to increased activity of the endocannabinoid system and changes in the gut microbiome. These factors may contribute to hedonic eating (eating for pleasure rather than hunger) and disrupted fat metabolism. Obesity is associated with higher endocannabinoid

levels in the blood and fatty tissues as well as increased receptor expression. Consuming high-fat foods appears to increase endocannabinoid production in the intestines and slow the rate at which endocannabinoids are broken down. Individuals with eating disorders or psychiatric symptoms often exhibit disrupted appetite, disrupted signaling of fullness, and altered eating behaviors.

The Microbiome and Mental Health

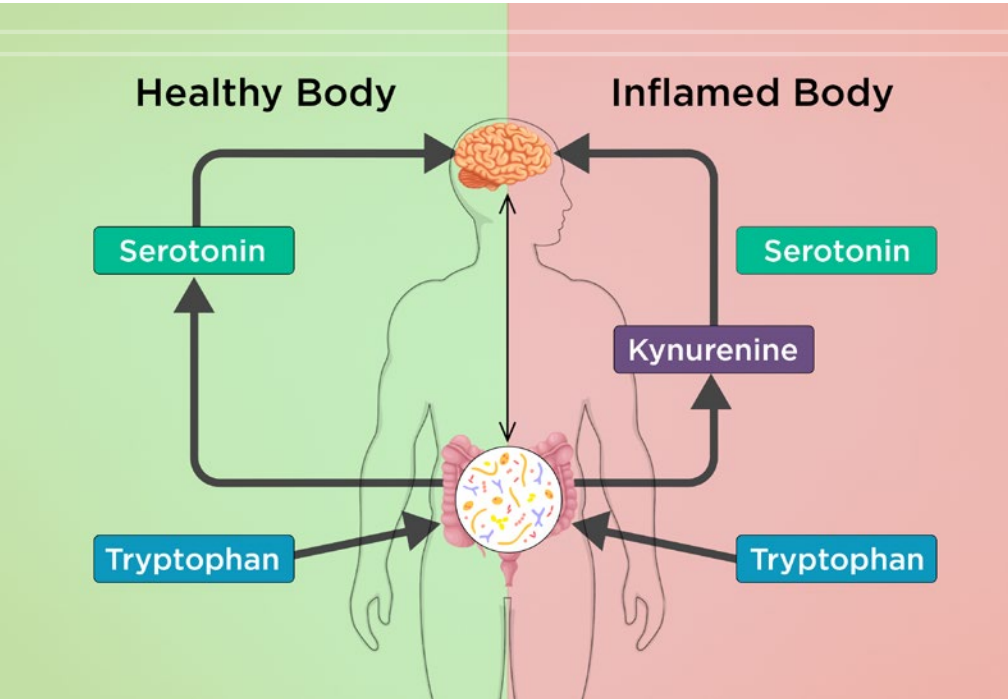
There's some evidence that psychological aspects of disordered eating could influence the microbiota. Specifically, psychological stress, both chronic and experienced early in life, has been shown to reduce microbial diversity in animals.

Research on the microbiome's connection to anxiety and depression is limited, but animal studies have turned up some compelling findings. Scientists have conducted experiments where they transferred fecal matter between mice with different personalities or studied mice that lacked a microbiome altogether. These studies have shown a link between the microbiome and anxiety. In one experiment, researchers transferred fecal matter between anxious and aggressive mice, and their personalities changed accordingly. Aggressive mice became more docile, while anxious mice became more exploratory. This suggests that the microbiome can have a significant influence on mouse behavior, even more so than their genes. Similar results were observed in germ-free mice that received fecal transplants from human donors diagnosed with major depressive disorder. Their behavior changed after the transplant, indicating that the microbiome might also play a role in human mental health.

Studies of mice have also suggested that the microbiome may impact the production of substances that support cognitive function. For example, although the exact mechanisms are unclear, the microbiome might affect levels of brain-derived neurotrophic factor, which is important for brain development, as well as levels of GABA, a neurotransmitter that regulates mood. However, note that the data from rodent studies are limited in their application to human biology. The germ-free mice used do not develop fully functioning immune or nervous systems, making them inadequate models for human physiology.

Recent studies have shown that the gut microbiome can influence the function of the kynurenine pathway, which plays an important role in the metabolism of tryptophan. Tryptophan is an essential amino acid obtained from our diet. The body metabolizes it to produce various molecules, including proteins, neurotransmitters, and other neuroactive compounds, such as quinolinic acid, which has an excitatory effect, or kynurenic acid, which can reduce signaling in the brain.

Changes in the kynurenine pathway could have significant implications for mood and mental health. For example, such alterations could reduce the availability of serotonin, which plays a crucial role in regulating mood. Tryptophan is also a precursor for serotonin, and when tryptophan is diverted toward the kynurenine pathway instead of toward serotonin synthesis, it can lead to reduced serotonin levels, which are often observed in individuals with depression. Certain bacteria in the gut have the ability to break down tryptophan and produce metabolites that can influence the activity of enzymes in the kynurenine pathway.



The Microbiome and Medications

Antidepressants are among the top three most prescribed and used therapeutic drug classes. While there's limited research on the effects of antidepressants on the gut microbiota, some preclinical studies are available. For example, one study looked at selective serotonin reuptake inhibitors (SSRIs)—one of the most commonly prescribed antidepressants. When researchers orally administered SSRIs to animals, they found that they reduced overall microbial diversity and decreased levels of specific taxa. Similar evidence has been noted for tricyclic antidepressants in animal models.

However, some observational studies in humans have provided opposing results regarding the effects of antipsychotic medications. For instance, one study observed higher indices of diversity in patients taking the drug risperidone, whereas another study found that diversity was lower in patients taking other forms of second-generation antipsychotic medications. Conflicting results like these leave researchers with no clear relationship between the use of antipsychotic drugs, microbial changes, and treatment outcomes or side effects. Moreover, some of these changes appear to be gender specific, often affecting females significantly more than males, which adds an extra layer of complexity.

There is some evidence that the gut microbiota can alter the effects of certain neuromodulatory drugs. For instance, when Levodopa (L-dopa) is taken orally, certain gut bacteria can convert it to a compound that isn't neuroactive. These microbial actions are thought to reduce the concentration of L-dopa that is absorbed and thus possibly alter its therapeutic properties.

Note that these studies are limited by their small study sizes, heterogeneous findings, and different definitions of "dysbiosis." They also rely on stool samples, which can't represent the entire gut microbiome. At most, it can be said that these psychotropic and neuromodulatory drugs interact with the microbiota in a bidirectional manner: The drugs seem to have some impact on the microbiome, and the microbiome seems to have some impact on the drugs' effectiveness. This two-way interaction likely accounts for some of the variability of both the microbiome and drug efficacy between individuals.

READING

Almeida, C., R. Oliveira, R. Soares, and P. Barata. "Influence of Gut Microbiota Dysbiosis on Brain Function: A Systematic Review." *Porto Biomedical Journal* 5, no. 2 (2020): 1–8. https://doi.org/10.1097/j.pbj.0000000000000059.

Clark, A., and N. Mach. "Exercise-Induced Stress Behavior, Gut-Microbiota-Brain Axis and Diet: A Systematic Review for Athletes." *Journal of the International Society of Sports Nutrition* 13, no. 1 (2016): 1–21. https://doi.org/10.1186/s12970-016-0155-6.

Cryan, J. F., and T. G. Dinan. "Mind-Altering Microorganisms: The Impact of the Gut Microbiota on Brain and Behaviour." *Nature Reviews Neuroscience* 13, no. 10 (2012): 701–712. https://doi.org/10.1038/nrn3346.

Fond, G., W. Boukouaci, G. Chevalier, A. Regnault, G. Eberl, N. Hamdani, F. Dickerson, et al. "The 'Psychomicrobiotic': Targeting Microbiota in Major Psychiatric Disorders: A Systematic Review." *Pathologie Biologie* 63, no. 1 (2015): 35–42. https://doi.org/10.1016/j.patbio.2014.10.003.

Furness, J. B. "The Enteric Nervous System and Neurogastroenterology." *Nature Reviews Gastroenterology & Hepatology* 9 (2012): 286–294. https://doi.org/10.1038/nrgastro.2012.32.

Lee, C., E. Doo, J. M. Choi, S. Jang, H.-S. Ryu, J. Y. Lee, J. H. Oh, et al. "The Increased Level of Depression and Anxiety in Irritable Bowel Syndrome Patients Compared with Healthy Controls: Systematic Review and Meta-Analysis." *Journal of Neurogastroenterology and Motility* 23, no. 3 (2017): 349–362. https://doi.org/10.5056/jnm16220.

Mayer, E. A., R. Knight, S. K. Mazmanian, J. F. Cryan, and K. Tillisch. "Gut Microbes and the Brain: Paradigm Shift in Neuroscience." *Journal of Neuroscience* 34, no. 46 (2014): 15490–15496. https://doi.org/10.1523/JNEUROSCI.3299-14.2014.

Ng, Q. X., W. Loke, N. Venkatanarayanan, D. Y. Lim, A. Y. S. Soh, and W. S. Yeo. "A Systematic Review of the Role of Prebiotics and Probiotics in Autism Spectrum Disorders." *Medicina* 55, no. 5 (2019): 1–10. https://doi.org/10.3390/medicina55050129.

Rao, M., and M. D. Gershon. "The Bowel and Beyond: The Enteric Nervous System in Neurological Disorders." *Nature Reviews Gastroenterology & Hepatology* 13, no. 9 (2016): 517–528. https://doi.org/10.1038/nrgastro.2016.107.

Rogers, G. B., D. J. Keating, R. L. Young, M. L. Wong, J. Licinio, and S. Wesselingh. "From Gut Dysbiosis to Altered Brain Function and Mental Illness: Mechanisms and Pathways." *Molecular Psychiatry* 21, no. 6 (2016): 738–748. https://doi.org/10.1038/mp.2016.50.

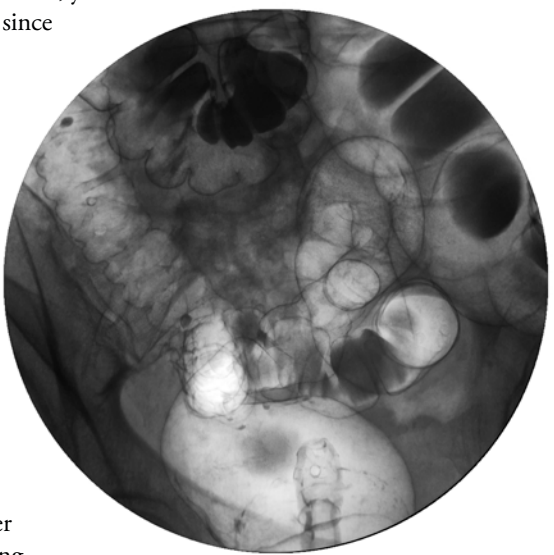
10

Common Digestive Complaints

Perhaps you've noticed that your digestion feels off lately, but you aren't sure what's going on. With so much information—and misinformation—circulating the internet, it can be confusing to pin down the problem. Everyone will experience some form of GI distress from time to time, and it's nothing to feel embarrassed about. Some of the most common symptoms include gas, bloating, and temporary changes in bowel habits. Today, you'll learn about these common GI disruptions: their causes, how to test for them, and how to treat them.

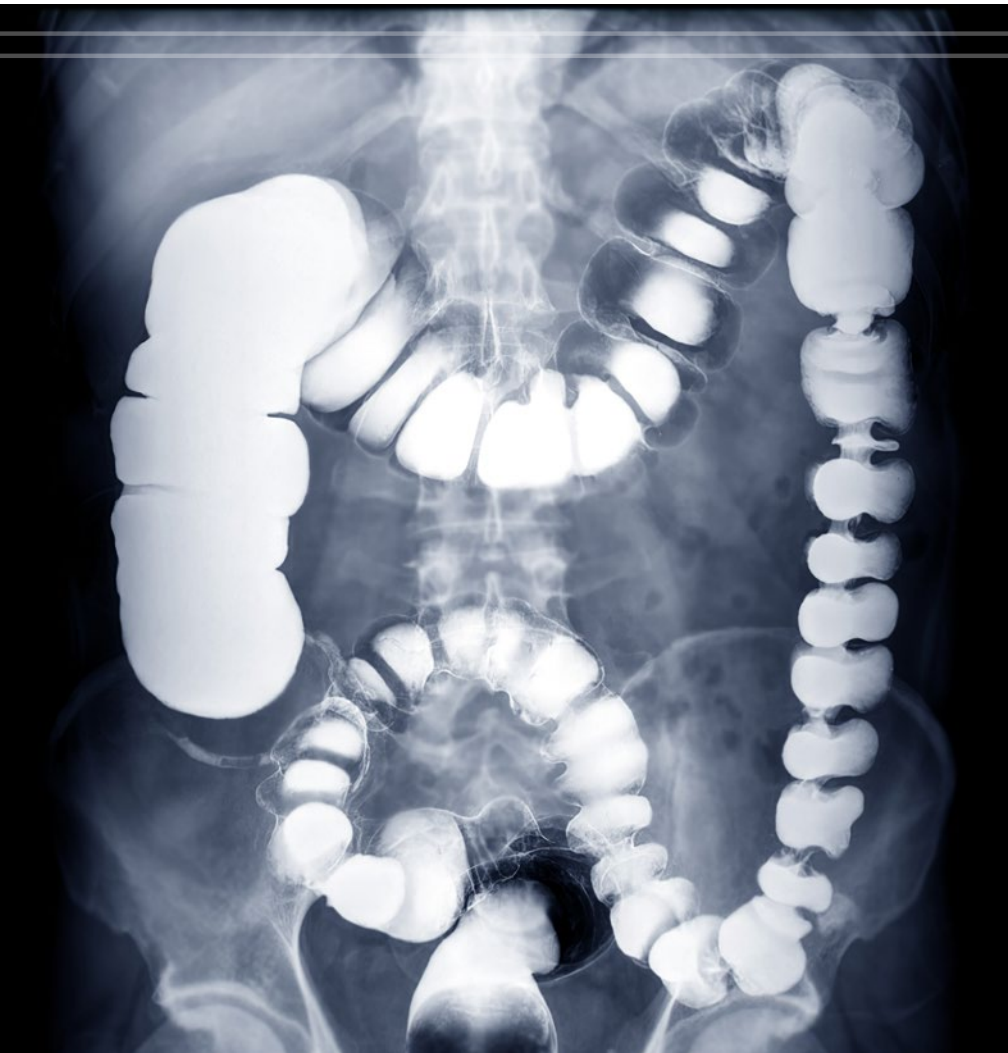
Examining the Gut

Although bouts of GI distress are a normal part of life, there are some red flags that could indicate the possibility of a serious illness. If you're experiencing the following, you should see a doctor: rectal bleeding or blood in the stool; unexplained weight loss or fatigue; persistent abdominal pain; persistent nausea or vomiting; a change in bowel habits (like ongoing constipation or diarrhea) that lasts more than a few weeks; jaundice (a yellowing of the skin and eyes); and anemia. Also, if you have a family history of GI diseases, such as colon cancer or IBS, you could be at increased risk, too, since genetics could also play a role.



Since many GI diseases share a number of symptoms, doctors often have to narrow down the potential causes of your symptoms before arriving at the most likely diagnosis. For that, your doctor may use several available diagnostic and screening methods to determine the health of your GI tract. One method is called an upper GI series. It involves swallowing barium—a fine powder that shows up on x-rays. When mixed with water and swallowed, barium coats the lining of your esophagus, stomach, and the first part of the small intestine. The doctor can use x-ray to visualize these structures and determine if there are any irregularities. This may be followed by imaging of the rest of the small intestine, and in some cases, a "double contrast" medium—like CO_2—is used to show the stomach lining. This is often used if symptoms indicate potential ulcers, tumors, or Crohn's disease of the small intestine.

A lower GI series includes a barium swallow followed by imaging, but it also requires an overnight fast and bowel cleansing. It can also include a double contrast to detect polyps, inflammation, and colon cancer. And recent advances in medical technology have led to the development of the capsule endoscopy, in which an individual swallows a tiny pill-sized camera that travels the entire length of the GI tract, capturing images as it goes.



Your doctor might also conduct stool tests. For example, a fecal occult blood test examines a stool sample for blood that isn't apparent to the naked eye. The blood could indicate polyps, hemorrhoids, ulcers, IBD, or other issues. Your doctor may also test your gallbladder or pancreas since diseases affecting these organs would lead to GI symptoms despite having a healthy digestive tract. They might take images of your gallbladder after you eat a fatty meal or measure the fat lost in your stool.

Breath tests are commonly used to check for small intestine bacterial overgrowth (SIBO). When bacteria ferment specific sugars in your small and large intestines, they produce high levels of methane and hydrogen. Abnormally high levels of these gases in your breath can indicate an overgrowth of bacteria or difficulty in digesting and absorbing these sugars. Keep in mind that the accuracy of these breath tests is limited, with a relatively high risk of producing both false positive and false negative results. Results depend on the dose and type of sugar used, the testing center's reference ranges, and even the patient's stress level. Individuals with IBS are more likely to exhibit positive breath tests, suggesting that bacteria might play a role in the disease. However, these results don't have useful applications in guiding dietary recommendations or predicting patient responses to IBS or SIBO therapies.

Food Intolerance and Allergy

Food intolerances and allergies are both adverse reactions to food, but they differ in their underlying mechanisms and severity. Food allergies involve the immune system and can cause severe and potentially life-threatening symptoms. In an allergic reaction, the immune system mistakenly identifies a food protein as harmful and launches an attack against it. This can trigger symptoms such as hives, swelling, difficulty breathing, and anaphylaxis. Different types of tests can screen for or diagnose allergies, like pricking the skin with a potential allergen. Treating a food allergy sometimes requires strict avoidance of the allergenic food. Peanut allergies are one example that can cause serious reactions and might require emergency treatment.

Food intolerances, however, don't involve the immune system and are usually less severe than allergies. They occur when the body has difficulty digesting a nutrient due to a lack of digestive enzymes. This can cause symptoms such as bloating, gas, diarrhea, constipation, and stomach pain. Lactose intolerance is one common example that leads to GI distress after eating dairy. Food intolerances are often diagnosed through a process of elimination and may require reducing or avoiding the problematic food.

Another source of GI discomfort may be your daily protein shake or low-carb dessert. Many "diet" products claim their diet creds by cutting sugar to or near zero. But a lot of these products still contain sugar alcohols, which are a type of FODMAP. In diet products, these poorly absorbed sugars, like maltitol or sorbitol, are often used in large amounts to substitute for regular sugar. Sugar alcohols can have a strong laxative effect, and some products are even required to disclose this on their label.

Another common cause of gas in many health foods is inulin, a FODMAP found in ingredients like chicory root. It's considered a functional fiber because it's a concentrated, isolated indigestible carbohydrate that's shown some benefit for humans. However, inulin can also cause a lot of GI distress, so check items like protein bars and powders if you think you could be sensitive to it.

And if you drink coffee or alcohol, you could be contributing to your GI distress. Coffee relaxes the lower esophageal sphincter, which can aggravate heartburn. It also leads to the release of hormones like gastrin, which increase intestinal contractions and lead to defecation in at least 30% of the population. Alcohol consumption causes food to move through the GI tract more rapidly, and frequent drinking can also lead to poorer nutrient absorption as well as damage to the intestinal lining, elevated intestinal permeability, and changes to the microbiota.

Medication and GI Issues

Although antibiotics are not typically targeted specifically at the gut microbiota, they still have unavoidable effects when taken orally. Some can reduce the diversity of the microbial population by up to 30%, and these effects can last from 1 month up to 6 years. Antibiotics that target certain types of bacteria can also affect other beneficial or neutral bacteria that may be present. Additionally, antibiotic use can change the metabolism and oxygen levels in the gut, which can impact the survival of the microbial community. The use of certain antibiotics has been associated with infections caused by *C. difficile*. And early or chronic antibiotic use has also been linked to an increased risk of diseases, such as colorectal cancer, IBD, and obesity.

Nonsteroidal anti-inflammatory drugs (NSAIDs) like ibuprofen can cause significant GI side effects, including GI bleeding, inflammation, ulceration, and permeability. The risk increases with long-term use. However, only 1% to 2% of chronic users face serious complications. And while NSAIDs can impact gut microbiota, the relationship is complicated and highly specific to the drug being used.

Some other drugs, like antacids and proton pump inhibitors, modify acidity in the GI tract. While these drugs primarily reduce the acidity of the stomach, they can also have this effect throughout the entire GI tract, especially in the small intestine. Proton pump inhibitor use has been associated with an increased risk of infection with *C. difficile*—a pathogenic bacterium that wreaks havoc on the GI tract. It is also associated with reduced microbial diversity as well as lower levels of certain beneficial bacterial populations, and some evidence suggests that it may increase the risk of developing SIBO.

Laxatives can affect the microbiota, too. The laxative polyethylene glycol (PEG) influences fluid distribution in the intestinal tract, causing a more rapid excretion of stool. Studies on PEG use in rodents have found that it alters various bacterial populations associated with the disruption of the mucus barrier and immune function. Its use is also associated with increased abundance of the genus *Bacteroides*, a mucin-degrading group.

Both human and rodent data provide strong evidence that the diabetes drug metformin significantly affects gut microbiome composition. Some changes are beneficial. For example, metformin can encourage the growth of butyrate-producing bacteria and microbiome-specific regulation of blood glucose levels. However, it is also associated with elevated levels of genes linked to gas production, and it's correlated with higher numbers of potential pathogens and virulence factors that could actually cause diseases.

Managing Common GI Issues

One of the most effective ways to manage gas discomfort is through dietary intervention, or managing the consumption of gas-producing foods. But did you know that low-intensity, rhythmic exercise like walking or cycling can also help to increase gastric motility and relieve gas discomfort? And if your problem is excessive gas, then an over-the-counter drug like simethicone may help by breaking up gas bubbles.

If you're thinking of increasing your fiber intake, it's important to do it gradually and to give your body time to adjust. Aim for 25 to 40 grams of fiber per day. Fiber can help with another common GI complaint: constipation. Some foods, like green vegetables, plain yogurt, and beans, contain specific types of fiber and fermentable carbohydrates that can make stools softer and easier to pass. However, be cautious with supplements like methylcellulose, which might interfere with nutrient absorption and even cause some cramping and diarrhea. Nonfiber osmotic laxatives and stool softeners might be helpful, but stimulant laxatives can often cause painful cramps and disrupt normal nervous signaling.

Regarding diarrhea management, it's important to maintain hydration with fluids that are of equal osmolality—or particle concentration—to the gut. Sports drinks are a common example; they're formulated with the best concentrations of glucose and electrolytes to facilitate absorption and prevent GI distress. Stimulating the GI tract with food is also important to maintain its functionality. Gradually increasing soluble fiber and resistant starch might help thicken the stool, but it's best to limit simple carbohydrates, sugar alcohols, caffeine, and gas-producing foods. If you're dealing with traveler's or antibiotic-associated diarrhea, you might want to consider a yeast-based probiotic called *Saccharomyces boulardii.*

Probiotics

Probiotics have become massively popular as a way to address common GI symptoms. But what's true and what isn't? There are generally three types of studies that look at probiotic interventions: The first involves healthy individuals who have no prior digestive issues, the second involves individuals who have known digestive issues, and the third involves healthy individuals who are given a challenge to induce digestive issues.

While these studies still lack standardized protocols, some probiotics have been consistently effective in reducing GI distress in multiple RCTs. They have been shown to help alleviate constipation and diarrhea associated with antibiotic use as well as IBS, IBDs, and communicable infections. Several have even been helpful in managing serious complications like pouchitis and necrotizing enterocolitis and preventing intestinal infectious diseases in children. However, note that in most cases, probiotics are used in addition to prescribed medications.

Probiotics that have consistently shown positive results include the yeast *S. boulardii,* which has been effective in preventing and treating both adult and pediatric diarrhea associated with travel, antibiotic use, and communicable infections, as well as in preventing *C. difficile* recurrence. However, the usefulness of probiotics for reconstituting the microbiome after antibiotic use or disease is still questionable. Some studies have even suggested that probiotics may delay the reestablishment of the native biome after antibiotic administration.

READING

Edakkanambeth Varayil, J., B. A. Bauer, and R. T. Hurt. "Over-the-Counter Enzyme Supplements: What a Clinician Needs to Know." *Mayo Clinic Proceedings* 89, no. 9 (2014): 1307–1312. https://doi.org/10.1016/j.mayocp.2014.05.015.

Harper, A., M. M. Naghibi, and D. Garcha. "The Role of Bacteria, Probiotics and Diet in Irritable Bowel Syndrome." *Foods* 7, no. 2 (2018): 1–20. https://doi.org/10.3390/foods7020013.

Iriondo-DeHond, A., J. A. Uranga, M. D. Del Castillo, and R. Abalo. "Effects of Coffee and Its Components on the Gastrointestinal Tract and the Brain-Gut Axis." *Nutrients* 13, no. 1 (2020): 88. https://doi.org/10.3390/nu13010088.

Lange, K., M. Buerger, A. Stallmach, and T. Bruns. "Effects of Antibiotics on Gut Microbiota." *Digestive Diseases* 34, no. 3 (2016): 260–268. https://doi.org/10.1159/000443360.

McFarland, L. V. "Use of Probiotics to Correct Dysbiosis of Normal Microbiota Following Disease or Disruptive Events: A Systematic Review." *BMJ Open* 4, no. 8 (2014): e005047. https://doi.org/10.1136/bmjopen-2014-005047.

Miller, L. E., A. C. Ouwehand, and A. Ibarra. "Effects of Probiotic-Containing Products on Stool Frequency and Intestinal Transit in Constipated Adults: Systematic Review and Meta-Analysis of Randomized Controlled Trials." *Annals of Gastroenterology* 30, no. 6 (2017): 1–11. https://doi.org/10.20524/aog.2017.0192.

Reding, K. W., K. C. Cain, M. E. Jarrett, M. D. Eugenio, and M. M. Heitkemper. "Relationship between Patterns of Alcohol Consumption and Gastrointestinal Symptoms among Patients with Irritable Bowel Syndrome." *The American Journal of Gastroenterology* 108, no. 2 (2013): 270–276. https://doi.org/10.1038/ajg.2012.414.

Suez, J., N. Zmora, G. Zilberman-Schapira, U. Mor, M. Dori-Bachash, S. Bashiardes, M. Zur, et al. "Post-Antibiotic Gut Mucosal Microbiome Reconstitution Is Impaired by Probiotics and Improved by Autologous FMT." *Cell* 174, no. 6 (2018): 1406–1423. https://doi.org/10.1016/j.cell.2018.08.047.

Weersma, R. K., A. Zhernakova, and J. Fu. "Interaction between Drugs and the Gut Microbiome." *Gut* 69 (2020): 1510–1519. https://doi.org/10.1136/gutjnl-2019-320204.

Yoon, M. Y., and S. S. Yoon. "Disruption of the Gut Ecosystem by Antibiotics." *Yonsei Medical Journal* 59, no. 1 (2018): 4–12. https://doi.org/10.3349/ymj.2018.59.1.4.

11

Eating for Gut Health

Are you tired of scrolling through endless lists of questionable supplements and trendy diets, all claiming to be the secret to a healthy gut? Scroll no further. In this lecture, based on the latest studies and expert opinions, you will examine the foods and supplements that have been shown to support a thriving gut microbiome. You will also learn about the impacts that even various dietary patterns and eating times can have on your microbiota.

Different Dietary Patterns

Your long-term dietary pattern plays a significant role in both the composition and function of your microbiome. Diet also impacts your exposure to health conditions like metabolic syndrome, which encompasses a group of factors, such as high blood pressure and high cholesterol, that can lead to heart disease, stroke, and diabetes. And it's estimated that a prudent dietary pattern—like the DASH diet—could reduce your risk of colorectal cancer by about 20%. The Mediterranean dietary pattern is also linked to health benefits and favorable changes to the gut microbiome.

The DASH and Mediterranean diets both include a wide variety of whole grains, fruits, and vegetables, which provide fiber to gut microbes and several plant compounds that help to regulate the body's immune system and inflammation. They also limit refined carbohydrates and red or processed meats, which have been linked to an increased risk of colorectal cancer. Instead, they recommend eating fish more often and including low-fat dairy, both of which are associated with a reduced colorectal cancer risk. They recommend fat sources like nuts, seeds, and olive oil, which provide polyunsaturated fats, which can also protect against this cancer.

While some research suggests that vegan or vegetarian diets are the most protective against colorectal cancer, other findings have shown that pescetarian diets are equally protective. These diets tend to have more complex carbohydrates, fiber, and plant-based protein compared to omnivorous diets, which could be why vegan diets may offer more protection against certain heart and metabolic diseases. But omnivorous diets that include small amounts of red meat can also be protective.

Recent studies have also looked into ketogenic diets, which are high in fat and low in carbohydrates. According to this research, a ketogenic diet may lead to a decrease in certain inflammatory immune cells. However, it can also reduce the numbers of certain beneficial gut bacteria. When a ketogenic diet is necessary for medical reasons, it can be modified to minimize the negative effects on the gut bacteria. For example, some high-protein foods like whey protein and pea protein promote the growth of beneficial bacteria, and other foods high in both fat and protein—like salmon—provide omega-3 fatty acids.

Studies have also examined Paleo dietary patterns, which restrict foods to those thought to have been eaten by our Paleolithic ancestors. Long-term, strict adherence to a Paleo-style diet has been associated with a reduction in beneficial gut bacteria, which may result from the low intake of fiber and resistant starch. Paleo dieting has also been associated with elevated levels of Trimethylamine-N-oxide, a compound produced by gut microbes that's been tentatively associated with cardiovascular problems. Its production may be influenced by the duration of the diet.

Fermented Foods

Fermented foods have been consumed by humans for a long time and were initially developed to make food last longer. However, the fermentation process and the live bacteria present in these foods offer other potential benefits. During fermentation, lactic acid bacteria (LAB) like *Lactobacillus* are allowed to flourish as they convert carbohydrates to lactic acid, and some strains of LAB are considered probiotic.

Currently, fermented dairy products like yogurt and kefir are the only foods considered probiotic. The World Health Organization (WHO) defines probiotics as "live microorganisms which when administered in adequate amounts confer a health benefit on the host." The term *adequate amounts* often refers to the presence of a significant number of live, active culture colony-forming units (CFUs). A common benchmark is a minimum of 1 billion CFUs for it to be considered a "true" probiotic. These fermented dairy products, as well as fermented milk from cows, goats, and camels, have higher levels of nutrients like folate, vitamin K, and riboflavin. They also contain compounds with antioxidant and antihypertensive properties.

So far, RCTs and epidemiological data suggest that consuming fermented dairy products is associated with lower LDL cholesterol levels and a reduced risk of cardiovascular disease and colorectal cancer. There is weaker evidence suggesting that fermented dairy might improve bone mineral density, and a few studies show potential benefits in reducing muscle soreness and potentially improving mood.

Grains and legumes can also be fermented to produce foods like sourdough bread, tempeh, and fermented lentils, quinoa, wheat, rye, and bran. These products present similar benefits to fermented dairy, including increased vitamin content, production of antioxidant and antihypertensive compounds, and a reduction in FODMAPs. Fermented fruits and vegetables, such as kimchi, have not been extensively studied. However, early findings suggest similar benefits, including higher levels of phenols, many of which may have antioxidant and anti-inflammatory effects. Emerging findings also indicate that kimchi, gochujang, and fermented soy may improve lipid profiles.

Pro-, Pre-, and Synbiotics

Probiotic supplements, which consist of live microorganisms, can have a positive impact on your gut and overall well-being. Although the exact mechanisms aren't clear, it appears that they might work by preventing the growth of harmful bacteria, improving the integrity of our intestinal cells, and influencing our immune and nervous systems. However, there are still questions about whether probiotics actually colonize the digestive tracts of those who take them and whether this matters.

Recent research has shown that even if probiotic bacteria are found in fecal samples, they may not have an impact on the tissues and the microbiota of the gut. The location where probiotics settle and their effects are not under our control. Due to the differences in people's microbiomes, diets, bacterial strains, and study designs, it is challenging to draw definitive conclusions about the effectiveness of over-the-counter probiotics. However, studies consistently show that probiotic supplementation can be beneficial in certain situations, like preventing traveler's diarrhea.

Probiotics are distinct from another common gut supplement: prebiotics. Prebiotics are nutrients that beneficial bacteria in our gut can use as food. They are usually in the form of fiber or carbohydrates that our bodies can't digest. These nutrients are fermented by microbes in our gut, producing energy and beneficial substances called "postbiotics," such as SCFAs. Studies in animals and humans have shown that prebiotics can increase the numbers of helpful bacteria like *Bifidobacteria,* reduce inflammation and toxins in the body, regulate the gut's permeability, and even affect our appetite.

You don't necessarily need to take prebiotic supplements because these carbohydrates are already present in many foods. Fruits, vegetables, whole grains, nuts, and legumes contain prebiotic fibers like inulin and beta-glucan, which promote the growth of beneficial bacteria. Resistant starch can be found in green bananas, cooked and cooled potatoes, and rice. Studies have suggested that consuming resistant starch may increase *Bifidobacteria*, *lactobacilli*, and the production of butyrate while reducing inflammation markers, although the results can vary depending on a person's existing microbes.

Synbiotics combine probiotics with prebiotics. For example, a synbiotic might combine a specific strain of *Lactobacillus* and fiber. Emerging evidence suggests that, just like probiotics, synbiotics may reduce inflammation markers in people with obesity or IBD and improve symptoms of IBS and IBD.

Considering the limitations and high cost of probiotic and prebiotic supplements, it is more advisable to meet the recommended daily intake of fiber by consuming a wide variety of whole grains, fruits, vegetables, nuts, and legumes (such as beans).

Chemicals and Additives

There's no shortage of alarming headlines and confident influencers proclaiming the dangers of "chemicals," but the vast majority of these claims are either completely fabricated or based on misinterpretations of studies done in cell culture or animal models. Artificial sweeteners, such as acesulfame K, aspartame, saccharin, and sucralose, have been approved by the FDA. The safety of these sweeteners, as well as of other sweeteners like stevia and sugar alcohols, has been extensively studied. However, their impact on the gut microbiome is a newer area of research. Studies on cells have shown that artificial sweeteners induce increased hormone production and inflammation. However, these findings have not been replicated in most rodent models or in human data. Overall, the evidence shows that artificial sweeteners are safe for both humans and their gut microbes.

Genetically modified organisms, so-called GMOs, have also gotten a bad reputation, but it might not be earned. Genetic modification (GM) involves adding specific genetic material to organisms to produce desired traits, such as resistance to insects. GM crops actually reduce the amount of pesticides needed to protect crops, resulting in higher yields and income for farmers and less pesticidal contamination in the end product. Studies have shown that the effects of GM proteins on the gut microbiota are similar to those of conventional versions, even at higher levels of ingestion. GM crops undergo rigorous safety and allergenicity testing before they are approved for consumption by the FDA.

Food additives are substances added to improve the quality and shelf life of foods. These additives must be authorized by government agencies and undergo rigorous testing before being approved for use. They have set limits on how much can be safely consumed, known as the acceptable daily intake. Some additives are classified as generally recognized as safe (GRAS) based on their long history of use or their inert nature. Recent studies, conducted on rodents, have shown that some food additives and GRAS substances can lead to intestinal inflammation, changes in the microbiota, metabolic issues, liver abnormalities, and behavioral changes. However, these studies used rodents or cell cultures to artificially model the human microbiome, and the doses used were sometimes unrealistic for human consumption. So, it remains uncertain whether these effects can be replicated in humans.

Circadian Rhythms

Gut microbiota follow the same daily rhythm as the rest of your body, called the circadian rhythm. Disruptions in the gut microbiota, such as completely removing them in rodents, can disrupt the daily rhythm of the host animals. In humans, disruptions in circadian rhythm, like those caused by shift work, are associated with metabolic syndrome. These studies suggest that the interaction between the host and the gut microbiome is bidirectional.

Researchers are now looking into the impact of nutrient timing on circadian rhythms. One approach gaining popularity is time-restricted eating (TRE). This involves fasting periods and reduced meal frequency to align with circadian rhythms and affect metabolism and inflammation. Specifically, people who follow TRE diets will eat their meals in a restricted window, usually between 6 and 12 hours, and fast for the rest. Studies in rodents and a few human trials have shown that different forms of TRE can change inflammatory markers and the gut microbiota, potentially providing protection to the GI and nervous systems.

However, there is conflicting data suggesting that the changes may be due to caloric restriction rather than timed eating. Starvation studies have shown mixed results in animals and humans. Microbial diversity either varied unpredictably or remained unchanged in response to food restriction.

When changes did occur, they typically involved an increase in taxa that can withstand low energy availability, supporting the idea that dietary changes play a role in the effect of fasting on the microbiome. Current evidence suggests that eating two to six meals per day, preferably during daylight hours, including breakfast when hungry in the morning, is a good way to structure meals for overall health and a normal circadian rhythm.

READING

Biesalski, H. K. "Nutrition Meets the Microbiome: Micronutrients and the Microbiota. *Annals of the New York Academy of Sciences* 1372, no. 1 (2016): 53–64. https://doi.org/10.1111/nyas.13145.

Cignarella, F., C. Cantoni, L. Ghezzi, A. Salter, Y. Dorsett, L. Chen, L. Fontana, et al. "Intermittent Fasting Confers Protection in CNS Autoimmunity by Altering the Gut Microbiota." *Cell Metabolism* 27, no. 6 (2018): 1222–1235. https://doi.org/10.1016/j.cmet.2018.05.006.

Frazier, K., and E. B. Chang. "Intersection of the Gut Microbiome and Circadian Rhythms in Metabolism." *Trends in Endocrinology & Metabolism* 31, no. 1 (2020): 25–36. https://doi.org/10.1016/j.tem.2019.08.013.

Kaczmarek, J. L., S. V. Thompson, and H. D. Holscher. "Complex Interactions of Circadian Rhythms, Eating Behaviors, and the Gastrointestinal Microbiota and Their Potential Impact on Health." *Nutrition Reviews* 75, no. 9 (2017): 673–682. https://doi.org/10.1093/nutrit/nux036.

Karl, J. P., A. M. Hatch, S. M. Arcidiacono, S. C. Pearce, I. G. Pantoja-Feliciano, L. A. Doherty, and J. W. Soares. "Effects of Psychological, Environmental and Physical Stressors on the Gut Microbiota." *Frontiers in Microbiology* 9 (September 2018): 1–32. https://doi.org/10.3389/fmicb.2018.02013.

Lang, J. M., C. Pan, R. M. Cantor, W. H. W. Tang, J. C. Garcia-Garcia, I. Kurtz, S. L. Hazen, et al. "Impact of Individual Traits, Saturated Fat, and Protein Source on the Gut Microbiome." *mBio* 9, no. 6 (2018): e01604-18. https://doi.org/10.1128/mBio.01604-18.

Laudisi, F., C. Stolfi, and G. Monteleone. "Impact of Food Additives on Gut Homeostasis." *Nutrients* 11, no. 10 (2019): 2334. https://doi.org/10.3390/nu11102334.

Le Roy, C., P. Wells, J. Si, J. Raes, J. Bell, and T. Spector. "Red Wine Consumption Associated with Increased Gut Microbiota α-Diversity in 3 Independent Cohorts." *Gastroenterology* 158 (2019). https://doi.org/10.1053/j.gastro.2019.08.024.

Spencer, M., A. Gupta, L. Van Dam, C. Shannon, S. Menees, and W. D. Chey. "Artificial Sweeteners: A Systematic Review and Primer for Gastroenterologists." *Journal of Neurogastroenterology and Motility* 22, no. 2 (2016): 168–180. https://doi.org/10.5056/jnm15206.

Suez, J., T. Korem, G. Zilberman-Schapira, E. Segal, and E. Elinav. "Non-caloric Artificial Sweeteners and the Microbiome: Findings and Challenges. *Gut Microbes* 6, no. 2 (2015): 149–155. https://doi.org/10.1080/19490976.2015.1017700.

12

A Lifestyle for Gut Health

When it comes to supporting gut health, your dietary choices are an obvious place to start, but you don't want to forget three other important factors: exercise, sleep, and stress management. In today's fast-paced society, it's easy to put these off because you're overwhelmed and feel like you don't have time. But that's exactly when you need these habits the most! A prudent diet is foundational to supporting your gut health, but the rest of your lifestyle matters, too. As you'll learn in this lecture, exercise, sleep, and stress can have a significant impact on your gut microbiome and your overall health.

Exercise and GI Issues

A growing body of research has been exploring how the microbiome could play a role in the immune and mental-health-related benefits of regular exercise. First, exercise has been shown to enhance immune function by increasing the production and circulation of various immune cells, and recent research has highlighted the role of the gut microbiome in this process. Remember, some microbes produce metabolites like SCFAs that help regulate the activity of immune cells, reduce inflammation, and enhance immune surveillance. Exercise seems to encourage these SCFA-producing microbes to flourish. So, in tandem with your gut microbiome, exercise could ultimately promote a more robust immune system.

Research has also recently shed light on the role of the gut-brain axis in mediating the mental health benefits of exercise. Exercise has been shown to modulate the gut microbiome composition, increasing the abundance of bacteria that produce various neurotransmitters. This could explain why exercise seems to improve some of the symptoms of mood disorders like depression and anxiety as well as IBS, which is tentatively considered to be a disorder of the gut-brain axis.

The WHO suggests that adults between the ages of 18 and 64 perform 150 to 300 minutes of moderate-intensity aerobic physical activity each week. This could be as simple as taking a brisk 30-minute walk each day. Alternatively, the WHO recommends 75 to 150 minutes of vigorous-intensity aerobic activity, like jogging, per week. If you're a regular exerciser, you may already be meeting or exceeding these recommendations, but you might have another concern: the GI distress you experience after a hard training session. A significant percentage of athletes often experience GI problems during intense physical activity, ranging from increased gas and stomach discomfort to nausea and even bloody diarrhea.

Exercise-associated GI distress tends to affect endurance athletes and females more frequently. Intense exercise seems to have a particularly strong influence on the GI tract. For example, some studies have associated GI symptoms with exercising close to VO_2 max, which is the maximum volume (V), or amount, of oxygen (O_2) that the body can utilize during exercise. The more intense the activity, the closer you tend to get to VO_2 max. One study found that even a

relatively short run exceeding 60% of VO_2 max can raise markers of intestinal permeability, while intense exercise above 85% VO_2 max can delay gastric emptying, impeding the movement of food from the stomach.

Several theories attempt to explain the causes behind these issues. For instance, elevated body temperature has been found to modify the gut microbiome. Exercise can also temporarily deprive gut microbes of oxygen while blood is sent to the muscles rather than the GI tract. Without oxygen, microbes turn to anaerobic bacterial metabolism. That can subtly change pH levels throughout the gut and potentially alter the composition of the microbiome on a nonpermanent basis.

In endurance athletes, some GI issues may also be caused by a spike in free radicals. Free radicals are a natural result of exercise, produced when we use oxygen to convert adenosine triphosphate to energy. But these highly reactive molecules can damage DNA, lipids, and proteins. An overabundance of free radicals, especially when the body struggles to get rid of them, can cause a state called oxidative stress, which can damage gut tissues. Endurance athletes also exhibit varying levels of endotoxins after exercise, which might be due to temporarily elevated intestinal permeability. Although it isn't always the case, those that require medical attention tend to show higher markers of inflammation and circulating endotoxins.

Limited research has explored the effects of resistance training on GI function, but one study found impaired protein digestion and absorption after an intense exercise session. Implementing dietary modifications, such as following a short-term low-FODMAP diet, eating less fat before exercise, and drinking enough carbohydrates around workouts, has shown promise in alleviating symptoms associated with exercise-induced GI distress. It's important to note that all these symptoms are temporary, short-term effects. The benefits of exercise for the gut and overall health certainly outweigh its risks.

Gut Health and Sleep

Recent research suggests that poor sleep can have a serious impact on gut health. Disrupted sleep patterns and insufficient sleep are linked to a variety of digestive problems and worsening symptoms, including IBD, IBS, and gastroesophageal reflux disease. Sleep deprivation and night shift work have also been linked to changes in the gut microbiome, perhaps in part because they disrupt the circadian rhythms of both the host and the microbes. Compared to people who sleep a full 8 hours, those who get 6 or fewer tend to have less-diverse microbiomes and higher levels of both intestinal permeability and circulating markers of inflammation.

Establishing a regular nightly routine before bed can help you wind down and fall asleep more quickly. Both caffeinated drinks and alcohol can prevent you from falling asleep or reduce your sleep quality by disrupting your normal

sleep cycles. Caffeine can affect your brain for several hours after you've ingested it, so you may need to switch to decaf 4 to 6 hours before you plan to go to sleep.

It's also a good idea to avoid screens before bed and to start your routine early enough that you have time to wind down before you actually lie down. Consider setting a timer for about 30 minutes before you want to fall asleep, and at that point, set your phone and other screens in another room or away from your bed. Consider reading, journaling, or performing deep breathing exercises along with a calming scent like lavender to relax even further. Lastly, keep your bedroom cool and dark, as heat and humidity can disrupt sleep.

Stress

The stress response is hardwired into our bodies to help us cope with challenging situations. However, when stress becomes chronic, it can impact our health. For one thing, it can seriously disrupt digestion. It can prevent the stomach from emptying or cause digestive material to pass rapidly through the system. Chronic stress is associated with an increased risk of developing IBS, and even short-term, or acute, stress is linked to worsening IBS symptoms, such as increased inflammation. Anecdotally, many people report GI distress during times of stress and anxiety, and there's a strong link between mood disorders like anxiety and IBS.

So, just like sleep and exercise, managing stress plays an important role in gut health. Yoga and mindfulness-based practices to reduce stress have been studied in patients with IBD and IBS, and both have the potential to reduce symptoms of anxiety and depression, improve mood, and enhance quality of life.

Sometimes, patients find that they have unhelpful thoughts and behaviors associated with their IBS symptoms. Cognitive behavioral therapy (CBT) can help patients address these habits and develop new ways of thinking about their stressors. When combining CBT with other practices, such as mindfulness, people with IBS can better understand and accept their abdominal sensations, which can then help them manage their IBS more effectively. Another new potential method for reducing stress to deal with GI

disorders is gut-directed hypnotherapy. This involves inducing a suggestible mental state, or hypnosis, with the help of verbal guidance from the therapist. The aim is to facilitate receptivity to therapeutic suggestions.

There's growing concern around how pollution and environmental toxins may impose another form of physical stress on our bodies. Certain chemicals, such as persistent organic pollutants (POPs), can accumulate in body tissues and potentially cause illness. While levels of POPs in the environment have decreased due to regulations, their effects on human health at the levels detected are still uncertain. These environmental chemicals could potentially affect the microbiome by interfering with enzyme activity, altering the growth of certain bacteria, or by being metabolized by the microbiota. Some studies have looked into these effects. However, they've mostly been conducted in unrealistic doses using rodents, fish, and cell cultures. The results vary depending on the model and chemical used and are not applicable to real-life human exposure.

The Future of Gut Research

There's still much more to discover about the relationship between your gut microbes and your health. For all that researchers don't know, there's quite a lot that they're learning. Consider the rapid advances in gene sequencing over just the past 10 years that have made it possible to identify more microbes and possibly even get an idea of what they're doing.

People are much more curious and concerned (and confused) about their gut health now. The near future is uncertain because the field is so new and turbulent. To put it into perspective, insulin was discovered 100 years ago, in 1921, and it revolutionized the treatment of diabetes. However, early insulin-based treatments were difficult to produce and came with many health risks. It took a little more than 50 years before synthetic insulin was developed, and the first artificial pancreas was only approved by the FDA within the last 10 years. Who knows how long it might take to come up with microbiome-centered therapies? That said, technology tends to improve exponentially, so researchers might find answers and solutions faster with each new generation.

READING

Benedict, C., H. Vogel, W. Jonas, A. Woting, M. Blaut, A. Schürmann, and J. Cedernaes. "Gut Microbiota and Glucometabolic Alterations in Response to Recurrent Partial Sleep Deprivation in Normal-Weight Young Individuals." *Molecular Metabolism* 5, no. 12 (2016): 1175–1186. https://doi.org/10.1016/j.molmet.2016.10.003.

Manor, O., C. L. Dai, S. A. Kornilov, B. Smith, N. D. Price, J. C. Lovejoy, S. M. Gibbons, et al. "Health and Disease Markers Correlate with Gut Microbiome Composition across Thousands of People." *Nature Communications* 11 (2020): 5206. https://doi.org/10.1038/s41467-020-18871-1.

Mawdsley, J. E., and D. S. Rampton. "Psychological Stress in IBD: New Insights into Pathogenic and Therapeutic Implications." *Gut* 54, no. 10 (2005): 1481–1491. https://doi.org/10.1136/gut.2005.064261.

Mikocka-Walus, A. A., V. Pittet, J. B. Rossel, R, von Känel, and the Swiss IBD Cohort Study Group. "Symptoms of Depression and Anxiety Are Independently Associated with Clinical Recurrence of Inflammatory Bowel Disease." *Clinical Gastroenterology and Hepatology* 14, no. 6 (2016): 829–835. https://doi.org/10.1016/j.cgh.2015.12.045.

Mortaş, H., S. Bilici, and T. Karakan. "The Circadian Disruption of Night Work Alters Gut Microbiota Consistent with Elevated Risk for Future Metabolic and Gastrointestinal Pathology." *Chronobiology International* 37, no. 7 (2020): 1067–1081. https://doi.org/10.1080/07420528.2020.1778717.

Nunan, D., T. Cai, A. D. Gardener, J. M. Ordóñez-Mena, N. W. Roberts, E. T. Thomas, and K. R. Mahtani. "Physical Activity for Treatment of Irritable Bowel Syndrome." *Cochrane Database of Systematic Reviews* 6, no. 6 (2022). https://doi.org/10.1002/14651858.CD011497.pub2.

Palsson, O. S., and S. Ballou. "Hypnosis and Cognitive Behavioral Therapies for the Management of Gastrointestinal Disorders." *Current Gastroenterology Reports*, 22, no. 7 (2020): 31. https://doi.org/10.1007/s11894-020-00769-z.

Smith, R. P., C. Easson, S. M. Lyle, R. Kapoor, C. P. Donnelly, E. J. Davidson, E. Parikh, et al. "Gut Microbiome Diversity Is Associated with Sleep Physiology in Humans." *PloS One* 14, no. 10 (2019): e0222394. https://doi.org/10.1371/journal.pone.0222394.

Sugaya, N., K. Shirotsuki, and M. Nakao. "Cognitive Behavioral Treatment for Irritable Bowel Syndrome: A Recent Literature Review." *BioPsychoSocial Medicine* 15, no. 1 (2021): 23. https://doi.org/10.1186/s13030-021-00226-x.