

## MICROBIOLOGY

## Girth and the Gut (Bacteria)

Mouse, human studies begin to clarify gut bacteria's role in obesity

Five years ago, a team headed by Jeffrey Gordon of Washington University in St. Louis (WUSTL) in Missouri made a surprising discovery: The guts of obese mice and people harbor an array of microbes different from that of their lean counterparts. More provocatively, when they gave lean mice certain gut-dwelling microbes, the rodents became fat (*Science*, 29 May 2009, p. 1136). The findings sparked headlines and fueled popular speculation that manipulating gut bacteria might keep weight down in people.

Already, Martin Blaser had been heading down a similar track. Blaser, a microbiologist at New York University in New York City, was struck by how successful farmers are at increasing the growth rates of livestock by adding low doses of antibiotics to their feed. “The earlier in life they start the antibiotic, the more profound the effect,” he points out. He began to wonder whether antibiotic use, particularly in children, might affect the long-term establishment of a balanced microbial community in the human gut, eliminating bacteria there that could help ward off obesity. He started conducting mouse studies to examine the hypothesis.

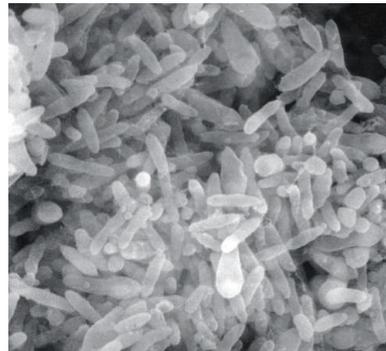
Since then, several other groups have joined in. A raft of intriguing obesity-related findings was presented at a meeting last month on the microbiome, the bacteria that live inside the guts and other tissues of animals. Yet many in the field caution that it remains difficult to determine whether changes in gut microbes drive or contribute to obesity or whether the excess weight itself triggers those changes. “The jury is still out [about] what the role of the gut microbiota may be in obesity in humans,” says Claire Fraser-Liggett, a microbiologist at the Uni-

versity of Maryland School of Medicine in Baltimore who has studied gut bacteria and obesity in the Amish.

### Case of the missing microbes

The farm animal–antibiotic connection was one clue that led Blaser to wonder about microbial causes of the obesity epidemic. Another was the fact that very few people now harbor the ulcer-causing bacterium *Helicobacter pylori* in their stomachs. *H. pylori*, which has also been linked to stomach cancers, is one of up to 1000 different microbes that call the human body home. Once ubiquitous in the human microbiome and still so in the guts of people from developing countries, it is now found in just 6% of U.S. children. That might seem like good news, as there should be fewer ulcers and cancers. But Blaser suspects that it is also bad news, as studies suggest that *H. pylori*'s presence in the gut helps regulate the stomach's production of the hormone ghrelin, which stimulates food intake.

This bacterium may not be the only species disappearing from our microbiome. After a person takes antibiotics, “it has always been presumed that the microbiota will spring back,” Blaser says. But the fate of *H. pylori* suggests otherwise. Its vanishing act and other shifts in the microbiome may contribute to an increased risk for weight gain, Blaser worries.



**Go with the gut.** Antibiotic use may adversely affect the long-term makeup of the intestine's bacterial communities.

He has started to investigate this theory by giving mice either low doses of antibiotics over long periods, akin to what farm animals receive, or short-term, high doses, more like what a sick infant or adult would get. He then compares the physiology and microbiomes of these treated rodents with those of mice raised under similar conditions but given no antibiotics. In one set of studies, the mice fed low doses of antibiotics long-term wound up with 15% more body fat than the control mice, Blaser reported last month at the International Human Microbiome Congress in Vancouver, Canada. The chubbier, antibiotic-fed mice also had about 25% more fat in their livers.

The treated mice also had a different set of bacterial species inhabiting their guts. And several hundred bacterial genes, including ones for fatty acid production, exhibited different levels of activity—some increasing, others decreasing—in these mice compared with the controls. Similar changes occur in the rodents given short pulses of antibiotics, he noted.

Antibiotics “may be driving the gut microbiome to a place where it shouldn't be,” Fraser-Liggett says. “We do not know the functional consequences, but with these miracle drugs now 60 years later, we may be seeing effects that change susceptibility to various diseases.”

Blaser will examine the gut microbiomes of children to see whether his results are applicable to humans. If so, “that would be a remarkable connection that could have a significant impact on medical care,” says genome scientist George Weinstock of WUSTL. But he's cautious: “In a lot of cases, the microbiome in mice doesn't translate into humans.”

### Patterns in genes

S. Dusko Ehrlich has avoided that issue, bypassing mice and instead directly examining whether patterns in the microbiome of people relate to body mass index and obesity. A microbiologist at the INRA Microbiology and Food Chain Division in Jouy-en-Josas, France, Ehrlich is part of a group, the Meta-

HIT consortium, investigating connections between microbial genes in human intestines and human health. By comparing such genes from obese and nonobese individuals, he and his colleagues have found that certain sets of bacterial genes and bacteria correlate with excess weight and insulin resistance.

The researchers first sequenced all the bacterial genes in stool samples of 177 Danes, 55 who were thin and 122 who were either overweight or obese. Although the researchers concluded that most participants in the study had roughly 600,000 distinct bacterial genes in their guts, almost one-third of the obese study participants had only about 360,000 such genes, 30% to 40% fewer. A similar percentage of 36 obese French people had a comparable dearth of gut bacteria genes, Ehrlich reported at the Vancouver meeting. Moreover, the obese people “don’t have as great a bacterial diversity” in their guts, Ehrlich reported. One missing microbe in that group was a methane producer, leading Ehrlich to wonder whether “the carbon that does not get out [of the body] as gas could be incorporated as fat.”

When they looked at medical histories of all their study subjects, Ehrlich and his colleagues found that the obese people with fewer gut bacteria genes were more likely to be insulin resistant than were the obese people who had a typical tally of intestinal microbial genes. These obese people also tended to have higher than normal white blood cell counts, suggesting that they were in a state of low-level inflammation, Ehrlich said. Some researchers have found evidence of a link between inflammation and obesity (*Science*, 17 December 2010, p. 1621).

Ehrlich and his colleagues have also tested whether the types of bacteria in a person’s gut can “diagnose” obesity. Using just six meta-species, they were able to correctly predict whether a person was lean or obese more than 80% of the time, he reported. When researchers try to make the same predictions by considering all of a person’s genetic risk factors for obesity, they are right only 58% of the time, Ehrlich pointed out.

At this point, however, it’s unclear whether the differences in intestinal microbes are “the cause, a contribution to, or the consequence”

of obesity, notes Ehrlich. “If we can provide evidence that they [at least] provide a contribution, then we can go and find a treatment.”

### Help from the Amish

Other work presented at the microbiome meeting indicates that sorting out this cause-and-effect puzzle will be tough. Frustrated by the inconsistent results others were getting when they looked for connections between the microbiome and obesity, Fraser-Liggett and her colleagues examined 400 adult Amish living in Pennsylvania. Amish marry within their group and have very similar lifestyles, environment, and eating habits—they even cook in communal kitchens. Thus, Fraser-Liggett hoped to eliminate some of the variables that might have confounded other studies.

The body mass index of Amish ranged from 16 to 51 (30 is obese), and some of the

rial genes are active in a person and not just at which bacteria are there. Scientists have found that although the species mix of the microbiome may vary significantly from one person to the next, those individuals often still have equivalent complements of bacterial genes at work inside them. Through such gene analyses, researchers can begin to better assess what the bacteria in the gut are really doing to, or for, their host, Fraser-Liggett notes.

Gordon suggests that more clarity on the obesity-microbiome issue will also come from using the guts of mice as bioreactors for human microbes. His group has pioneered the study of germ-free mice, which are grown in a sterile environment from birth, and he is now exposing such mice to bacteria from human guts. Once those human microbes have established residence in the guts of the mice, Gordon then feeds

the animals a variety of human diets. Using these rodent proxies, he can thus track how different diets affect the “human” microbiomes, assessing the bacteria as often as he needs to to get a dynamic picture. “You can sample many features between the microbial community and the host,” he says.

Despite his role in igniting the study of obesity and microbiomes, Gordon resists the idea that our gut bacteria are the sole explanation for the growing number of obese people. “It’s not the dominant part of the problem; excessive energy intake is,” he contends.

The simplistic notion of only changing one’s gut bacteria to lose weight has been “hyped a lot,” he complains.

Others, such as microbial ecologist Liping Zhao of Shanghai Jiao Tong University in China, are more convinced that the microbiome will prove important when it comes to obesity. “Increased abundance of ‘bad genes’ and [a] decrease of ‘good genes’ in our diet-disrupted gut microbiome [may] be the primary driving force for this obesity epidemic,” Zhao says.

Even if Zhao’s prediction proves right, the studies to date make clear that the connection between the microbiome and excess weight is complex, Fraser-Liggett says. For those looking to bacteria to stem the obesity epidemic, she concludes, “there’s clearly no magic formula.”

—ELIZABETH PENNISI



**Personalized microbiota.** Germ-free mice grown in sterile environments and given human gut microbes provide a way to test links between bacteria and obesity.

obese ones also had metabolic syndrome. Fraser-Liggett and her colleagues captured a snapshot of the gut microbiome of each Amish by obtaining stool samples, sequencing the DNA in them, and using the 16S ribosomal subunit gene often used to tell bacteria apart, identifying any microbial components. Although the scientists did detect some differences in certain bacteria between obese and lean Amish, they didn’t find the dramatic shifts that Gordon had documented between lean and obese mice, Fraser-Liggett reported at the meeting.

One problem may be that simply taking a census of the bacteria present in a person’s gut may not be enough. “16S [analysis] is not very informative,” Ehrlich says. “We need to go to more precise measures.” Increasingly, microbiome researchers are looking at what bacte-