WN *Hot topic*

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Microbiota
Bugged





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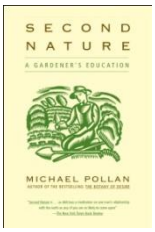
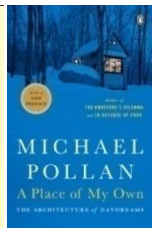
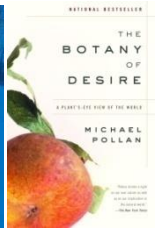
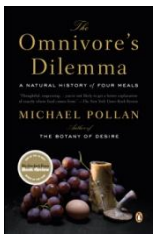
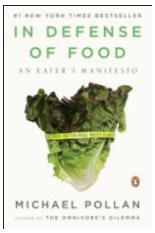
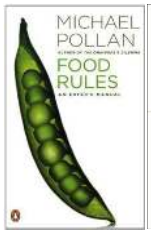


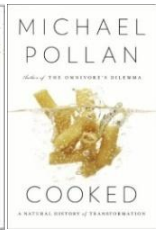




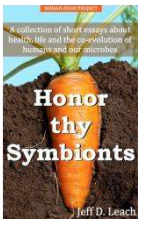








Michael Pollan's books written over more than 20 years: *Second Nature* (1991), *A Place of my Own* (1997), *The Botany of Desire* (2001); *The Omnivore's Dilemma* (2006), *In Defense of Food* (2008), *Food Rules* (2010), *Cooked: a Natural History of Transformation* (April 2013)



Jeff Leach's Naked Pizza advertisement; *New York Times* with op-ed piece on the biome; and his books: *Bloom. An Eater's Guide to a Healthy Biome* (2012), *Honor Thy Symbionts* (2013)

Summary

Hot topic is the title of a new *WN* occasional series. As the name implies, it will address areas and issues of importance to public health and nutrition that are controversial or high profile, and that need to be discussed and better understood. Such topics may be regularly exposed in popular print and electronic media, but relatively ignored, overlooked or neglected in the scientific and academic media, or else addressed extensively on the internet or in publications sometimes supposed to be outside the scope of nutrition science. So it may be best to say that the general title of *Hot topic* is of commentaries of a speculative nature, even though the topics may well not be seen as such by their protagonists.

In this first of the *Hot topic* series Michael Pollan, and the anthropologist, explorer, entrepreneur and campaigner Jeff Leach, report on and outline the importance of gut microbial ecology, including what they see as the crucial and central important of the nutrition, nourishment and protection of the trillions of bacteria and scores of important species of bacteria that live on us and in us, notably inside our lower gut – our colon. Their functions are still not well understood.

Jeff Leach and Michael Pollan both infer what amounts to a revolution in conventional thinking about nutrition. It may well be that we should all think first about the nourishment of our gut biota. This means diets that preferentially protect our 'friendly flora', those bacterial species that are commensal, that live with us with no harm done, and those that are symbiotic, positively protective against the alien species that can cause infective and infectious diseases. Deranged gut microbial ecology may even be implicated in various serious chronic diseases of the gut such as Crohn's disease, diverticular disease and ulcerative colitis.

If what follows was in the form of a conventional paper, Jeff Leach would be the first author, because the original research is his, and that of colleagues and co-workers whose findings are mostly in the anthropological literature. Michael Pollan is the reporter, including on himself, and has like all outstanding investigative journalists, become somewhat of an authority on the topic along the way. One of Jeff Leach's statements is below.

As humans have evolved, so, too, have our diseases. Autoimmune disease affects an estimated 50 million people at an annual cost of more than $100 billion. And the suffering, and monetary costs, are sure to grow. Maybe it's time we talk more about human ecology when we speak of the broader environmental and ecological concerns of the day. The destruction of our inner ecosystem surely deserves more attention, as global populations run gut-first into the buzz saw of globalisation and its microbial scrubbing diet. But more important, we should seriously consider making evolutionary biology a basic science for medicine, or making its core principles compulsory in secondary education. Currently they are not.

Geoffrey Cannon writes:

WN published 15 appraisals of Michael Pollan's life and work in our April and May issues, ending with his own reviews of his favourite current books

As a *World Nutrition* reader, you don't need more about Michael Pollan here. He was on the cover of our April issue, with appraisals of his life and work by a serious of writers, together with an extract from his new book *Cooked*. We received so many appraisals, almost all illuminating, that we ran another series last month in May. Readers may wonder if *WN* has got shares in Michael Pollan Inc? No, it's not that.

This is the story. For some time now we have been planning commentaries with the general title *Hot topic*. The purpose is to bring to light areas of relevance and interest that are overlooked, or neglected, often because orthodox professionals feel that these are 'way out'. This applies to most areas that are new, unless like -omics they are rewarded with massive grants because of their commercial potential.

Our intention was to begin the series with a commentary on gut microbial ecology. This is an voyage to a new found land or, to mix images, an exploration of our own dark continents – our guts. We had decided to feature Jeff Leach, an archeologist who is somewhat of a nutritional Indiana Jones. He has credentials (see Box 3 below). He believes, in common with eminent researchers such as the Karolinska Institute's Tore Midtvedt, that the impact of gut bacteria, which among many functions form the first line of our immune defences, is grossly underestimated. He believes that nutrition needs to include nourishment and protection of our gut flora, as a first priority. Big stuff. He has put his money where his mouth is, by co-founding the Naked Pizza chain based in New Orleans, now also as NKD Pizza in Dubai, whose products are made with a whole new formula, and also co-founding *The Human Food Project*, designed to track and check world gut health.

Then we discovered that Michael Pollan and Jeff Leach had met, and that Michael, after completing his book *Cooked*, had become a subject for the Human Food Project. We had no choice! Below is the *New York Times* feature by Michael published on 15 May. And Jeff Leach? He was somewhere in the interior of Africa, in search of the perfect human biome. He still is, as far as we know.

Our second genome


Counted as cells, about 1 of us is human and about 10 of us is bacterial, all over us but mostly in our guts. At right, one commensal bacterial species

Michael Pollan writes:

I can tell you the exact date that I began to think of myself in the first-person plural – as a superorganism, that is, rather than a plain old human being. It happened on 7 March. That's when I opened my e-mail to find a huge, processor-choking file of charts and raw data from a laboratory located at the Bio-Frontiers Institute at the University of Colorado, Boulder.

As part of a new citizen-science initiative called the *American Gut Project*, the lab sequenced my microbiome – that is, the genes not of 'me', exactly, but of the several hundred microbial species with whom I share this body of mine. These bacteria, which number around 100 trillion, are living (and dying) right now on the surface of my skin, on my tongue and deep in the coils of my intestines, where the largest contingent of them will be found, a pound or two of microbes together forming a vast, largely uncharted interior wilderness that scientists are just beginning to map.

I clicked open a file called Taxa Tables, and a colorful bar chart popped up on my screen. Each bar represented a sample taken (with a swab) from my skin, mouth and feces. For purposes of comparison, these were juxtaposed with bars representing the microbiomes of about 100 'average' Americans previously sequenced.

Here were the names of the hundreds of bacterial species that call me home. In sheer numbers, these microbes and their genes dwarf us. It turns out that we are only 10 percent human: for every human cell that is intrinsic to our body, there are about 10 resident microbes – including commensals (generally harmless freeloaders) and mutualists (favor traders) and, in only a tiny number of cases, pathogens. To the extent that we are bearers of genetic information, more than 99 percent of it is microbial. And it appears increasingly likely that this 'second genome,' as it is

sometimes called, exerts an influence on our health as great and possibly even greater than the genes we inherit from our parents. But while your inherited genes are more or less fixed, it may be possible to reshape, even cultivate, your second genome.

Our old friends

Justin Sonnenburg, a microbiologist at Stanford University, suggests that we would do well to begin regarding the human body as 'an elaborate vessel optimized for the growth and spread of our microbial inhabitants.' This humbling new way of thinking about the self has large implications for human and microbial health, which turn out to be inextricably linked. Disorders in our internal ecosystem – loss of diversity, say, or a proliferation of the 'wrong' kind of microbes — may predispose us to obesity and a whole range of chronic diseases, as well as some infections. 'Fecal transplants', which install a healthy person's microbiota into a sick person's gut, have been shown to effectively treat an antibiotic-resistant intestinal pathogen named *Clostridium difficile*, which kills 14,000 people in the USA each year. (Researchers use 'microbiota' to refer to all the microbes in a community, and 'microbiome' to refer to their collective genes.)

We've known for a few years that obese mice transplanted with the intestinal community of lean mice lose weight and vice versa. (We don't know why.) A similar experiment was performed recently on humans by researchers in the Netherlands: when the contents of a lean donor's microbiota were transferred to the guts of male patients with metabolic syndrome, the researchers found striking improvements in the recipients' sensitivity to insulin, an important marker for metabolic health. Somehow, the gut microbes were influencing the patients' metabolisms.

Our resident microbes also appear to play a critical role in training and modulating our immune system, helping it to accurately distinguish between friend and foe and not go nuts on, well, nuts and all sorts of other potential allergens. Some researchers believe that the alarming increase in autoimmune diseases in the West may owe to a disruption in the ancient relationship between our bodies and their 'old friends' – the the microbial symbionts with whom we coevolved.

These claims sound extravagant, and many microbiome researchers are careful not to make the mistake that scientists working on the human genome did a decade or so ago, when they promised they were on the trail of cures to many diseases. We're still waiting. Yet whether any cures emerge from the exploration of the second genome, the implications of what has already been learned – for our sense of self, for our definition of health and for our attitude toward bacteria in general – are difficult to overstate. Human health should now 'be thought of as a collective property of the

human-associated microbiota,' as one group of researchers recently concluded – that is, as a function of the community, not the individual.

Such a paradigm shift comes not a moment too soon, because as a civilization, we've just spent the better part of a century doing our unwitting best to wreck the human-associated microbiota with a multi-fronted war on bacteria, and with a highly processed diet, both notably detrimental to its well-being. Researchers now speak of an impoverished 'Westernized microbiome' and ask whether the time has come to embark on a project of 'restoration ecology' – not in the rain forest or on the prairie but right here at home, in the human gut.

Colonisation begins at birth

In March I traveled to Boulder to see the Illumina HiSeq 2000 sequencing machine that had shed its powerful light on my own microbiome, and to meet the scientists and computer programmers who were making sense of my data. The lab is headed by Rob Knight, a rangy, crew-cut 36-year-old biologist who first came to the US from his native New Zealand to study invasive species, a serious problem in his home country. Knight earned his PhD in ecology and evolutionary biology from Princeton when he was 24 and then drifted from the study of visible species and communities to invisible ones. Along the way he discovered he had a knack for computational biology.

Knight is regarded as a brilliant analyst of sequencing data, skilled at finding patterns in the flood of information produced by the machines that 'batch sequence' all the DNA in a sample and then tease out the unique genetic signatures of each microbe. This talent explains why so many of the scientists exploring the microbiome today send their samples to be sequenced and analyzed by his lab; it is also why you will find Knight's name on most of the important papers in the field.

Over the course of two days in Boulder, I enjoyed several meals with Knight and his colleagues, postdoctoral and graduate students. I was a little taken aback by the table talk. I don't think I've ever heard so much discussion of human feces at dinner, but then one thing these scientists are up to is a radical revaluation of the contents of the human colon. I learned about Knight's 16-month-old daughter, who has had most of the diapers to which she has contributed sampled and sequenced. Knight said at dinner that he sampled himself every day, as does his wife.

A result of the family's self-study has been a series of papers examining family microbial dynamics. The data helped demonstrate that the microbial communities of couples sharing a house are similar, suggesting the importance of the environment in shaping an individual's microbiome. Knight also found that the presence of a family

dog tended to blend everyone's skin communities, probably via licking and petting. One paper, 'Moving pictures of the human microbiome,' tracked day-to-day shifts in the microbial composition of each body site. Knight produced animations showing how each community – gut, skin and mouth – hosted a fundamentally different cast of microbial characters that varied within a fairly narrow range over time.

Knight's daily sampling of his daughter's diapers (along with those of a colleague's child) also traced the remarkable process by which a baby's gut community, which *in utero* is sterile and more or less a blank slate, is colonized. This process begins shortly after birth, when a distinctive infant community of microbes assembles in the gut. Then, with the introduction of solid food and then weaning, the types of microbes gradually shift until, by age 3, the baby's gut comes to resemble an adult community much like that of its parents.

Breastmilk nourishes gut bugs

The study of babies and their specialized diet has yielded key insights into how the colonization of the gut unfolds and why it matters so much to our health. One of the earliest clues to the complexity of the microbiome came from an unexpected corner: the effort to solve a mystery about milk. For years, nutrition scientists were confounded by the presence in human breastmilk of certain complex carbohydrates, called oligosaccharides, which the human infant lacks the enzymes necessary to digest. Evolutionary theory argues that every component of mother's milk should have some value to the developing baby, or natural selection would have long ago discarded it as a waste of the mother's precious resources.

It turns out the oligosaccharides are there to nourish not the baby, but one particular gut bacterium within babies called *Bifidobacterium infantis*, which is uniquely well-suited to break down and make use of the specific oligosaccharides present in mother's milk. When all goes well, the bifidobacteria proliferate and dominate, helping to keep the infant healthy by crowding out less savory microbial characters before they can become established and, perhaps most important, by nurturing the integrity of the epithelium – the lining of the intestines, which plays a critical role in protecting us from infection and inflammation.

'Mother's milk, being the only mammalian food shaped by natural selection, is the Rosetta stone for all food,' says Bruce German, a food scientist at the University of California, Davis, who researches milk. 'And what it's telling us is that when natural selection creates a food, it is concerned not just with feeding the child but the child's gut bugs too.'

Where do these all-important bifidobacteria come from, and what does it mean if, like me, you were never breastfed? Mother's milk is not, as once was thought, sterile: it is both a 'prebiotic' – a food for microbes – and a 'probiotic,' a population of beneficial microbes introduced into the body. Some of them may find their way from the mother's colon to her milk ducts and from there into the baby's gut with its first feeding. Because designers of infant formula did not, at least until recently, take account of these findings, and did not include prebiotic oligosaccharides or probiotic bacteria in their formula, the guts of bottle-fed babies are not optimally colonized.

Most of the microbes that make up a baby's gut community are acquired during birth – a microbially rich and messy process that exposes the baby to a whole suite of maternal microbes. Babies born by Caesarean section however, a comparatively sterile procedure, do not acquire their mother's vaginal and intestinal microbes at birth. Their initial gut communities more closely resemble that of their mother's (and father's) skin, which is less than ideal, and may account for higher rates of allergy, asthma and autoimmune problems in C-section babies: not having been seeded with the optimal assortment of microbes at birth, their immune systems may fail to develop properly.

At dinner, Knight told me that he was sufficiently concerned about such an eventuality that, when his daughter was born by emergency C-section, he and his wife took matters into their own hands: using a sterile cotton swab, they inoculated the newborn infant's skin with the mother's vaginal secretions to insure a proper colonization. A formal trial of such a procedure is under way in Puerto Rico.

Western bugs, Western diseases?

While I was in Boulder, I sat down with Catherine Lozupone, a microbiologist who had just left Knight's lab to set up her own at the University of Colorado, Denver, and who spent some time looking at my microbiome and comparing it with others, including her own. Lozupone was the lead author on a 2012 paper in *Nature*, 'Diversity, stability and resilience of the human gut microbiota,' which sought to approach the gut community as an ecologist might, trying to determine the 'normal' state of the ecosystem and then examining the various factors that disturb it over time.

How does diet affect it? Antibiotics? Pathogens? What about cultural traditions? So far, the best way to begin answering such questions may be by comparing the gut communities of various far-flung populations, and researchers have been busy collecting samples around the world and shipping them to sequencing centers for analysis. The American Gut Project, which hopes to eventually sequence the

communities of tens of thousands of US citizens, represents the most ambitious such effort to date; it will help researchers uncover patterns of correlation between people's ways of life, diet, health status and the makeup of their microbial community.

It is still early days in this research. As Lozupone (and everyone else I interviewed) underscored; scientists can't even yet say with confidence exactly what a 'healthy' microbiome should look like. But some broad, intriguing patterns are emerging. More diversity is probably better than less, because a diverse ecosystem is generally more resilient – and diversity in the Western gut is significantly lower than in other, less-industrialized populations. The gut microbiota of people in the West looks very different from that of a variety of other geographically dispersed peoples. So, for example, the gut community of rural people in West Africa more closely resembles that of Amerindians in Venezuela than those of people from the USA or Europe.

These rural populations not only harbor a greater diversity of microbes, but also a different cast of lead characters. Guts in the US and Europe contain relatively high levels of bacteroides and firmicutes and low levels of the prevotella that dominate the guts of rural Africans and Amerindians. Why are the microbes different? It could be the diet, which in both rural populations features a considerable amount of whole grains (which prevotella appear to like), plant fiber and very little meat. (Many firmicutes like amino acids, so they proliferate when the diet contains lots of protein; bacteroides metabolize carbohydrates.)

The lower biodiversity in the West could be a result of our profligate use of antibiotics (in health care as well as the food system); our diet which includes so much processed food (which has generally been cleansed of all bacteria, the good and the bad); and environmental toxins and generally less 'microbial pressure' (exposure to bacteria) in everyday life. All of this may help explain why, though these rural populations tend to have greater exposures to infectious diseases and lower life expectancies than those in the West, they also have lower rates of chronic disorders like allergies, asthma, type 2 diabetes and cardiovascular disease.

'Rural people spend a lot more time outside and have much more contact with plants and with soil,' Lozupone says. Another researcher, who has gathered samples in Malawi, told me 'In some of these cultures, children are raised communally, passed from one set of hands to another, so they're routinely exposed to a greater diversity of microbes.' The nuclear family does not help the health of the microbiome.

As it happens, Lozupone and I had something in common, microbially speaking: we share unusually high levels of prevotella for Americans. Our gut communities look more like those of rural Africans or Amerindians than like those of our neighbors. Lozupone suspects that the reasons for this might have to do with a plant-based diet; we each eat lots of whole grains and vegetables and relatively little meat. (Though neither of us is a vegetarian.)

Box 1

Michael Pollan's guts

Edited from interview by Rachel Nolan in *The New York Times*, 20 May 2013.

RN What first got you interested in writing about the microbiome – the ecosystem of the several hundred microbial species that live inside a person's body?

MP In my research for my new book *Cooked*, I delved into various kinds of fermentation, and began hanging out with *fermentos* — passionate picklers, cheese-makers and brewers. These people all have relaxed attitudes toward bacteria, which they regard as friends. I describe them as 'pacifists in the war on bacteria' in which the rest of us have been foot soldiers. They introduced me to the importance of the fermentation going on inside us, and as I began looking into it, I realized that managing internal fermentation was an important aspect of health.

What surprised you most about the microbiome's effects on humans?

Our gut microbes can acquire genes from our food, changing them at the genetic level. A recent study found that the Japanese have a common gut bacterial species — one we share — but that in the Japanese, it has a gene that allows it to metabolise seaweed. This gene comes from a marine bacterium that lives on seaweed, so was acquired in the diet and has given the Japanese a digestive ability we lack. The other big surprise was that gut bacteria appear to influence our mood and temperament. They manufacture neurotransmitters like serotonin, for example. If you transplant the gut microbiota of relaxed and adventurous mice into the guts of timid and anxious mice they become less stressed and more adventurous.

Were scientists wary of exaggerating claims about the microbiome?

I encountered a great deal of reluctance to make recommendations about how best to manage our gut microbiome, and a deep caution about overpromising. But as soon as I asked scientists about how they had changed their own behaviour in light of what they were learning, they were happy to talk about their attitudes toward antibiotics, fermented foods and domestic hygiene.

Aside from eating fermented foods, and not going nuts with household cleaners, is there anything else that you recommend to improve your microbiome?

Garden. The exposure to soil is probably a good thing. Gardening is a good way to safely increase your 'microbial pressure' on a daily basis. Having a dog may be a good thing too.

The Western microbiome

Is there any possibility that the Western microbiome may hold some advantages? Did you hear any scientists speak out in favour of it?

No. No one spoke in praise of the Western microbiome — I heard many more expressions of concern, especially about its relative lack of diversity. But it's possible we're overlooking some important adaptation it has made to modern life, like microbes that detoxify various environmental toxins, for example. These may be keeping us healthy. The interactions of gut bugs and novel chemicals, including pharmaceuticals, is probably very important but not very well understood.

You write that bacterial makeup changes rapidly in response to antibiotics. How fast does a person's microbiome snap back to normal?

Not much work has been done on the resilience of the microbiome in the face of antibiotics, but recovery does appear to happen; the concern is whether after repeated courses of antibiotics, recovery is more fitful or less complete. Even after thoroughly washing your hands, your skin community swiftly recovers from that insult, and looks much like it did before the washing. The bugs are hiding in the grooves and crevices in your skin, just waiting for the storm to pass. That community is like another fingerprint — recognisably you. The user of a computer mouse or keyboard can be identified by the distinctive community of his or her skin microbes.

There's a possibility of connecting the microbiome to autoimmune disease. If that can be more firmly established, what sort of treatments might help?

It seems likely that there are key windows of development, in childhood and perhaps at puberty, where exposure to the 'right' microbes educates the immune system. Whether it's possible to reset those patterns of recognition and response later in life is not clear.

If probiotics become more and more important, is it possible they might be regulated more closely?

Most of the researchers were sceptical of the probiotics now being marketed. If we do develop next-generation probiotics, it's likely they will be treated more like — and regulated more like — pharmaceuticals than supplements.

Restricting antimicrobials

Does eating different types of food — 'dirtier' or more fermented — help our weak microbiomes, or will food not help us enough?

If indeed we have damaged or impoverished the Western microbiome, the problem needs to be addressed with changes in the diet, and with changes in our attitude and exposure to bacteria. We'll need to develop new norms for our everyday interactions with bacteria, and more rigorous protocols governing the use of antibiotics. The routine use of antibiotics in animal feed needs to be regulated and possibly banned. The argument for getting antibiotics out of agriculture is the rise of antibiotic-resistant microbes, which are a serious public health threat. The profligate use of antibiotics in agriculture is likely to be damaging the human microbiome.

Down and dirty

Michael Pollan has the disease-protective microbiome of an African. This good news is probably because he ferments food and digs dirt in his garden

Two other features of my microbiome attracted the attention of the researchers who examined it. First, the overall biodiversity of my gut community was significantly higher than that of the typical Westerner, which I decided to take as a compliment, though the extravagantly diverse community of microbes on my skin raised some eyebrows. 'Where have your hands been, man?', Jeff Leach of the American Gut Project [featured here with Michael – Ed] asked after looking over my results. My skin harbors bacteria associated with plants, soil and a somewhat alarming variety of animal guts. I put this down to gardening, composting (I keep worms too) and also the fact that I was fermenting *kimchi* and making raw-milk cheese, 'live-culture' foods teeming with microbes.

Compared to a rain forest or a prairie, the interior ecosystem is not well understood, but the core principles of ecology – which along with powerful new sequencing machines have opened this invisible frontier to science – are beginning to yield some preliminary answers and a great many more intriguing hypotheses. Your microbial community seems to stabilize by age 3, by which time most of the various niches in the gut ecosystem are occupied.

That doesn't mean it can't change after that; it can, but not as readily. A change of diet or a course of antibiotics, for example, may bring shifts in the relative population of the various resident species, helping some kinds of bacteria to thrive and others to languish. Can new species be introduced? Yes, but probably only when a niche is

opened after a significant disturbance, like an antibiotic storm. Just like any other mature ecosystem, the one in our gut tends to resist invasion by newcomers.

You acquire most of the initial microbes in your gut community from your parents, but others are picked up from the environment. 'The world is covered in a fine patina of feces', as the eminent Stanford microbiologist Stanley Falkow tells his students. The new sequencing tools have confirmed his hunch: Did you know that house dust can contain significant amounts of fecal particles? Or that, whenever a toilet is flushed, some of its contents are aerosolized? Knight's lab has sequenced the bacteria on toothbrushes. This news came during breakfast, so I didn't ask for details, but got them anyway: 'You want to keep your toothbrush a minimum of six feet away from a toilet,' one of Knight's colleagues told me.

What or who are 'we'?

Some scientists in the field borrow the term 'ecosystem services' from ecology to catalog all the things that the microbial community does for us as its host or habitat. The services rendered are remarkably varied and impressive. 'Invasion resistance' is one. Our resident microbes work to keep pathogens from gaining a toehold by occupying potential niches or otherwise rendering the environment inhospitable to foreigners. The robustness of an individual's gut community might explain why some people fall victim to food poisoning while others can blithely eat the same meal with no ill effects.

Our gut bacteria also play a role in manufacture of substances like neurotransmitters (including serotonin); enzymes and vitamins (notably the B vitamins, and vitamin K) and other essential nutrients (including important amino acid and short-chain fatty acids); and a suite of other signaling molecules that talk to, and influence, the immune and the metabolic systems.

Some of these compounds may play a role in regulating our stress levels and even temperament. For example, when gut microbes from easygoing, adventurous mice are transplanted into the guts of anxious, timid mice, they become more adventurous. The expression 'thinking with your gut' may contain a larger kernel of truth than we thought.

The gut microbes are looking after their own interests, chief among them getting enough to eat and regulating the passage of food through their environment. The bacteria themselves appear to help manage these functions by producing signaling chemicals that regulate our appetite, satiety and digestion. Much of what we're learning about the microbiome's role in human metabolism has come from studying

'gnotobiotic mice' raised in labs like Jeffrey Gordon's at Washington University, in St. Louis, to be microbially sterile, or germ-free.

Recently, Gordon's lab transplanted the gut microbes of Malawian children with kwashiorkor, an acute form of malnutrition, into germ-free mice. The lab found those mice with kwashiorkor who were fed the children's typical diet could not readily metabolize nutrients, indicating that it may take more than calories to remedy malnutrition. Repairing a patient's disordered metabolism may require reshaping the community of species in his or her gut.

Keeping the immune system productively engaged with microbes, exposed to lots of them in our bodies, our diet and our environment, is another important ecosystem service and one that might turn out to be critical to our health. 'We used to think the immune system had this fairly straightforward job,' Michael Fischbach, a biochemist at the University of California, San Francisco, says. 'All bacteria were clearly "non-self" so simply had to be recognized and dealt with. But the job of the immune system now appears to be far more nuanced and complex. It has to learn to consider our mutualists, our resident bacteria, as self too. In the future we won't even call it the immune system, but the microbial interaction system.'

The absence of constructive engagement between microbes and immune system (particularly during certain windows of development) could be behind the increase in autoimmune conditions in the West.

So why haven't we evolved our own systems to perform these most critical functions of life? Why have we outsourced all this work to a bunch of microbes? One theory is that, because microbes evolve so much faster than we do (in some cases a new generation every 20 minutes), they can respond to changes in the environment, to threats as well as opportunities, with much greater speed and agility than 'we' can. Exquisitely reactive and adaptive, bacteria can swap genes and pieces of DNA among themselves.

This versatility is especially handy when a new toxin or food source appears in the environment. The microbiota can swiftly come up with precisely the right gene needed to fight it, or eat it. Researchers found that a common gut microbe in Japanese people has acquired a gene from a marine bacterium that allows the Japanese to digest seaweed, something the rest of us can't do as well.

Such plasticity serves to extend our comparatively rigid genome, giving us access to a tremendous bag of biochemical tricks we did not need to evolve ourselves. 'The bacteria in your gut are continually reading the environment and responding,' says Joel Kimmons, a nutrition scientist and epidemiologist at the Centers for Disease Control and Prevention in Atlanta. 'They're a microbial mirror of the changing world. And because they can evolve so quickly, they help our bodies respond to changes in our environment.'

Bad wars on bugs

A handful of microbiologists have begun sounding the alarm about our civilization's unwitting destruction of the human microbiome and its consequences. Important microbial species may have already gone extinct, before we have had a chance to learn who they are or what they do. What we think of as an interior wilderness may in fact be nothing of the kind, having long ago been reshaped by unconscious human actions. Taking the ecological metaphor further, the 'Westernized microbiome' most of us now carry around is in fact an artifact of our particular type of civilization.

To obtain a clearer sense of what has been lost, María Gloria Dominguez-Bello, a Venezuelan-born microbiologist at New York University, has been traveling to Amazonia to collect samples from native peoples who have had little previous contact with Westerners or Western medicine. 'We want to see how the human microbiota looks before antibiotics, before processed food, before modern birth,' she told me. 'These samples are gold.'

Preliminary results indicate that a pristine microbiome, of people who have had little or no contact with Westerners, features much greater biodiversity, including a number of species never before sequenced, and, as mentioned, much higher levels of prevotella than is typically found in the Western gut. Dominguez-Bello says these vibrant, diverse and antibiotic-naïve microbiomes may play a role in Amerindians' markedly lower rates of allergies, asthma, atopic disease and chronic conditions like Type 2 diabetes and cardiovascular disease.

One bacterium commonly found in the non-Western microbiome is a corkscrew-shaped inhabitant of the stomach by the name of *Helicobacter pylori*. Dominguez-Bello's husband, Martin Blaser, a physician and microbiologist at NYU, has been studying *H. pylori* since the mid-1980s and is convinced that it is an endangered species, the extinction of which we may someday rue. According to the 'missing microbiota hypothesis,' we depend on microbes like *H. pylori* to regulate various metabolic and immune functions, and their disappearance is disordering those systems. The loss is cumulative: 'Each generation is passing on fewer of these microbes,' Blaser told me, with the result that the Western microbiome is being progressively impoverished.

He calls *H. pylori* the 'poster child' for the missing microbes. Medicine has actually been trying to exterminate it since 1983, when Australian scientists proposed that the microbe was responsible for peptic ulcers; it has since been implicated in stomach cancer as well, as a co-factor with diets high in salt. But *H. pylori* is a most complicated character, the entire spectrum of microbial good and evil rolled into one bug. It also plays a role in regulating acid in the stomach. Presumably it does this to render its preferred habitat inhospitable to competitors, but the effect on its host can

be salutary. People without *H. pylori* may not get peptic ulcers, but they frequently do suffer from acid reflux. Untreated, this can lead to Barrett's esophagus and, eventually, a certain type of esophageal cancer, rates of which have soared in the West as *H. pylori* has been zapped.

I have an interest here. When after a recent bout of acid reflux, my doctor ordered an endoscopy, I discovered that, like most people in the US today, my stomach has no *H. pylori*. My gastroenterologist was pleased. But after talking to Blaser, the news seemed more equivocal, because *H. pylori* also does us a lot of good. The microbe engages with the immune system, quieting the inflammatory response in ways that serve its own interests as well as our own. This calming effect may explain why populations that still harbor *H. pylori* are less prone to allergy and asthma. Blaser's lab has also found evidence that *H. pylori* plays an important role in human metabolism by regulating levels of the appetite hormone ghrelin. 'When the stomach is empty, it produces a lot of ghrelin, the chemical signal to the brain to eat,' Blaser says. 'Then, when it has had enough, the stomach shuts down ghrelin production, and the host feels satiated.' The disappearance of *H. pylori* may be contributing to obesity by muting these signals.

But what about the diseases *H. pylori* is blamed for? Blaser says these tend to occur only late in life. He says this microbe's evolutionary role might be to help shuffle us off life's stage once our childbearing years have passed. For those of us who want a long healthy life he has proposed not one but two unconventional therapeutic interventions: inoculate children with *H. pylori* to give them the benefit of its services early in life, and then exterminate it with antibiotics at age 40.

Easier said than done. These days Blaser is most concerned about the damage that antibiotics, even in tiny doses, are doing to the microbiome, and particularly to our immune system and weight. 'Farmers have been performing a great experiment for more than 60 years,' Blaser says, 'by giving subtherapeutic doses of antibiotics to their animals to make them gain weight.' Scientists aren't sure exactly why this practice works, but the drugs may favor bacteria that are more efficient at harvesting energy from the diet.

'Are we doing the same thing to our kids?' he asks. Children in the West receive, on average, between ten and twenty courses of antibiotics before they turn 18. And those prescribed drugs aren't the only antimicrobials finding their way to the microbiota. Scientists have found antibiotic residues in meat, milk and surface water as well. Blaser is also concerned about the use of antimicrobial compounds in our diet and everyday lives — everything from chlorine washes for lettuce to hand sanitizers. 'We're using these chemicals precisely because they're antimicrobial,' Blaser says. 'We need to ask, what are they doing to our microbiota?' Antibiotics have helped us to treat infectious diseases. But, as in any war, the war on bacteria appears to have had some unintended consequences.

Box 2

Jeff Leach's pizzas

In a different but associated mode, Jeff Leach is also a pizza parlour entrepreneur. His big and potentially commercial idea is that all the theory about what diet the human species is really evolved to eat, can be translated into one of the fast food favourites in the US – take-out pizzas. He has formulated a pizza whose crust is made with ingredients that he believes, based on his fieldwork with the remains of primaeval meals, are a basis for the best human food. Success also depends on he product being delicious and delivered promptly, in a cut-throat franchised business.

The following story appeared some time ago in a newspaper in New Orleans, in the first incarnation as 'World's Heatlhiest Pizza'. This approach was not altogether successful, so Jeff Leach and his partners started again with 'Naked Pizza', which has attracted capital and is expanding in the US, and also in Dubai with the more discreetly named NKD Pizza.

Jeff Leach says the real health culprit of the pizzas most people get from the big pizza delivery chains is the highly processed dough underneath all that tasty stuff. So his pizzas pile cheese, meat and vegetables on a crust that he and his partners promote as a fiber-rich tool for helping fix what's wrong with the typical US diet. This crust is made from at least ten different grains with the addition of inulin, a fiber-rich supplement extracted from chicory root and known as a 'smart fiber' for its benefits to the digestive system.

Leach says 'It's the fiber, stupid' In addition to this venture in the pizza business, Leach is an anthropologist and a science writer whose syndicated column is published in newspapers and magazines around the country and online. He has delivered a keynote address at Harvard University Medical School. Pizza, he says, is another delivery channel for him to talk about fiber.

'You get a pizza, but you're also getting education', Leach says at his take-out-only pizza shop in the Carrollton area of New Orleans. 'That's why we put messages on the box, messages on the menu, messages on our Web site. We're getting it to you any way we can'.

World's Healthiest Pizza is certainly the only shop I've ever seen that brings up the specter of death on its pizza boxes. 'Our pizzas will not kill you. Promise', it reads in bright red print. There's a lot more printed on there, offering a large-print primer on the effects of proper fiber intake on the digestive system, but with the type of urgency you usually get from religious tracts handed out on the street. Leach also describes his company's modus operandi as 'fanatical'.

'We hear things like, well, what if people copy you?' says Leach. 'That's exactly what we want. That's what we're trying to do. We want to raise the bar on what is the most popular food in America. We're trying to turn it on its head and make it healthy'.

The trouble with antibiotics

One of the more striking results from the sequencing of my microbiome was the impact of a single course of antibiotics on my gut community. My dentist had put me on a course of amoxicillin as a precaution before oral surgery. In a week, my impressively non-Western 'alpha diversity' had plummeted, and had come to look very much like the US average. My (possibly) healthy levels of prevotella had also disappeared, to be replaced by a spike in bacteroides, much more common in the West, and an alarming bloom of proteobacteria, a phylum that includes a great many weedy and pathogenic characters, including *E coli* and salmonella. What had appeared to be a pretty healthy, diversified gut was now raising expressions of concern among the microbiologists who looked at my data.

'Your *E coli* bloom is creepy,' Ruth Ley, a Cornell University microbiologist who studies the microbiome's role in obesity, told me. 'If we put that sample in germ-free mice, I bet they'd get inflamed.' Great. Just when I was beginning to think of myself as a promising donor for a fecal transplant, now I had a gut that would make mice sick. I was relieved to learn that my gut community would eventually bounce back to something resembling its former state. All the same though, one recent study has found that when subjects were given a second course of antibiotics, the recovery of their interior ecosystem was less complete than after the first.

General metabolic syndrome theory

The scientists I interviewed had little doubt that the Western diet is altering our gut microbiome in troubling ways. Some, like Blaser, are concerned about all the antimicrobials we're ingesting with our meals; others were concerned with the sterility of processed food products. Most agreed that the lack of dietary fiber in the Western diet was deleterious to the microbiome. Others voiced concerns about the additives in processed foods, few of which have ever been studied for their specific effects on the microbiota.

According to a recent article in *Nature* by the Stanford microbiologist Justin Sonnenburg, 'Consumption of hyper-hygienic, mass-produced, highly processed and calorie-dense foods is testing how rapidly the microbiota of individuals in industrialized countries can adapt.' As our microbiome evolves to cope with the Western diet, Sonnenburg says he worries that various genes are becoming harder to find, as the microbiome's inherent biodiversity declines along with our everyday exposure to bacteria.

Catherine Lozupone in Boulder and Andrew Gewirtz, an immunologist at Georgia State University, directed my attention to the emulsifiers commonly used in many processed foods. These are ingredients with names like lecithin, *Datem*, CMC, and polysorbate 80. Gewirtz's lab has done studies in mice indicating that some of these detergent-like compounds may damage the mucosa, the protective lining of the gut wall, potentially leading to leakage and inflammation.

A growing number of medical researchers are coming around to the idea that the common denominator of many, if not most, of the chronic diseases from which we suffer today may be inflammation, which is to say, a heightened and persistent immune response by the body to a real or perceived threat. Thus, various markers for inflammation are common in people with metabolic syndrome, the complex of abnormalities that predisposes people to illnesses like cardiovascular disease, obesity, Type 2 diabetes and perhaps some forms of cancer. While health organizations differ on the exact definition of metabolic syndrome, the name for the syndrome of conditions that include various of these diseases, a 2009 report from the Centers for Disease Control and Prevention found that 34 percent of American adults are afflicted with the condition. Is inflammation yet another symptom of metabolic syndrome, or is it perhaps the cause of it? And if it is the cause, what is its origin?

One theory is that the problem begins in the gut, with a disorder of the microbiota, specifically of the all-important epithelium that lines our digestive tract. This internal skin, the surface area of which is large enough to cover a tennis court, mediates our relationship to the world outside our bodies. More than 50 tons of food pass through it in a lifetime. The microbiota play a critical role in maintaining the health of the epithelium: some bacteria, like the bifidobacteria and *Lactobacillus plantarum* (common in fermented vegetables), seem to directly enhance its function. These and other gut bacteria also contribute to its welfare by feeding it. Unlike most tissues, which take their nourishment from the bloodstream, epithelial cells in the colon obtain much of theirs from the short-chain fatty acids that gut bacteria produce as a by-product of their fermentation of plant fiber in the large intestine.

But if the epithelial barrier isn't properly nourished, it can become more permeable, allowing it to be breached. Bacteria, endotoxins – toxic by-products of certain bacteria – and proteins can then slip into the blood stream, thereby causing the body's immune system to mount a response. This resulting low-grade inflammation, which affects the entire body, may lead over time to metabolic syndrome and a number of the chronic diseases that have been linked to it. That's the theory.

Evidence in support of this theory is beginning to accumulate, some of the most intriguing coming from the lab of Patrice Cani at the Université Catholique de Louvain in Brussels. When Cani fed a high-fat, 'junk food' diet to mice, the community of microbes in their guts changed much as it does in humans on a fast-food diet. But Cani also found the junk-food diet made the animals' gut barriers

notably more permeable, allowing endotoxins to leak into the bloodstream. This produced a low-grade inflammation that eventually led to metabolic syndrome. Cani concludes that, at least in mice, 'gut bacteria can initiate the inflammatory processes associated with obesity and insulin resistance' by increasing gut permeability.

These and other experiments suggest that inflammation in the gut may be the cause of metabolic syndrome, not its result, and that changes in the microbial community and lining of the gut wall may produce this inflammation. If Cani is correct, and there is now some evidence indicating that the same mechanism is at work in humans, then medical science may be on the trail of a Grand Unified Theory of Chronic Disease, at the very heart of which we will find the gut microbiome.

A new health dawn?

My first reaction to learning all this was to want to do something about it immediately, something to nurture the health of my very own personal microbiome. Most of the scientists I interviewed were reluctant to give me any practical recommendations; it's too soon, they told me, we don't know enough yet. Some of this hesitance reflects an understandable abundance of caution. The microbiome researchers don't want to make the mistake of overpromising, as the genome researchers did.

They are also concerned about feeding a gigantic bloom of prebiotic and probiotic quackery. Rightly so: probiotics are already being hyped as the new panacea, even though it isn't at all clear what these supposedly beneficial bacteria do for us, or how they do what they do. There is some research suggesting that some probiotics may be effective in a number of ways: modulating the immune system; reducing allergic response; shortening the length and severity of colds in children; relieving diarrhea and irritable bowel symptoms, and improving the function of the epithelium.

A problem is that, because the probiotic marketplace is largely unregulated, it's impossible to know what, if anything, you're getting when you buy a 'probiotic' product. One study tested 14 commercial probiotics and found that only one contained the exact species stated on the label.

But some of the scientists' reluctance to make recommendations surely flows from the institutional bias of science and medicine: that the future of microbiome management should remain firmly in the hands of science and medicine. Down this path, which holds real promise, lie improved probiotics and prebiotics, fecal transplants (with better names) and related therapies. Jeffrey Gordon, one of those scientists who peers far over the horizon, looks forward to a time when disorders of the microbiome will be treated with 'synbiotics'. These would be suites of targeted,

next-generation probiotic microbes administered along with the appropriate prebiotic nutrients to nourish them.

Then, the fecal transplant would give way to something far more targeted. This would be a purified and cultured assemblage of a dozen or so microbial species that, along with new therapeutic foods, will be introduced to the gut community to repair 'lesions' – important missing species or functions. Bur, alas, assuming it all works as advertised, such an approach will also allow Big Pharma and Big Food to stake out and colonize the human microbiome for profit.

When I asked Gordon about do-it-yourself microbiome management, he said he looked forward to a day 'when people can cultivate this wonderful garden that is so influential in our health and well-being'. That day awaits a lot more science. So he declined to offer any gardening tips or dietary advice. 'We have to manage expectations,' he said.

Alas, I am impatient. So I gave up asking scientists for recommendations. Instead, I began to ask them how, in light of what they've learned about the microbiome, they have changed their own diets and ways of life. Most of them have made changes. They were for example slower to take, or give their children, antibiotics. Some spoke of relaxing the sanitary regime in their homes, encouraging their children to play outside in the dirt and with animals, deliberately increasing their exposure to the great patina.

Many researchers told me they had eliminated or cut back on highly processed food products, either because of their lack of dietary fiber or out of concern about additives. In general they seemed to place less faith in probiotics (which few of them used) than in prebiotics – foods likely to encourage the growth of 'good bacteria' already present. Several, including Justin Sonnenburg, said they had added fermented foods to their diet such as natural yogurt, kimchi, or sauerkraut. These foods can contain large numbers of probiotic bacteria, like *L plantarum* and bifidobacteria. While most probiotic bacteria don't appear to take up permanent residence in the gut, there is evidence that they might leave their mark on the community, sometimes by changing the gene expression of the permanent residents, in effect turning on or off metabolic pathways within the cell, and sometimes by stimulating or calming the immune response.

Need to feed gut bugs

What about increasing our exposure to bacteria? 'There's a case for dirtying up your diet,' Sonnenburg told me. 'I view it as a cost-benefit analysis' he wrote in an e-mail. 'Increased exposure to environmental microbes likely decreases chance of many

Western diseases, but increases pathogen exposure. Certainly the costs go up as scary antibiotic-resistant bacteria become more prevalent.' So wash your hands in situations when pathogens or toxic chemicals are likely present, but maybe not after petting your dog. 'In terms of food, I think eating fermented foods is the answer, as opposed to not washing food, unless it is from your garden,' he said.

With his wife, Erica, also a microbiologist, Sonnenburg tends a colony of gnotobiotic mice at Stanford, examining among other things, the effects of the Western diet on their microbiota. (Removing fiber drives down diversity, but the effect is reversible.) He's an amateur baker, and when I visited his lab, we talked about the benefits of baking with whole grains.

'Fiber is not a single nutrient' Sonnenburg said, which is why fiber supplements are no magic bullet. There are hundreds of different polysaccharides' (complex carbohydrates, including fiber) 'in plants, and different microbes like to chomp on different ones.' To boost fiber, the food industry has added lots of a polysaccharide called inulin to hundreds of products, but that's just one kind (often derived from the chicory-plant root) and so may only favor a limited number of microbes. I was hearing instead an argument for a variety of whole grains and a diverse diet of plants and vegetables as well as fruits. 'The safest way to increase your microbial biodiversity is to eat a variety of polysaccharides,' he said.

His comment chimed with something a gastroenterologist at the University of Pittsburgh told me. 'The big problem with the Western diet,' Stephen O'Keefe said, 'is that it doesn't feed the gut, only the upper gastrointestinal tract. All the food has been processed to be readily absorbed, leaving nothing for the colon, the lower intestinal tract. But it turns out that one of the keys to health is fermentation in the large intestine.' And the key to feeding the fermentation in the large intestine is giving it lots of plants with their various types of fiber, including resistant starch (found in bananas, oats, beans); soluble fiber (in onions and other root vegetables, nuts); and insoluble fiber (in whole grains, especially bran, and avocados).

With our diet of swiftly absorbed sugars and fats, we're eating for one, just ourselves, and we and depriving the trillion, all our bacteria, of the food they like best, which is made up largely from complex carbohydrates and fermentable plant fibers. The by-product of fermentation is the short-chain fatty acids that nourish the gut barrier and help prevent inflammation. And there are studies suggesting that simply adding plants to a fast-food diet will mitigate its inflammatory effect.

The outlines of a diet for the new super-organism of us plus bugs, or rather us as also being bugs, were coming clear, and it didn't require the ministrations of the food scientists at Nestlé or General Mills to design it. Big Food and Big Pharma probably do have a role to play, as will Jeffrey Gordon's next-generation synbiotics, in repairing the microbiota of people who can't or don't care to simply change their

diets. This is going to be big business. Yet the components of a microbiota-friendly diet are already on the supermarket shelves and in farmers' markets.

Viewed from this perspective, the foods in the markets appear in a new light, and I began to see how you might begin to shop and cook with the microbiome in mind, the better to feed the fermentation in our guts. The less a food is processed, the more of it that gets safely through the upper gastrointestinal tract and into the eager clutches of the microbiota. Al dente pasta, for example, feeds the bugs better than soft pasta does; steel-cut oats better than rolled; raw or lightly cooked vegetables offer the bugs more to chomp on than overcooked. And so on. This is at once a very old and a very new way of thinking about food: it suggests that all calories are not created equal and that the structure of a food and how it is prepared may matter as much as its nutrient composition.

It is a striking idea that one of the keys to good health may turn out to involve managing our internal fermentation. Having recently learned to manage several external fermentations, of bread and kimchi and beer — I know a little about the vagaries of that process. You depend on the microbes, and you do your best to align their interests with yours, mainly by feeding them the kinds of things they like to eat. But absolute control of the process is too much to hope for. It's a lot more like gardening than governing.

Our garden within

The successful gardener has always known that you don't need to master the science of the soil, which is yet another hotbed of microbial fermentation, in order to nourish and nurture it. You just need to know what it likes to eat, which is basically, organic matter, and how, in a general way, to align your interests with the interests of the microbes and the plants. The gardener also discovers that, when pathogens or pests appear, chemical interventions 'work,' that is, solve the immediate problem, but at a cost to the long-term health of the soil and the whole garden. The drive for absolute control leads to unanticipated forms of disorder.

This, it seems to me, is pretty much where we stand today with respect to our microbiomes, our teeming, quasi-wilderness. We don't know a lot, but we probably know enough to begin taking better care of it. We have a pretty good idea of what it likes to eat, and what strong chemicals such as aniobotics and detergents do to it. We know all we need to know, in other words, to begin, with modesty, to tend our garden within.

Box 3

Jeff Leach's guts

Jeff Leach (left) in Africa with the !Kung people in search of uncontaminated guts

Jeff Leach writes: My interest in modern diet and the gut microbiome began a decade ago when my daughter was diagnosed with type 1 diabetes. As with other autoimmune diseases, an underlying genetic susceptibility must exist for type 1 diabetes to manifest, but an environmental component (trigger) is necessary. It's becoming increasingly clear that the gut microbiome plays a significant if not causal role in the development of type 1 diabetes, other autoimmune diseases, and modern (ecological) diseases in general.

In an effort to raise awareness about the changes in human ecology that have given rise to diseases of the modern world, with others I have launched the *Human Food Project.* See also our recently launched crowd sourcing project *American Gut*. We hope you can join us as we look to the past to better understand why we get sick.

My research interests include the impact of acculturation on the gut microbiome among traditional groups in Africa, public health policy, ethnography, indigenous rights, hot-rock technology, site formation processes, foraging and pastoral societies, and evolution of the human micro-biome from our Mio-Pliocene ancestors to modern primates and humans.

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