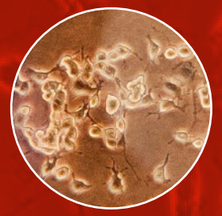
By Alex Hutchinson
Photographs by Marcelle Deichev

WHAT YOUR

GUT KNOWS



OUR MICROBIOME MAY HOLD THE KEY TO BRAIN DISEASES, STROKE RECOVERY AND MORE. NOVEL RESEARCH IS REVEALING SURPRISING CONNECTIONS

In recent months, headlines have trumpeted the news that our microbiome — the community of microorganisms that lives in and on our bodies — can be affected by factors and conditions as disparate as alcohol and artificial sweeteners. It is a familiar theme. Since the early 2000s, there has been a steady drumbeat of hype about the links between microbes and human health. Advances in DNA sequencing technology have made it possible to assemble huge catalogues of the bacteria present in any given organism. It is such an exciting and rapidly advancing field, says neurobiologist Eran Blacher, that it is easy to overlook a basic question: "What is the biological meaning of all this?"

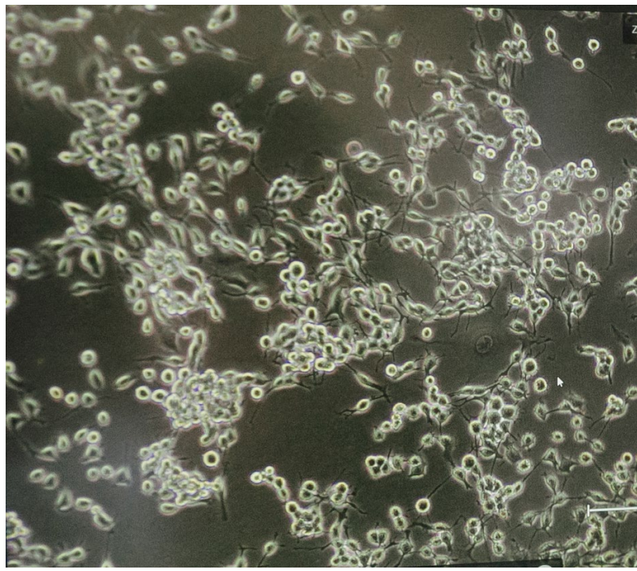
Blacher heads the Gut–Brain Axis Laboratory at the Hebrew University of Jerusalem, which he established in 2023 with support from an Azrieli Early Career Faculty Fellowship. He is interested in understanding brain maladies such as Alzheimer's disease, amyotrophic lateral sclerosis (ALS) and stroke, all of which are influenced by the two-way communication paths connecting the brain with the gut and its associated microbes. He has new findings, for example, linking stroke recovery to gut permeability (the extent to which the gut "leaks") in aging.

But he is not convinced that solutions will take the form of fortified yogurts or probiotic pills, or that simply making lists of what organisms are present in healthy and diseased microbiomes will get us there. "We don't want to spend our entire careers just cataloguing bacteria without knowing if they're doing something clinically useful or not," he says.

Instead, he's pursuing a more holistic and multidisciplinary approach to microbiome research that focuses on understanding the links between form and function, as well as tracking how that changes with age. And he's harnessing new techniques in the emerging field of spatial biology to make a big bet on the future of microbiome research: that it's not just which bacteria are present that matters, but also where they are.

Mirror image: Problems in the gut may reflect problems elsewhere in the body, says Erin Blacher, who heads the Gut–Brain Axis Laboratory at the Hebrew University of Jerusalem, established with support from an Azrieli Early Career Faculty Fellowship.

The gut is not just a digestive organ. It is a communications hub, relaying signals that influence how our brain operates, how our bodies age and how we recover from disease. That's why promising microbiome research focuses on both form and function



Inspired by nature from a young age, Blacher fell in love with the brain. "It's made out of atoms and molecules, and in that sense it's not different from any other object in nature. And yet it mediates wills and emotions and aspirations and motivations." (Above right) Cells from specially bred mice.

Blacher grew up in Holon, a suburb south of Tel Aviv, the son of a high school art history teacher and an urban planner. He was an obsessive reader, immersed in philosophy, literature and the arts, but he was also fascinated by nature. "I remember, from a very young age, looking down at the ground and digging for invertebrates and asking many, many questions," Blacher recalls. He was fascinated by the link between how things look and their function — one of the key questions in biology.

When the time came to go to university, he was torn between studying science or becoming a physician. He ended up choosing the former, in part because it promised to combine his fascination with the natural world and his more humanistic interests. Collecting data is a seemingly impartial process: you assemble objective facts and numbers that describe the world. "But the way we put those pieces together to construct an argument, connecting them to make the ideas flow, makes us a bit like storytellers or authors," he says. "I like the work of putting the scientific pieces together to unravel the mechanisms by which our body works."

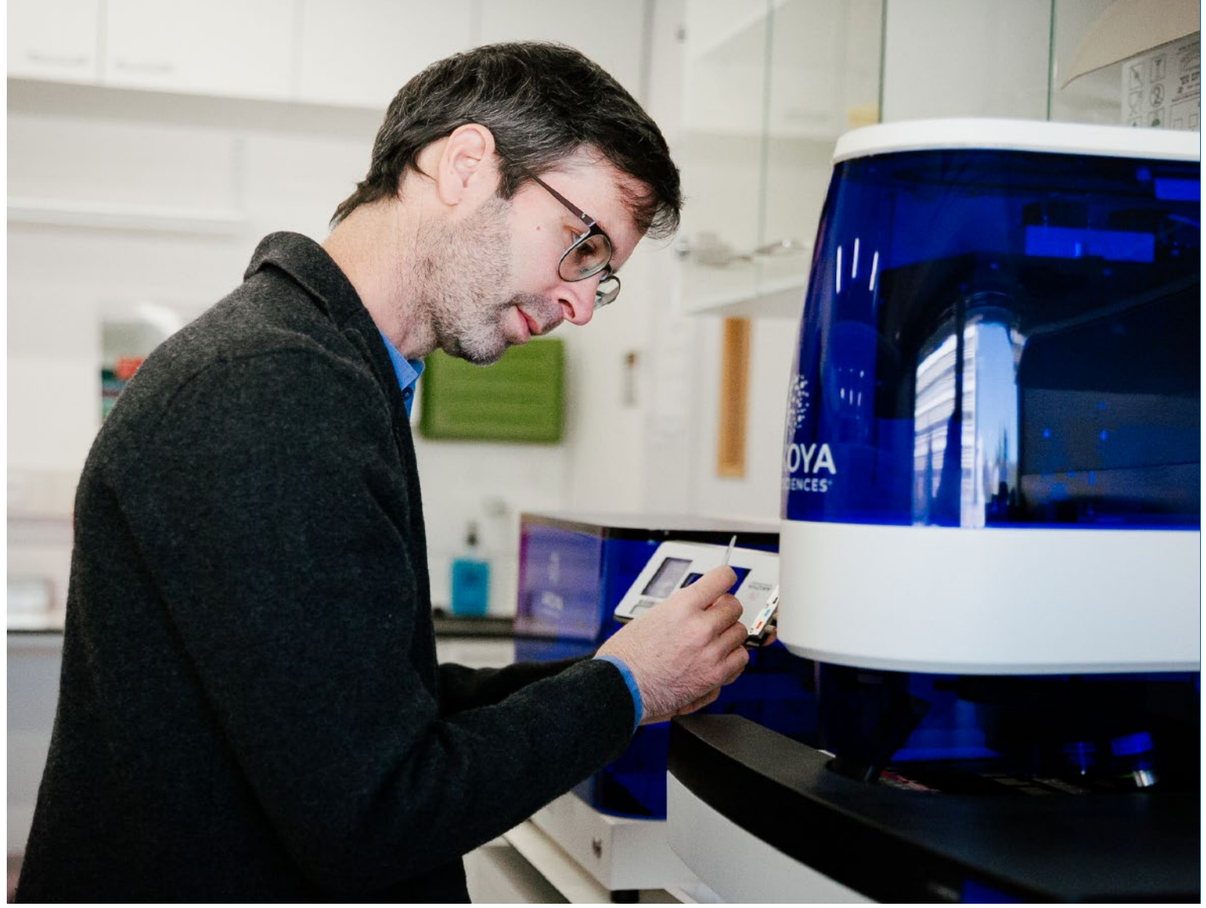
It was while studying undergraduate biology at Tel Aviv University that Blacher fell in love with the brain — its complexity and ineffability. "The brain is not that different from this table," he points out, gesturing at the surface in front of him. "It's made out of atoms and molecules, and in that sense it's not different from any other object in nature. And yet it mediates wills and emotions and aspirations and motivations. And we don't know how these atoms and molecules drive those very big concepts."

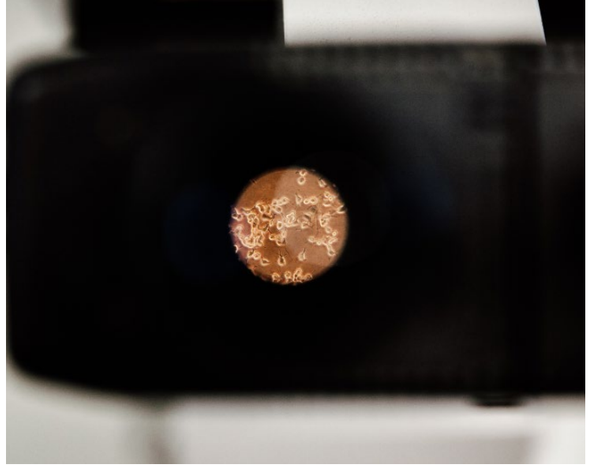
When Blacher finished his biology degree, he decided to stay at Tel Aviv University for a PhD in neurobiology, studying the brain's innate immune system. Unlike the so-called adaptive immune system, which targets specific pathogens and learns from experience, the innate immune systems functions as a rapid, non-specific first line of defence against disease.

Working in the lab of neurobiologist Reuven Stein between 2011 and 2016, Blacher studied how a class of innate immune cells called microglia influence the progression of diseases such as cancer and Alzheimer's, using pre-clinical models of these conditions. He found, for example, that a particular microglia-related enzyme accelerates the progression of glioma tumours, an insight that is still being pursued as a possible treatment target.

But by the time he finished his PhD, he couldn't shake off a feeling of disappointment. Despite all the progress scientists had made in understanding the genetics and mechanisms of brain diseases, there were still no cures and few effective treatments, and the rates of these diseases continued to rise. "So there must be something other than genetics," he concluded. "And I thought, what can explain this? Which part of the body takes information from the outer world, from the environment, processes it, and relays it to other systems in the body. And one of the best answers is the gut."

Armed with this insight, he moved to the Weizmann Institute of Science for a postdoctoral fellowship with Eran Elinav, one of the world's foremost microbiome researchers. It was a big leap. "I'm a brain scientist, and I want to study how microbes affect the brain," Blacher told Elinav by way of introduction. Combining strengths, they assembled a diverse team of microbiologists, mathematicians, computer scientists and other experts. Blacher was the team's





neurobiologist. “Each of us had a different question,” Blacher recalls, “but we all faced the same challenges, so we worked together.”

One of the projects Blacher worked on explored the link between gut microbes and ALS. Mice bred to have an ALS-like disease seemed to have different microbial strains in their gut compared to healthy mice. In particular, they had lower levels of bacteria that produce a molecule called nicotinamide. Inoculating the mice with more nicotinamide-generating bacteria or simply giving them nicotinamide directly improved their ALS-like motor symptoms. In humans, too, ALS patients turned out to have a different microbiome composition and lower levels of nicotinamide in their blood and brain fluids compared to healthy family members. The results of this study were published in Nature, and Blacher was awarded the NOSTER and Science Microbiome Prize. This line of research may yield a novel therapeutic approach, though the road to a successful clinical treatment remains long (see “Dude, Where’s My Probiotic Superpill?”).

Blacher’s experiences in Elinav’s lab cemented his interest in the gut–brain axis as a means of understanding brain conditions. But there was one final piece of his research agenda that clicked into place during a second postdoctoral fellowship, this one with neurologist Katrin Andreasson at Stanford University. His insight was that many of the conditions he was interested in — Alzheimer’s, Parkinson’s, ALS, stroke — share a common thread. “What is the biggest risk factor for all these diseases?” he asks. “Aging.”

The biology of older animals — their immune system, hormonal system, nervous system and so on — is very different from that of their younger counterparts. Something about the biological changes associated with aging makes us more susceptible to brain diseases as time passes. And yet the vast majority of basic research is conducted on young mice. They have been specially bred to have conditions akin to the diseases being researched, but the rest of their body is still operating as a young animal. During his time at Stanford, Blacher began using older mice aged between 20 and 22 months (equivalent to about 80 years old in a human), comparing their results to those of the more commonly studied younger mice.

This approach has already yielded promising insights. Doctors, for example, have found that older individuals who have a stroke are more susceptible to infections. “We all know that physicians may say, ‘I’m sorry, we did our best, but your father was very old and he got this infection, and we couldn’t save him,’” Blacher says. “But what’s the reason? I mean, being old is not an explanation. What really goes wrong when we age?”

Blacher’s gut-brain research shows the promise of spatial biology, an emerging field that studies where cells and molecules fit within an organism and what their environment says about their behaviour

Blacher’s use of advanced multiplexed imaging allows him to create a composite picture of many types of proteins and cells in a mouse’s gut (above, top and bottom). It’s a nuanced view of what’s happening in the body, and where. “In the gut, location matters a lot.”

In experiments comparing young and old mice experiencing the equivalent of a stroke, Blacher and others found that aged mice have a prolonged breach of the gut lining, allowing bacteria to leak into the blood and cause damage to the brain and elsewhere. There is an initial distress signal that travels from the brain to the gut via the nervous system that causes a biological “zipper” to open in the gut lining. In young mice, it zips closed again quickly. But in older mice, it stays open. Blacher and his colleagues have identified several candidate bacterial strains that appear to contribute to the prolonged gut leakage, opening the possibility of developing treatment approaches.

At the Hebrew University, Blacher’s Gut–Brain Axis Laboratory is combining the insights that he gained during his PhD and postdoctoral fellowships: the importance of communication between the gut and brain, the contribution of microbes to the gut’s function and the crucial role of aging. It is an approach with the potential to advance not just theoretical knowledge about the microbiome, but also clinical treatments that help patients, says Professor Zahava Vadasz, a physician-researcher at Technion–Israel Institute of Technology and the head of research and innovation at the Bnai Zion Medical Center in Haifa.

Vadasz says Blacher stands out in his relationships with people and willingness to collaborate. She says he is adept at combining different techniques, technologies and bodies of knowledge. “This integrative approach makes Eran’s projects so unique.”

Vadasz points to Blacher’s use of a novel technique called advanced multiplexed imaging. To image proteins and cells in, say, a cross-section of a mouse’s gut, standard imaging techniques involve using a handful of specially targeted markers to “illuminate” their location. But the gut is enormously complex, so seeing only a few types of protein gives a very incomplete picture. New multiplexing technology lets Blacher apply three markers, take a picture, then wash those markers away and apply three more markers, take another picture, and keep repeating the process until he has assembled a composite photo of a mouse gut with dozens or perhaps even a hundred different types of proteins imaged simultaneously.

This new approach provides a much more nuanced way of probing what is happening in the body, and where. “In the gut, location matters a lot,” Blacher says. Perhaps diseases are more likely when certain microbes are located in a particular place, or in proximity to certain other microbes, or when they communicate with certain immune cells. This spatial biology approach is still in its infancy. Figuring out how to image the bacteria themselves is an ongoing challenge, since they are not firmly attached to the tissue and can sometimes be washed away during the imaging process. But Blacher believes the approach has enormous potential to illuminate the questions about structure and function that he first wondered about as a child, digging in the dirt for invertebrates. And even if the path isn’t straightforward, which it almost certainly won’t be, he is prepared for it. In fact, he is looking forward to it.

“This is the essence of being a scientist,” he says. “After you fail so many times, and your hypotheses, as beautiful as they may be, clash with reality and are proven wrong — we have beautiful ideas, but the mice never seem to agree with them — then suddenly you have this eureka moment where for a very short period of time, you know something about the world that nobody else knows. “These small moments make the arduous scientific journey worthwhile and rewarding.” ▲●■

DUDE, WHERE'S MY **PROBIOTIC SUPERPILL?**

There is no doubt our health is heavily influenced by the microbes living in our gut, with certain diseases linked to the presence or absence of certain combinations of bacteria. But that does not mean we can simply eat the "good" bacteria in the form of a probiotic supplement, says microbiome researcher Eran Blacher.

For one thing, most strains of gut bacteria cannot survive in an oxygen-rich environment outside the body. "The probiotics that you see everywhere, they are just a big group of Lactobacillus species that are in found in yogurt," Blacher says. "They're not harmful, but they may not be so beneficial either, although they can be formulated and capsulated, and you can keep them on the shelf and sell them."

It is also hard to predict what would happen once you take a probiotic supplement. Some strains of bacteria may colonize the gut if you ingest them, while others may simply be washed away. That outcome could depend on your diet and exercise habits and other individual differences. And once established there, the bacteria could function differently in one person's gut compared to another's. "So even if we identify the magic bug that delays aging and solves all the problems," Blacher says, "it will be very hard to manage it as a drug."

Instead, the real promise of probiotics, and of microbiome research generally, is as a tool to identify metabolic or immune-related pathways and molecules, and test promising drugs. Blacher's ALS research found that ALS patients had lower levels of gut bacteria that produced a molecule called nicotinamide. "We would never, ever have dreamt that nicotinamide had a role in the context of ALS without the microbiome study," he says. "But now that we have identified nicotinamide as a potentially beneficial molecule in ALS treatment, we don't need the nicotinamide-producing bacteria anymore. We can treat ALS-prone mice with nicotinamide directly and delay the disease's symptoms." ▲●■