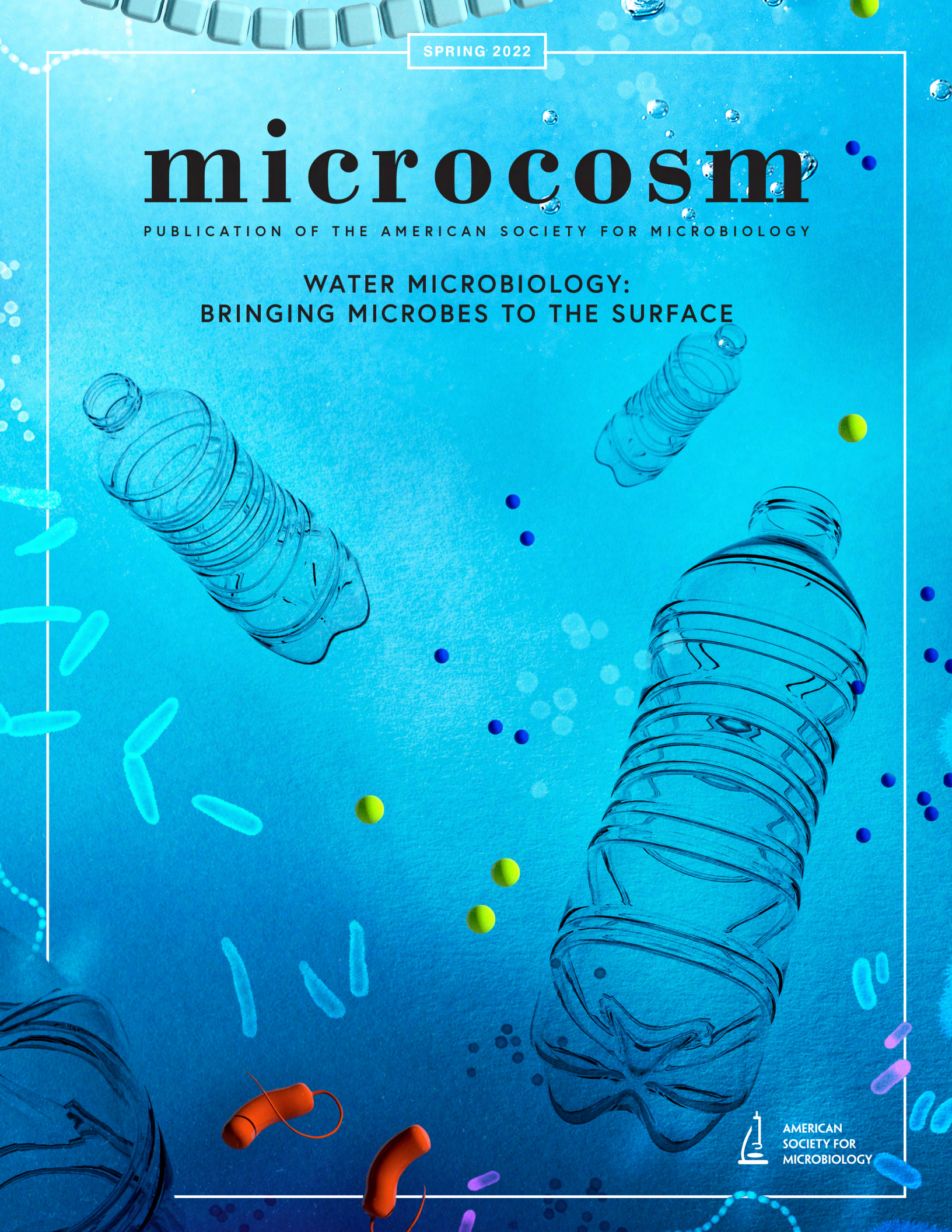


SPRING 2022

# microcosm

PUBLICATION OF THE AMERICAN SOCIETY FOR MICROBIOLOGY

WATER MICROBIOLOGY:  
BRINGING MICROBES TO THE SURFACE



AMERICAN  
SOCIETY FOR  
MICROBIOLOGY

# microcosm SPRING 2022

Explore the latest groundbreaking research in the microbial sciences, stay up-to-date with what's happening at ASM and read cutting edge scientific articles in *Microcosm*, ASM's flagship, members-only magazine.

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# From the Editor

BY STANLEY MALOY, PH.D., EDITOR-IN-CHIEF

In 1936, the Society of American Bacteriologists, the precursor to the American Society for Microbiology, began publishing a newsletter to inform members about Society affairs. The newsletter evolved, over time, to include feature articles and book reviews. After the name change to the American Society for Microbiology in 1960, the membership newsletter transitioned into the monthly *ASM News* magazine.

*ASM News* served as the ASM membership magazine for many years. It included information about board activities and branch meetings; membership updates, including letters, book reviews and obituaries; and short articles on new discoveries and the history of microbiology.

I've enjoyed our membership magazine from the time I was a student. Autobiographical stories like Stanley Falkow's "I'll Have the Chopped Liver Please, or How I Learned to Love the Clone," Amy Vollmer's "In Love With My Job," and Jon Beckwith's "Genetics at the Institute Pasteur: Substance and Style," provided fun insights into a wide variety of scientific journeys. Historical perspectives, like Lynn Margulis and Betsy Palmer Eldridge's "What a Revelation Any Science Is!," and descriptions of ASM Milestones in Microbiology sites, provided broad insights into the development of our discipline. As a bibliophile, I always consumed the book reviews, admiring those that provided interesting insights rather than a pedantic book report. And the letters, often written in response to a particular peeve about nomenclature, were great fodder for conversations with lab mates.

Important, but less enticing, were the descriptions of Society business, which were about as exciting as reading the tax code. Likewise, many of the feature articles seemed like dehydrated versions of a review article, like early astronaut food that provided essential nutrients, but wasn't very satisfying.

In response to feedback from readers, *ASM News* was reinvented as the magazine *ASM Microbe* in 2006. Like its predecessor, *ASM Microbe* was published monthly and included ASM leadership and membership news and articles about microbiology topics. Taking over from my good friend Elio Schaechter, I began as chair of the editorial advisory board of *ASM Microbe* in 2012. As *ASM Microbe* continued to evolve, we sought to focus on feature articles that had broader appeal, and in 2016, we began regular editorials to provide thought-provoking perspectives about microbiology.

The ASM General Meeting was renamed ASM Microbe in 2016, and *ASM Microbe*, the magazine, was reinvented as *Microcosm* the following year. I followed the transition from *Microbe* to *Microcosm* and have had the pleasure of serving as editor in chief since then.

Along with the change in name, we reimagined the style and substance of our membership magazine. *Microcosm* was redesigned from bottom to top, and its content became focused on advances in microbiology and inspirational stories about members that reflect the "big tent" of microbiology. The news sections shifted to the ASM website, allowing Society news to be updated as it happens. These changes were made in response to feedback from the ASM membership, and implemented by the creative, collaborative efforts of the ASM communications staff.

The first issue of *Microcosm* was focused on transformations, reflecting both the changes in *ASM Microbe*, as well as the many other transformational shifts at ASM. Each of the following issues has focused on a particular concept, like picturing the diversity of the giants of microbiology in Fall 2017, the intersection of science and technology in Spring 2019, the beneficial roles of microbes in Spring 2020, the ins and outs of vaccines in Fall 2020, One Health in fall 2021 and the microbiology of water in this issue. Now published biannually, each issue includes interesting reads that are timely, succinct and integrate multiple aspects of microbiology. And, like ASM Journals, *Microcosm* has shifted to an online format to allow for direct links to other resources and increased member access.

Your input and suggestions will ensure that *Microcosm* continues to evolve as a membership magazine that engages new and long-time ASM members from the many subdisciplines of microbiology. Please give us your feedback in-person at ASM Microbe in Washington D.C., send an email or connect via social media. I look forward to hearing from you!

**Stanley Maloy, Ph.D.**  
*Microcosm Editor-in-Chief*

# Microbes and Meltwater

BY STEPHEN ORNES

*Melting glaciers in Antarctica.* [Source](#)

**G**laciers are vast, frozen rivers of snow, ice and rock, collected often over thousands, even hundreds of thousands, of years. As an area of scientific research, glaciers have long landed in the domain of physical geochemists. It's easy to see why: these researchers study glaciers to investigate how water shapes the earth and track the increasingly observable signs of climate change. Many liken the annual growing and shrinking of glaciers to the deposits and withdrawals of a watery bank account, and warn that warming is driving the world dangerously close to being overdrawn.

But physical geochemists are not the only ones with a vital stake in predicting what the future will bring. Over the last 20 years or so, a growing group of microbiologists has also taken a critical interest in glaciers, due to the tiny passengers hitching a ride on these dense bodies of ice. Microorganisms can flourish on top of ice, beneath its surface (between the ice and the land) or even within—trapped, but not dead. Glaciers are ecosystems; they are not devoid of life. When they melt, their inhabitants change too.

"Most people think, 'It's frozen, it's dead, there's nothing there.' But [glaciers are] not just frozen nothingness," said microbiologist Dr. Christine Foreman, Ph.D., a professor at Montana State University in Bozeman, whose work looks at ways that microbes and climate change shape each other. "We now have this ability to think of glaciers as living systems. There's a deep reservoir of living things there." The deeper one goes, the older the ice, and presumably the more ancient the microorganisms. That means that to drill deep into the ice is to travel back in time. Studying glaciers can reveal both the biodiversity of life today and how it has shifted over centuries.

Glaciers hold roughly 75% of the freshwater on the planet, and as temperatures rise due to climate change, these "ice rivers" are quickly shrinking—in ways that will change how water cycles through the environment. Rates of retreat, and the impacts on life, vary by location. For example, according to [research published in December 2021](#), glaciers in the Himalayan mountains, which run along the northern edge of the Indian subcontinent, are shrinking at an exceptionally fast rate. Given that glaciers feed rivers that supply water to more than 1 billion people, more melting may limit accessibility to drinking water and accelerate disasters like floods and landslides. In Antarctica, which has the most glaciers worldwide, the loss of large amounts of ice will likely boost sea levels. So will the melting of Greenland's glaciers. A rising sea threatens coastal communities by eroding beaches and increasing the risk of devastating floods.

Melting glaciers won't only impact human communities; they'll also reshape microbial life. Viruses, bacteria and fungi that have been buried for hundreds of thousands of years could again see the sun. Those that live in, or on, the ice may be washed downstream by meltwater, changing the biological composition of the ecosystems they leave behind and the ecosystems where they land.

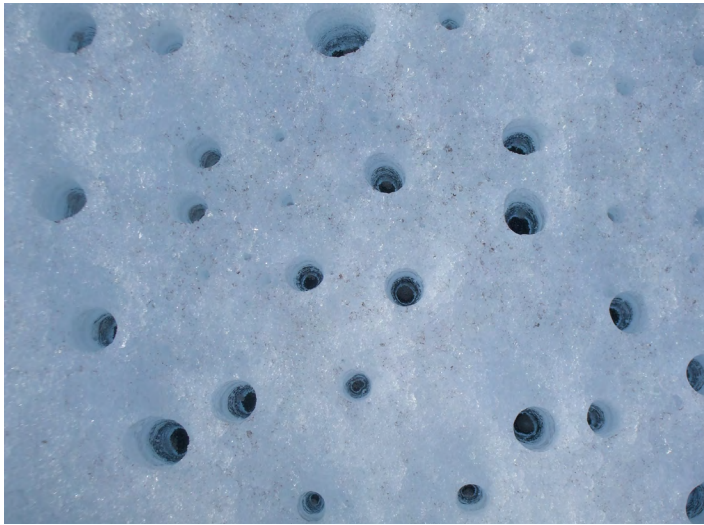
Microorganisms could also influence the changing climate in ways that aren't well understood. For example, they might release methane, a potent greenhouse gas or, through biological-physical interactions, accelerate the rate at which snow and ice melt. "There's essentially an amplifying process that we haven't accounted for," said microbiologist Dr. Arwyn Edwards, Ph.D., a researcher at Prifysgol Aberystwyth University in Wales, who has been studying ways that meltwater connects microbial populations in Greenland and Antarctica. "Glaciers respond to anthropogenic climate warming in ways that are entirely predictable. And then biology starts to drive feedbacks."

The influence of minuscule microbes on big climate processes has long gone unappreciated—and likely underestimated. "We have yet to move past the tip of the iceberg in terms of understanding the diversity of microbes and microbial interactions in those environments," said Edwards. But there's a shift underway, as a growing number of studies investigate life on ice and how it's changing. "These organisms can be searingly important," said Edwards. "They are a missing piece in the jigsaw puzzle."

## MICROBIAL LIFE ON GLACIERS, BUT NOT FROM SPACE

Today's efforts to connect meltwater to microbes can, in a way, be traced back to a startling discovery made [more than 150 years](#) ago by Adolf Erik Nordenskiöld, a Swedish-Finnish aristocrat, scientist and explorer. Nordenskiöld was the first explorer to cross the Northeast Passage; throughout his career, he led 10 expeditions to the Arctic north. During one journey in 1870, he observed that the glacial ice surrounding his team wasn't a uniform field of white. Instead, it was pitted with small dimples, each 1-3 feet deep and containing a dark mixture. He called it a "clayey mud," and worried that hidden holes (like potholes on an interstate) posed a danger to his expedition.

He named the mud "kryokonite" (the word is now spelled cryoconite) and speculated about its origin. Because the dimples were evenly distributed, he ruled out the possibility that the material had washed down from surrounding mountains or been deposited by rivulets of flowing water. He rightly concluded that the substrate had been carried by the wind; he also wrongly concluded that the presence of iron meant the goo included cosmic dust from space. When Sven Berggren, a Swedish botanist who was along on the voyage, examined the cryoconite, he observed a "peculiar ice-flora, consisting of a quantity of microscopical plants (algae)." Snow algae wasn't new; observations of snow-fields colored pink, red or green by various types of algae—sometimes called "watermelon snow"—[date back to Aristotle](#). Clearly, the cryoconite could hold life.



Cryoconite in Greenland. [Source](#)

However, Nordenskiöld saw something else in the small holes scattered across the ice. He surmised that the dark color of the cryoconites absorbs more heat from the sun than the bluish ice and, as a result, would accelerate the melting of the ice sheet. "Undoubtedly we have, in no small degree, to thank these organisms for the melting away of the layer of ice which once covered the Scandinavian peninsula," [he wrote in an 1883 publication](#).

Nordenskiöld's ideas were among the first to connect the glacial microbes to larger climate processes. Now, scientists have a better handle on the finer details. Gusts of wind deposit dust from local and distant sources, from industrial emissions and natural landscapes. The dust coats the snow and becomes trapped by filamentous cyanobacteria with super-sticky polysaccharides on their surfaces. "Cryoconite microbes do a good job of organizing that dust," Edwards said.

Recent studies have helped researchers gauge the contribution of snow algae to glacial melting. One gauge of that contribution is how algae changes the albedo, which measures the degree to which a material can reflect radiation from the sun. Snow, which has a high albedo, can reflect up to 90% of incoming sunlight. Cryoconites, which have a low albedo, absorb much of that energy, heat up as a result and, necessarily, heat the surrounding snow as well (a newish term for their contribution is "bioalbedo").

An international, interdisciplinary team of microbiologists, glaciologists and others recently used satellite data, runoff modeling and other tools to try to estimate the microbial contribution to melting. Their study, [published in January 2020](#), found that algal growth in 2017 boosted annual runoff from bare ice by around 5 gigatons, more than 10% of the total. In patches dense with biomass, the algae accelerated melting by more than 25%.

"The dark color of many species of snow algae," said Edwards, "acts as protective sunscreen for the microbes, shielding them from harmful radiation." But it's also clearly contributing to glacial warming. "They produce meltwater; they reduce the albedo," he explained, adding that climate models don't always account for this contribution, but they should.

## WAKING UP MICROBES FROZEN WITHIN GLACIERS AND ICE SHEETS

Microbes can also be trapped within glaciers and ice sheets, but just a few decades ago it would have been difficult to convince anyone of their existence. "Until 15-20 years ago, no one really thought there was life in ice," said Dr. John Priscu, Ph.D., Regents Professor Emeritus at Montana State University in Bozeman and senior research scientist, Polar Oceans Research Group. Priscu has been studying microbes in icy environments for more than 40 years.

Priscu helped bring attention to those frozen critters. In the 1990s, Priscu and his team studied an ice core collected by Russian drillers from more than 3,500 meters beneath the ice's surface—and about 150 meters above Lake Vostok, a subglacial lake discovered in the 1970s. The core represented lake water that had frozen, or accreted, on the bottom of the ice sheet. Antarctica has about 400 known subglacial lakes,

but Vostok is believed to be the deepest (at about 1,000 meters) and oldest. It may have formed more than 15 million years ago, even before the continent was covered in ice. When Priscu's group thawed the core and studied the meltwater, they [discovered](#) genetic material from bacteria and showed that the organisms were capable of active metabolism. The microbes could have lived in the lake and been trapped in water that had accreted to the bottom of the glacier, providing a glimpse of life in an icy world that has not seen sunlight for many millions of years, or they may have blown across the ice when the glacier was forming.



Ice sheet in Greenland. [Source](#)

The Lake Vostok ice cores ignited interest in deep, frozen life, and since then, researchers have not only sequenced the genomes of the collected microorganisms to gauge biodiversity, but also analyzed the abundance and metabolic potential of living things in ice cores taken from within glaciers and ice sheets around the world.

In ice cores, "we indeed have a time machine—2 miles [deep] of time machine," Priscu said. "When a microbe gets blown onto the surface of the ice, it eventually becomes part of the climate record as new layers form at the surface. It's like cosmic fly paper." Later, when scientists thaw that ice, sometimes the microbes wake up. "We can take ice that's 300,000 years old and melt it, and in 5 minutes they're metabolizing."

Studies of ice-dwelling microbes also reveal chemical clues to past and present climate patterns. Foreman, a former postdoctoral student of Priscu's, recently launched a project to study englacial ice, designed to learn what exactly those microbes are doing. "What if they are actively respiring in there?" she asked.

Though fears of ancient zombie pathogens have largely gone unfounded, more of these microbes will again see the light of day and be ferried away by meltwater. "As ice sheets are melting and ice shelves are breaking up, the melted water goes into the ocean," Priscu said. "It's taking these ancient genomes and pumping them back into the sea." But that mixing raises an important question: what happens when ancient microorganisms find themselves in a new ecosystem, commingling with newer ones? Can they live in that habitat?

This process is an example of what scientists call "genomic recycling," and it's not limited to the Southern Ocean. It happens globally, even at high altitudes. "In the Himalayas, we're getting a lot of mixing too," Priscu said. But in most cases, it's still not clear where the microbes end up, or what happens next. "What we do know is that ice-bound microorganisms can tell us what the climate was like when they were deposited at the surface.

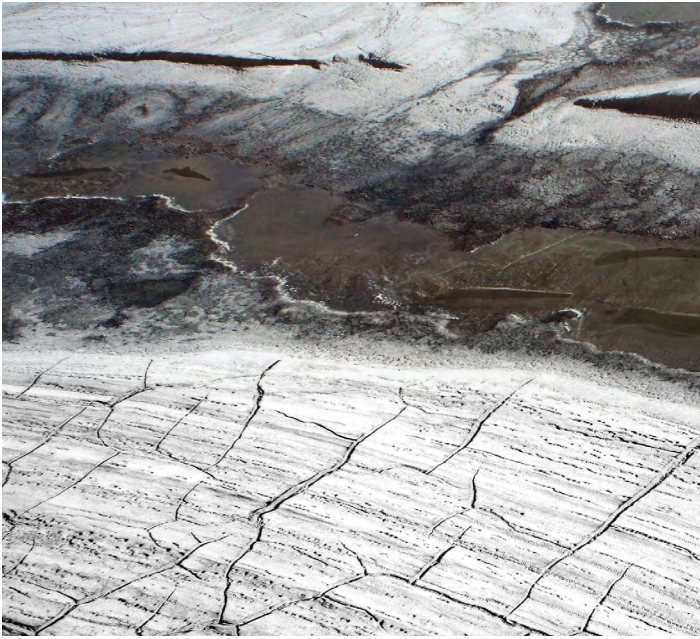
## TRACING THE THROUGHLINE

One place where scientists are trying to answer questions about melting and mixing is in and around the fjords of the Svalbard archipelago in Norway. These stunning, watery and remarkably deep channels wind through craggy mountains and pass by enormous glaciers. When the glaciers melt, the water drains into the fjord. Microbiologist Dr. Karen Lloyd, Ph.D., an associate professor at the University of Tennessee at Knoxville, wants to know how shrinking glaciers will shape microbial communities living on the floor of the fjords.

"We don't totally know what's happening with the retreat," she said. In the Svalbard fjords, glaciers play a critical role in feeding the microbial communities living at the bottom. "They sort of bulldoze iron into the seafloor," she said. But that influx of iron doesn't cause an algal bloom on the surface; instead, the iron settles in the deep, where it is processed by microorganisms. "The question is: What's going to happen when the bulldozers stop bulldozing?" said Lloyd.

"If glaciers stop dumping iron in the water, more of the mineral might be available to the microbes on land," she said. They may flourish, but right now it's not clear how the shifting availability will affect climate change. If these land-dwelling microbes became more active and produced more greenhouse gases, for example, they could amplify warming.

As part of her work, Lloyd is also collaborating with scientists who are looking for connections between land microbes and those that live at the bottom of the fjords. In March 2021 and 2022, she and her collaborators traveled by snowmobile to drill through frozen permafrost. The soil in Svalbard isn't as carbon-rich as that in places like Siberia, so scientists don't know as much about how the exposure and thawing of permafrost will affect the atmosphere.



A permafrost pattern in the Arctic. [Source](#)

Permafrost microbes may produce carbon dioxide, methane or possibly both. Yet how much of these compounds they produce—and at what rates—remains unclear. By studying carbon-degrading genes in the microbes, researchers may be able to answer a slew of lingering questions around which genes control the activity of microbes and attenuate in their CO<sub>2</sub> production. Predictions of carbon emissions from thawing permafrost are often based on gross carbon levels, but those predictions could be more accurate if they take into consideration the constraints posed by microbes.

For almost every ice-related process known to scientists, a better understanding of microbes and meltwater will improve models of how life in ice will affect and respond to global warming. Researchers in the U.K. are now quantifying the bioalbedo and incorporating the results into models of the Greenland Ice Sheet to determine the magnitude of ice melt caused by microbes. Right now, its contribution remains unclear. "You might have 2 or 3 data points but can't extrapolate to the entire system," Foreman explained. For a more accurate idea, researchers simply need more data.

And to better understand the influence of microbes within and beneath melting glaciers, she said, "we need more ice, more material." Priscu agreed, adding that a typical sample of meltwater from an ice core contains only about 1,000-10,000 organisms per milliliter. In order to collect enough data (e.g., genomic sequences, enzymatic analyses), researchers will need access to more cores removed from some of the planet's most remote places. "Including microbial research in studies of those cores," Priscu said, "is critical to understanding how microorganisms participate in global systems."

"Arctic and glacier environments are changing rapidly," said Edwards, the microbiologist from Wales. "The big question now is, how do these microorganisms matter, in a century where we will see a significant increase in climate change, for every human on the planet? Our overriding priority is on understanding this impact."

# Beware the Plastics

BY GEOFF HUNT, PH.D.

[Image Source](#)

**W**e live in a world of plastic. More than 400 million tons of the stuff, used in materials ranging from packaging to medical devices to automobile parts, [are now produced every single year](#). This staggering amount of pollution is having a deleterious impact on the environment, food chains, disease prevention and the global economy. Plastic is a health hazard to marine animals, a potential toxin to humans and a disruptive presence to seaborne commerce. What role can the microbiology community play in helping to ameliorate, and ultimately solve, this challenge?



Plastic waste from the ocean washed up on the shore. [Source](#)

## HOW MUCH IS ENOUGH?

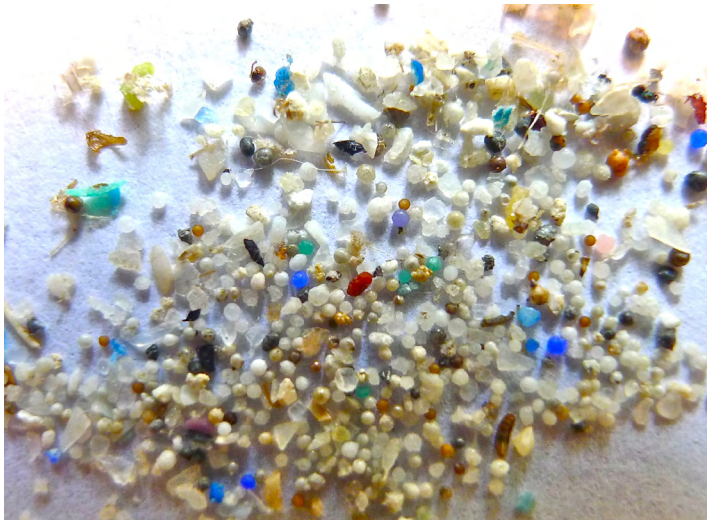
[Less than 10% of plastic material ends up getting recycled](#). The rest is either sitting in landfills around the world or clogging up the planet's marine environments. [Plastics decompose through several pathways](#), including physical breakdown, chemical reactions and biodegradation by microbes. As plastic degrades and decomposes on land, it eventually finds its way into waste streams and groundwater, [ultimately being carried by rivers to the world's oceans](#). Plastic was [first reported in oceans in 1972](#); 50 years later, the amount of plastic entering the sea is [estimated to be somewhere between 5 and 13 million metric tons annually](#).

Mainstream attention related to marine plastic pollution tends to focus on visible "trash islands." The most notorious example is the "Great Pacific Garbage Patch" (GPGP), located in the Pacific Ocean and [estimated to be approximately 1.6 million km<sup>2</sup> in size \(and growing\)](#). However, these observable pockets are only scratching

the surface of the problem. Studies estimate that barely 0.5% of aqueous plastic is on the ocean's surface. Much like an iceberg, [the bulk volume is either lying beneath the surface or has sedimented on the ocean floor](#).

Scarily enough, all of these estimates, which surely undercount the true scope of the problem, also fail to account for microplastics, [plastic particles that are less than 5 millimeters in diameter](#). Generated primarily by the fragmentation and breakdown of bulk items like water bottles, plastic bags and automobile tires, these invisible pollutants are exponentially more difficult to detect and remove due to their small size.





Microplastics found in the sand. [Source](#)

## MICRO PROBLEMS

Microplastics in the ocean also create artificial surfaces to which microbes can sorb. Researchers are working to characterize the nature of these interactions, and the findings have been far from conclusive. [Several reports](#) have shown that different types of aqueous microplastic surfaces attract different microbial species, with [distinct microbial colonies being found between polyethylene, polystyrene and polypropylene surfaces](#).

Other research [shows either no effect of microplastic composition](#) on the makeup of the adherent microbial community, or [ascribes effects to microplastic morphology](#) rather than composition. In some studies, [a distinct geographic microbial signature has been observed](#) based on where microplastic samples were collected; in others, there was [no reported difference in adherent microbial surface makeup](#) between different sample sites.

Perhaps a more important question is, what are microbes doing on these microplastic surfaces? One rising concern within the scientific community is that microplastics found in bodies of water can provide new platforms for biofilm formation. Unfortunately, this fear seems to be playing out. A recent report demonstrated that [bacteria congregating on aquatic microplastics engaged in increased amounts of horizontal gene transfer](#) compared to either free-living bacteria or microbes aggregating on natural surfaces. The obvious implication, on which the authors speculate, is that this behavior will lead to increased spread of antimicrobial resistance (AMR) genes, already a major challenge being tackled by the microbiology community.

## HOW TO RESPOND

With so many different actors and such variable results coming in from around the world, a critical first step for the field would be to agree upon common methodologies that could be used to conduct experiments about microbial behavior connected to plastics. As Dr. Nicole Fahrenfeld, Ph.D., associate professor of Civil and Environmental Engineering at Rutgers University, points out, "If we have a contaminant (i.e., microplastic) that can move to all these different places, it would be useful to have some universal standards in order to collect information from across the world."

Unfortunately, according to Fahrenfeld, "sampling and analysis methods for microplastics themselves are still in development." Because current methods for monitoring, studying and reporting these phenomena are not consistent across locations and institutions, "there's a pretty wide range of information in databases of microplastic occurrence," Fahrenfeld said.

Merely improving communication efforts will not be enough. Combating the microplastic-induced spread of AMR will require going beyond [current efforts searching for new antibiotics](#). The looming sense of urgency around this multi-faceted issue suggests the need for a more radical approach. One idea is to experiment with the selective deployment of genetically engineered microbes that could potentially out-compete the disease-causing organisms.

Elise Phillips, a researcher at the University of Tennessee Knoxville, suggests "shifting to actual application of knowledge" about the organisms that are responsible for spreading AMR genes, especially those found sorbed to microplastics. "How," she asked, "do we use these communities or alter them in a way that will help us with the problem" of microplastic-induced spread of AMR? Phillips suggests that researchers investigate this line of inquiry as a way to kickstart the search for potential solutions.

## PUTTING MICROBES TO WORK

Such radical problem-solving approaches are already taking place in other spheres, and they are aimed at eliminating the plastic that already exists while also minimizing (or altering) the production of new plastic products. A large amount of research is currently focused on [characterizing](#) and [applying the capacity of microbial enzymes](#) to break down plastics, in particular the widely used polyethylene terephthalate (PET). For example, a [2016 report from researchers in Japan](#) identified two enzymes, PETase and MHETase, that are capable of breaking down PET into molecules that can be metabolized by different microbes.

However, microbial degradation of plastics, which is dependent on a wide variety of biological, chemical and environmental factors, is [highly variable in terms of its efficiency](#). Simply allowing this process to proceed naturally will not remove plastic pollution at a scale, or over a timespan, that would allow humans to continue with their way of life. Instead, microbiologists are hard at work looking for ways to kick this process into a higher gear. Researchers are [ramping up their efforts to identify previously unknown microbes and novel enzymes](#) that can contribute to the biodegradation process. Meanwhile, other scientists are [using machine learning to design novel biodegradation enzymes](#) that can be deployed at scale.

## SPEEDING THINGS UP

Unfortunately, science moves slowly, and its findings and solutions tend to have limited reach beyond the scientific community without external interventions. This is where policy comes in. Adoption of science-based policies, whether at the local, national or international level, can have significant impact by raising mass awareness of an issue and effecting change quickly on a large scale.

An example of a simple, yet effective, policy that has had a huge impact with respect to plastic pollution has been the implementation of plastic bag tax laws. [Municipalities around the world have instituted regulations](#) that charge consumers a nominal amount (usually on the order of \$0.05-\$0.10) for each plastic bag they use when shopping. While the cost is small, the mere thought of having to pay for a bag apparently is enough of a nudge to induce widespread behavioral change. [Studies indicate](#) that taxes adopted in Chicago led to a 30% reduction in plastic bag use, while a similar policy adopted in Montgomery County, Md., led to a 42% drop. Other research shows that [implementing bag taxes is correlated with significant decreases in the volume of plastic bags collected](#) from municipal waterways, suggesting that the taxes are working as intended.

Higher-level policy change is also in the works. In March 2022, the United Nations announced that 175 countries will be working to [develop a legally binding agreement on ending plastic pollution](#). Signatories will look to promote sustainable production and consumption of plastics, enhance the development of comprehensive measuring and reporting tools on plastic pollution, and implement educational and awareness efforts [at both the national and international levels](#). Such international agreements have worked in the past. The most notable (and successful) model is the [1987 Montreal Protocol](#), which outlined concrete steps, such as phasing out the use of hydrochlorofluorocarbons, that have put the planet on track to have a fully restored ozone layer by 2050.

Outside of the lab and beyond the political arena, microbiologists can contribute through their personal actions. Potential steps include using fewer plastic products, making sure to recycle when possible, and [participating in clean-up days](#) that remove trash from the environment and prevent plastics from getting into the water supply in the first place.

Cleaning up the Earth's plastic problem may seem intractable, yet achievable solutions are within reach, especially through the lens of microbiological research. Members of the microbiology community have the ability, and the responsibility, to do their part as scientists and as citizens to take on, and ultimately overcome, the challenge of plastic pollution.

# Symbiosis on the Reef: Healthy Relationships Under Stress

BY ASHLEY M. ROBBINS, M.E.L.P.

A healthy coral reef at Palmyra National Wildlife Refuge. [Source](#)

It's no secret that healthy relationships can disintegrate in stressful environments. Like any breakup, the separation between corals and their symbionts can be caused by a multitude of factors, with some relationships proving stronger or more tolerant than others.

Coral reefs are among the world's most beautiful and biodiverse ecosystems, forged by a series of unlikely partnerships between organisms large and small. Though typically found in nutrient-poor environments, coral reefs are [immensely productive and biodiverse](#), providing habitat for an estimated [25% of marine life](#). Additionally, reefs serve as nurseries for fish species of commercial value and as hot spots for ecotourism. Yet these ecosystems also face immense pressure on multiple fronts—from [climate change](#) and [ocean acidification](#) to overfishing and damage by [certain sunscreens](#). In stressful environments, relationships cannot survive, much less thrive. [News stories](#) over the last few decades have reported significant losses in coral reefs and described sobering bleaching events associated with corals ejecting their symbionts. This dysbiosis occurs first on a microbial level and, though not initially evident, could result in a deserted, lifeless reef, if not addressed.

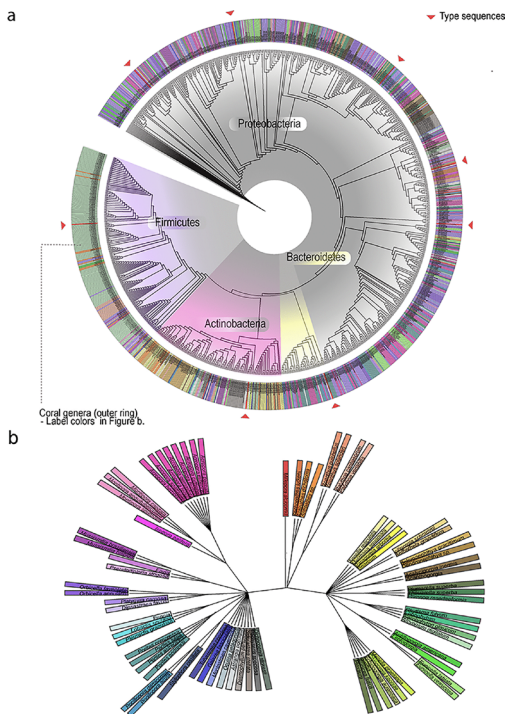


Figure 2 Phylogenetic trees of bacterial strains and coral species. The colors on the outer ring refer to the coral genus from which the bacteria were isolated, and the background colors in the center refer to the bacteria phyla. [Source](#)

## CORALS AND THEIR SYMBIONTS

Perhaps the [best-known relationship](#) on the reef is the one between the tropical stony coral species and *Symbiodiniaceae* zooxanthellae, photosynthetic algae that provide a source of food for the sessile coral animal. However, corals form relationships with a wide variety of bacteria, algae, fungi, archaea and even viruses, which can be found in the coral's surface mucus layer, in its tissue and within its calcium carbonate skeleton. For obligate symbionts, like some stony corals (class *Hexacorallae*) and zooxanthellae, the relationship is essential to survival. In other cases, microbial symbionts provide nonessential, but highly valuable, benefits to the host, like antimicrobial properties or nutrient exchanges. A diverse microbiome can therefore increase a coral's resilience and provide abundant alternative nutritional sources if symbioses with zooxanthellae break down.

"We know that [bacteria] aid the coral with nutrition and growth, mitigation of toxic compounds or stress, early life development and even pathogen control," said Dr. Michael Sweet, a microbial ecology professor at the University of Derby's Environmental Sustainability Research Centre, whose [recent paper](#) was the first study to synthesize decentralized data of cultured microbes associated with corals. "Some bacteria increase the bioavailability of iron to the algal symbionts or supply inorganic carbon for the host's all-important calcification reaction—production of the skeletons which produce the structures that make reefs so important for life in our oceans," he said.

The strength of the relationship depends on the health of both coral and symbiont. High taxonomic diversity in both partners allows multiple coral-symbiont combinations with unique costs and benefits that influence the overall fitness of the holobiont. Environmental stressors, [particularly temperature fluctuations](#), can quickly disintegrate symbiotic relationships, stressing either the coral or the symbiont to the point that it dies or physically separates from its partner. Some symbionts even have the potential to turn pathogenic if their host becomes vulnerable—[endoliths like \*Ostreobium\*](#), a genus of green algae, may begin to break down the internal structure of the coral, eating it from the inside out. Loss of a symbiont can mean loss of a food source, and a weakened immune system that leaves the coral susceptible to disease and death.

## A DEEPER DIVE

Earth contains over 6,000 species of coral, and no 2 relationships are alike, although "closely related species and genera exhibit similar microbiomes," said U.S. Geological Survey research microbiologist Dr. Christina Kellogg, who leads the coral microbial ecology laboratory and has studied both tropical corals and deep-sea corals hundreds of meters below the surface. The work is challenging and cost-prohibitive. Collecting samples from deep-sea corals, which make up the majority of coral species, typically requires submersibles or robotic devices. Kellogg brings the samples to the surface in individual, thermally-insulated containers to minimize contamination and reduce thermal stress responses in the transition.

"All deep-water corals and some mesophotic (middle-light zone) corals lack photosynthetic algal symbionts, so in that sense, they have different microbiomes from tropical reefs," said Kellogg. "As far as bacteria, there are, in fact, some that are the same across corals, no matter where they are or how they live." For example, the chemoorganotrophic marine bacterial group [Endozoicomonas](#) commonly

associated with tropical coral species and shallow-water [Mediterranean gorgonians](#), but recently Kellogg detected them in deep-water corals. *Endozoicomonas* exhibits a high amount of genetic and functional diversity and tends to dominate coral microbiomes, suggesting there might be a correlation between symbionts and their coral hosts, similar to the various species of zooxanthellae observed in stony versus soft corals.

Each symbiont likely confers unique benefits on its host species. Kellogg speculates that the non-algal symbionts of deep-sea corals help make carbon and nitrogen cycling as efficient as possible in an environment where nutrient availability is based on unpredictable currents from the surface. "[The corals] don't know when their next meal is coming, and they don't know what the quality of it is. How much more important must their microbial companions be?" Kellogg wondered.



Several species of deep-sea corals form a colorful garden 165 meters (540 feet) below the ocean's surface off the coast of Alaska's Aleutian Islands. [Source](#)

## SIGNS OF CORAL STRESS

An obvious sign of stress is coral bleaching—when a coral turns white upon expelling its zooxanthellae, which give the otherwise-clear coral its color. Disease, warming temperatures and cold spells have all been associated with bleaching. Although zooxanthellae can survive in the open ocean, bleaching indicates a very likely death for its coral host, as it enters a starvation period. Establishing what a healthy coral holobiont looks like may help scientists diagnose stress or illness in the early stages, allowing them to intervene before it's too late. However, there are challenges to doing so; [each coral species is unique in its baseline measurements](#), and little data exists thus far. Moreover, as more coral reefs die, there are fewer "healthy reefs" by which to compare findings.

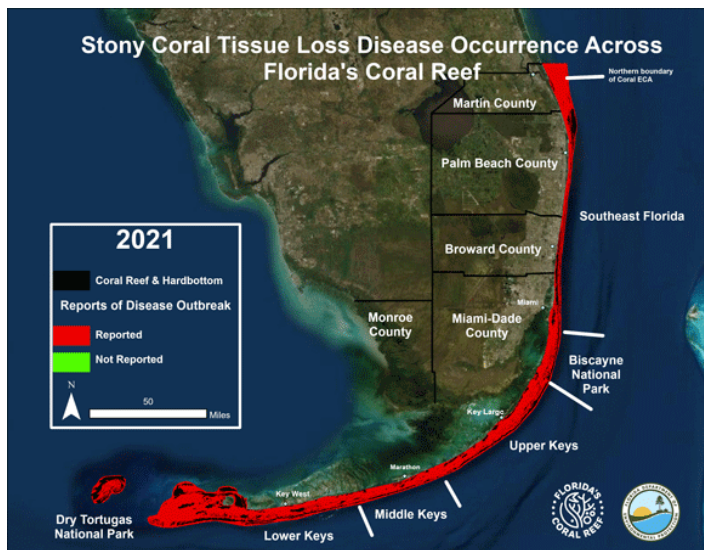
Scientists like biogeochemist Dr. Colleen Hansel are working to identify chemical signatures that serve as early warning indicators of coral stress. Hansel, a senior scientist at Woods Hole Oceanographic Institution, monitors chemicals involved in the physiology and immune systems of corals and the chemical signatures that occur when coral physiology is impaired or stressed.

"For the most part, corals and symbionts have a mutualistic interaction," Hansel said. "There is evidence that a trigger (can disrupt) that happy situation. One of the members may become pathogenic or initiate antagonistic behavior, such as sequestering nutrients that the coral needs and breaking down that relationship that was critical for keeping the happy ecosystem together. A lot of those interactions are based on chemical exchange."

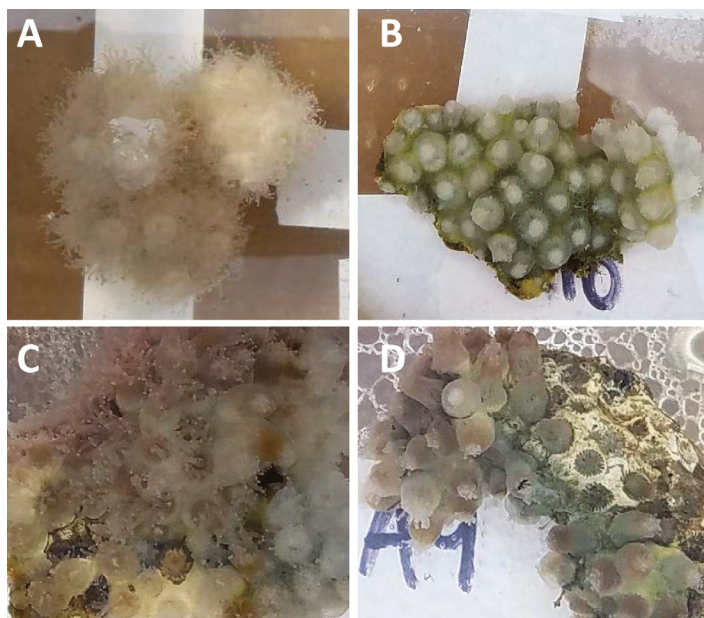
One chemical signature that Hansel's lab focuses on is the [reactive oxygen species \(ROS\) superoxide](#), a fundamental chemical produced by both corals and their symbionts on a regular basis. Under normal conditions, superoxide plays an important role in cell signaling, reproduction and tissue repair. However, an overabundance of intracellular superoxide, produced in response to sudden stress, leads to the breakdown of DNA and other biomolecules, and ultimately results in death. It has been historically assumed that a disruption in ROS homeostasis by triggering algae to overproduce superoxide causes corals to eject their symbionts. "Since superoxide does not pass biological membranes, the link between superoxide and bleaching is unresolved," said Hansel.

Superoxide has a half-life outside of the cell of only a few seconds, posing a challenge for making in-situ measurements. "By the time water samples are collected and brought to the surface," Hansel said, "the superoxide is long gone." She and her team at Woods Hole, with funding from Schmidt Marine Technology Partners, developed a unique submersible sensor, [DISCO, to allow for superoxide monitoring underwater at the source](#). Using this sensor, the team recently showed that healthy corals make superoxide outside of their cells, which they predict is providing essential physiological roles for the coral and symbionts. A deep-sea version of the technology, [SOLARIS](#), accomplished the first superoxide measurements associated with deep-sea corals and sponges (a paper on this is forthcoming).

"Each coral has a basal level of ROS always present that is part of their normal physiological function," said Hansel. "We need to quantify these baseline values so that when we have measurements over the healthy level, we will know the organism is stressed internally, it is fighting off a pathogen or it's struggling with tissue degradation." As ROS are part of the immune system response, elevated ROS production can precede more obvious visual symptoms of stress or disease. "[Identifying these signatures and developing low-cost deployable sensors](#) will allow us to implement a global early warning system for coral reef health," Hansel said. "Let's do some bloodwork. Let's take its pulse."



Map of stony coral tissue loss disease. [Source](#)



*Astrangia poculata* colonies during the course of treatment. [Source](#)

## DEVELOPING SOLUTIONS TO CORAL DIE-OFFS

Scientists are still far from understanding the causes of most coral diseases, like the rapidly moving, highly virulent [stony coral tissue loss disease](#) (SCTLD) currently decimating Caribbean corals. According to University of North Carolina Wilmington professor Dr. Blake Ushijima, the cause of SCTLD is likely complex, but probiotic treatments composed of microbial cultures from resistant corals may be the solution.

Ushijima found that corals less susceptible to SCTLD are colonized by beneficial bacteria, some of which produce broad-spectrum antibacterial compounds. "At least some of these very specific isolates, if put onto the coral, will actually protect them from disease and sometimes treat the disease directly," he said. "Right now, we are at the stage where [probiotic applications] seem to work in lab and aquarium trials, and we need to see if they can work in the field." His lab is also currently studying opportunistic infections by the bacterium [Vibrio coralliilyticus](#), a pathogen [implicated in a large number of diseases of corals](#) and shellfish. "Though *V. coralliilyticus* is likely not the cause of SCTLD, it may play a role in coinfections that exacerbate existing SCTLD lesions," he explained.

Kellogg said that the Caribbean/U.S. coral community has responded en masse to the topical pastes impregnated with chlorine or antibiotics and culling diseased colonies. Another effort has involved rescuing unaffected corals from the reefs, then housing them in zoos or aquaria for safekeeping until the disease passes and the reefs can be restored.

Lab settings allow for controlled experiments, although there is no way to replicate the complete holobiont and its dynamics as one could in an ocean environment. A lab does, however, provide a safe area in which to test proposed probiotic treatments before deploying them in the field as tools for reef restoration

or conservation. Sweet is now exploring ways to enrich the media with host tissue or mucus, which may unlock more of these symbiotic microbes in culture collections.

Is recovery possible? [Research has shown](#) that the *Astrangia poculata* microbiome can recover from antibiotic disturbance, and that individuals with algal symbionts reestablish their microbiomes in a more consistent manner compared to corals lacking symbionts. New studies are examining whether symbionts from more heat-resistant corals could transfer heat resistance to more vulnerable individuals, inviting the concept of "[microbiome transplantation](#)" treatments using inoculations of homogenized coral tissues. However, these studies are still in the beginning stages.

"There is a cost to inaction," Sweet said. "Reefs are dying all over the world, and our 'tool' can and does help (a little). I always describe our probiotics as a sticky plaster—they can stem the bleeding to some degree (reefs or individual coral colonies dying from bleaching or disease), but it won't really help unless we deal with the cause of the bleed (climate change). We have shown it works, now it is time to deploy and save a few corals—[those] which may be key to repopulating reefs in the not-too-distant future."

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To protect coral communities, support for microbiome research, data sharing and a collaborative approach that includes residents who depend on the ecosystem are critical. Learn about community approaches to coral conservation and what is needed to: [Save the Coral](#)



# Shark Epidermis Microbiome with Elizabeth Dinsdale

BY ASHLEY HAGEN, M.S.

**D**r. Elizabeth Dinsdale, Matthew Flinders Fellow in Marine Biology in the College of Science and Engineering at Flinders University in Adelaide, Australia, uses genomic techniques to investigate the biodiversity of microbial communities in distinct ecological niches, including coral reefs, kelp forest and shark epidermis. She discusses how shotgun metagenomics is being used to characterize the architecture of microbial communities living in the thin layer of underlying mucus on shark's skin, and how understanding the function of these microbes is providing clues to important host-microbe interactions, including heavy metal tolerance.

## ASHLEY'S BIGGEST TAKEAWAYS

Sharks belong to a subclass of cartilaginous fish called elasmobranchs and are unique in that their epidermises are covered in dermal denticles—overlapping tooth-like structures that reduce drag and turbulence, helping the shark to move quickly and quietly through the water. These dermal denticles are sharp (if you're going to pet a shark, make sure you go from the head to the tail to avoid getting cut!), and depending on the species of shark, may be more or less spread out across the epidermis.

Where do microbes enter the story? Dermal denticles overlay a thin layer of mucus, which provides a distinctive environment for microbial life. Collecting microbial samples from underneath a shark's dermal denticles is quite difficult, and the technique varies by shark species (shark size, water depth and ability to bite all factor into the equation). Liz's team uses a specially designed tool that the group affectionately calls a "supersucker," to create and capture a slurry of microbes and water for analysis.

The team then uses shotgun metagenomics to identify and characterize the microbes in their collected samples. Sequencing has revealed biogeographical difference, as well as similarities in microbial architecture of whale sharks across the globe.

There are 2 populations of whale sharks—one in the Atlantic Ocean and the other in the Indian Pacific Ocean. Samples collected from both populations have revealed that each individual whale shark, from within each aggregation, shares many of the same microbes. In fact, unlike algae which may share 1 to 2 microbial species, whale sharks share about 80% of microbes across every individual. Since many of the sharks don't cross aggregations, Liz's team is investigating the possibility of coevolution between microbes and hosts.

Metagenomic sequencing also provides information about the function of the sequenced microbes. High presence of heavy metal-tolerant microbes has been found in the epidermis of all shark species that the team has analyzed. Sharks are known to carry high levels of heavy metals in their skin, muscle and even blood. However, muscle tissue samples contain lower concentrations than skin, indicating that there may be a density gradient in place, and raising questions about how microbes might be involved in this regulation. Is there a pathway by which the microbes metabolize and help to remove concentrations of heavy metals across the epidermis? Liz and her team are hoping to find out.

## FEATURED QUOTES:

"I chose sharks because they're a very old lineage of vertebrate organisms. In fact, the oldest ones are still around today. They're quite different from Teleost fish, which while they were evolved early on, had a radiation in more recent times."

"I'm wondering if the sharks actually have a longer-term relationship with their microbes than the Teleost fish and other vertebrates that we see today. And that might tell us something about the evolution of microbes and viral interactions with their hosts."

"We've been able to collect samples from around 76 whale sharks from across the world in 5 different locations. They're the most spectacular animal you've ever seen because they're about 6-8 meters long. And we've gone out with collaborators in Cancún, Mexico; La Paz, Mexico; Philippines; Tanzania and Ningaloo reef off Northern Australia, collecting the microbes on the skin of all of these sharks in all of these locations, which is really quite a big sample size."

"We're wondering whether the microbes on the epidermis change across the globe—whether they show biogeography—which they do. However, when we do a more robust analysis like a network analysis, we see that they have the same architecture, so the microbial community is structured in the same way, which is also quite interesting. So we're wondering if that means that there's a certain level of community or community structure that occurs on, or between, the dermal denticles."

"Because we do shotgun metagenomics, we can actually identify what functions the microbes have. And the one really interesting standout that appears to occur is a high level of heavy metal associated genes. If you can compare the level of heavy metal associated genes on the sharks to the water column, it's about 4x as much."

"The other quite cool thing is when you do create these metagenomes, you do identify novel species. Many of our constructed genomes we can identify that they're a gamma proteobacteria, full stop. They're not matching any of the phylogeny any further, so that means we have some very novel microbes out there."

"We've started looking at the viruses associated with the skin of the sharks, and most of them are phage as opposed to eukaryotic viruses, which is once again different from what people have seen on the skin of Teleost fish. Sharks are known as long-lived and health animals, so whether that is one of the mechanisms by which they're not getting those high levels of viral infections is another outstanding question."

## HISTORY OF MICROBIOLOGY

Next generation sequencing (NGS), high-throughput sequencing methods that can process millions of individual DNA or cDNA fragments at the same time, became available and began to gain traction in the mid-2000s. In 2005, the 454 sequencing system became the first next generation sequencing platform to come to market, and Illumina acquired Solexa in 2007.

Metagenomic sequencing (mNGS) is a type of next generation sequencing that provides a hypothesis-free approach to detecting all nucleic acid in a given sample. Nothing specific is targeted, resulting in both host and microbial nucleic acid being sequenced. This gives researchers the ability to look at any portion of the genome sequenced, determine whether a microbe of interest is present in the sample, identify new or unexpected organisms and learn important information about the host.

The coral holobiont, is a dynamic group of algae, fungi, bacteria, archaea and viruses that form stable and species-specific associations with corals. In 2005, the group went to the Northern Line Islands, a group of Equatorial islands straddling the North and South Pacific Ocean that are all incorporated territories of the United States, and used metagenomics to investigate the microbes that were associated with a more pristine coral reef compared to a more degraded one. In addition to detection of 10 times more microbial cells in the water column of reefs on inhabited versus uninhabited islands, the group observed that the benthic communities on the most populated island had the highest prevalence of coral diseases. As Liz noted, shark numbers decreased in stressed coral populations where microbial communities increased in number and pathogenicity.

Prior to the research that Liz, Forest, Linda and Rob conducted between 2005-2009, disruptions in the holobiont were known to be associated with coral disease, but little was known about the specifics of how and why certain microbial species disassociate from their coral hosts in different ecological niches, a process called adaptive bleaching, and/or why they suddenly switch from mutualistic to pathogenic behavior. In order to learn more about these host-microbe interactions, the scientists exposed a species of marine stony coral, called *Porites compressa*, to a variety of known stressors, including increased temperature, elevated nutrients, dissolved organic carbon and lowered pH. They then isolated microbial communities and performed metagenomic sequencing (using a 454 Life Sciences system) and found that all types of stress increased microbial pathogenesis and the abundance of pathogen-associated genes, such as those involved in motility and virulence. Understanding how microbes are involved in the maintenance and degradation of coral reef ecosystems is a first step in preventing decline of one the most productive and biodiverse ecosystems on our planet.



# Waterborne Diseases in a Changing Climate

BY RODNEY ROHDE, PH.D.

*Naegleria fowleri* is commonly referred to as the "brain-eating amoeba." [Source](#)

**P**lentiful and essential, water is considered the universal solvent for biological life. Indeed, life is thought to have originated in the aqueous environment of the planet, and living organisms rely on blood, extra- and intracellular solutions and digestive juices for biological processes. Yet, [water can also be a place where dangerous and deadly microbes](#) reside, waiting for a host to arrive.

Waterborne illnesses are likely to occur more frequently with the ongoing changes to our weather and climate patterns. The health effects of climate change [could undercut public health advances and developments made in the last 50 years](#). Rain events, storm surges and the rise and fall of temperatures and humidity levels all [impact the survival of microbes](#) on land and in our seas. Some have evolved strategies to survive at temperatures higher than ambient body or host temperatures. Furthermore, changes in precipitation patterns can mobilize or concentrate pathogens and compromise water and sanitation infrastructure. Flooding and runoff can lead to the dissemination of raw sewage, harmful chemicals and pathogens, and drought can concentrate pathogens in areas of limited water supply. These factors favor the growth, reproduction, spread and survival of the major microbial taxa (parasites, helminths, bacteria, viruses and toxins created by harmful algae). Ultimately, global populations will likely meet these agents, leading to a rise in waterborne illnesses through ingestion, inhalation and skin contact, as well as via the consumption of contaminated fish and shellfish.

## EXAMPLES OF WATERBORNE PATHOGENS

In the [December 2021 issue](#) of *Emerging Infectious Diseases*, the Centers for Disease Control and Prevention (CDC) reported that a 56-year-old Maryland woman hospitalized on Sept. 20, 2019 [had likely acquired melioidosis](#), a disease characterized by a range of symptoms, including localized, pulmonary, bloodstream and disseminated infection, via novel transmission of *Burkholderia pseudomallei* from a freshwater home aquarium. The woman is the first person known to have acquired the severe tropical infection by this transmission route.



A freshwater aquarium with plants, congo tetra, harlequin rasbora, siamese algae eater, neon tetra and Bronze corydoras. [Source](#)

One of the reasons that aquatic infections are not immediately considered in differential diagnoses is that many aquatic zoonoses, including *B. pseudomallei*, are not endemic in the U.S. [Melioidosis](#), the disease caused by *B. pseudomallei* and formerly known as Whitmore's disease, was first described in 1912. Cases were historically identified primarily in northern Australia and areas of Southeast Asia, such as Thailand, and almost all cases of infection reported in the U.S. have been related to international travel to endemic regions.

In the U.S., [11.5 million households have pet fish](#), and most exotic aquarium animals are caught in the wild and transported to a major hub for transport to the U.S., where a [quarantine may not be in place](#). This could be why [outbreaks have recently been reported in the U.S.](#)



*Burkholderia pseudomallei* on sheep's blood agar. [Source](#)

*Mycobacterium marinum* is another bacterium that is commonly found in both salt and fresh water. It is the causative agent of disease in many species of fish, and the disease occasionally reaches humans. The infection is commonly known as [fish tank granuloma](#), and it produces nodular or ulcerating skin lesions on the extremities of healthy hosts. Fishhook injury is a common route of infection. Diagnosis is usually delayed because individuals do not consider this a likely microbial route of infection, and invasion into deeper structures, such as synovia, bursae and bone, occur in approximately 30% of reported cases. *M. marinum* is not a nationally notifiable condition in the U.S.

Fishing and/or cleaning and preparing fish or shellfish may provide additional exposure risk to water-loving bacteria. For example, *Streptococcus iniae* has caused cellulitis, arthritis, endocarditis and meningitis following superficial or puncture injuries, notably from cleaning tilapia. Other infections from contact with fish include, primarily topical skin infections from *Erysipelothrix rhusiopathiae*, and gastroenteritis from *Plesiomonas shigelloides*, *Campylobacter* spp. and *Salmonella* spp.

## HOW DO ENVIRONMENTAL CONDITIONS INFLUENCE PATHOGENS?

How does water become contaminated with pathogens in the first place? Most people are aware that water is usually [treated to ensure safe drinking quality](#), so how do outbreaks of waterborne illness occur?

Much of the existing research, including a [recent extensive literature review](#) from a team of collaborators at Emory University and University of Colorado, examines historical relationships between observed weather and disease incidence, with a more limited number of studies [projecting future disease rates](#). Positive associations between ambient temperature and diarrheal diseases (excluding viral diarrhea), as well as an increase in diarrheal disease following heavy rainfall and flooding events, have been recognized. The effects of drought on diarrhea have been less supported, due to a lack of evidence.

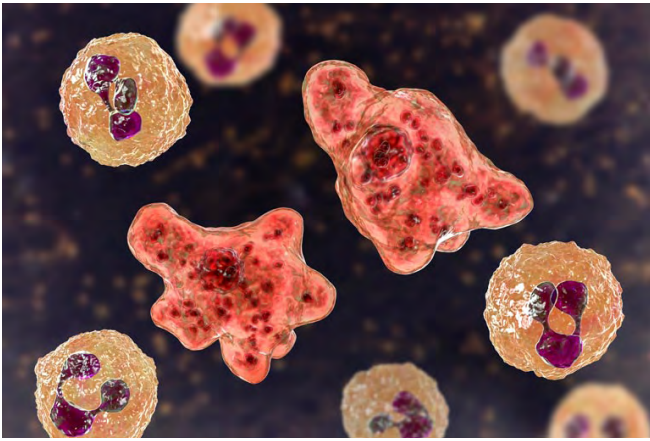
Heavy rainfall typically increases in intensity, duration and frequency due to climate changes. With increased intensity and/or amount of rainfall come increased runoff and flooding, especially in river and coastal environments. As one might expect, massive rainfalls can wash substances at the surface level, like chemicals, animal waste containing pathogens, gasoline and other dangerous items, into sources of drinking water. Changes in the Great Lakes' rainfall rates, higher lake temperatures and low lake levels have all been linked to increases in fecal bacteria levels in those waters. During summers following heavy rainfalls, freshwater streams in the southeastern U.S. show increased levels of the bacterial pathogens *Salmonella* and *Campylobacter*.

Additionally, *Cryptosporidium parvum*, a parasitic protozoan that causes diarrhea, cramping, abdominal pain and fever, is responsible for the deadliest waterborne outbreak in U.S. history in Milwaukee. *Cryptosporidium* infects the intestines of people and animals and is resistant to chlorination. [Due to this resistance](#), one often must filter the water and utilize ultraviolet radiation or ozone to make the drinking water safe. [Possible sources of this outbreak](#) included cattle, slaughterhouses and human sewage along 2 rivers that flow into the Milwaukee harbor. Rivers that were swelled by spring rains and snow runoff may have transported oocysts into Lake Michigan, and from there to the intake of the southern water plant. The outbreak caused 403,000 illnesses and over 50 deaths.

In 2000, [heavy rainfall in Walkerton, Ontario, Canada](#), carried agricultural runoff containing *E. coli* into the town's primary water source: a shallow well. This extreme weather-related event caused 2,300 illnesses and 7 deaths. Since groundwater wells receive limited water treatment because [the rock and sediment layers act as natural filters](#), they are more at risk for contamination with heavy precipitation events, increasing the possibility of waterborne illnesses for anyone who drinks water from those common wells.

[Data indicate](#) that the intersection of flooding and higher temperatures will not only transport pathogens into recreational waters, but may also help facilitate a microbial population explosion. Numerous examples of temperature-induced microbial growth have been reported for *Naegleria fowleri*, commonly referred to as the "brain-eating amoeba," *Salmonella*, *Campylobacter* and *Legionella*, which causes Legionnaires' Disease, a respiratory illness consisting of cough, shortness of breath, high fever, muscle aches and headaches.

Massive sewage overflow from events such as heavy rainfalls can also contaminate seafood. This type of event occurs repeatedly in



*Naegleria fowleri* is commonly referred to as the "brain-eating amoeba." [Source](#)

California, where an outdated sewage treatment plant is unable to accommodate heavy rainfall and subsequently [causes sewage to flow from the Tijuana River into the adjacent Pacific Ocean](#), spreading pathogens across regions of beaches along the California-Mexico border. Fish and shellfish can accumulate viruses found in sewage and put consumers at risk for gastrointestinal viruses, such as norovirus and hepatitis A. Each of these can result in stomach pain, nausea, diarrhea and vomiting. Hepatitis A can also cause acute liver failure, which is associated with high mortality.

Increases in the frequency and range of harmful algal blooms is another concern associated with rising sea surface temperatures and altered rainfall distribution. Certain algal blooms produce potent toxins that can contaminate seafood and lead to dangerous health issues. [Paralytic shellfish poisoning](#) is the most common and severe form of shellfish poisoning and is caused by ingesting shellfish contaminated with

saxitoxins that are produced by phytoplankton. Symptoms include numbness, tingling sensations in the body, headache, nausea, vomiting and diarrhea. A large dose of the toxin may lead to the inability to control bodily movements, difficulty swallowing, change in mental status, flaccid paralysis (feeling weak or paralyzed) and respiratory failure. [Ciguatera fish poisoning](#), or ciguatera, is an illness caused by eating fish that contain toxins produced by a marine microalga called *Gambierdiscus toxicus*. People who have ciguatera may experience nausea, vomiting and neurologic symptoms such as tingling fingers or toes.

## PREVENTION AND TREATMENT OF WATERBORNE INFECTIONS

Keeping some [basic safety measures in mind while](#) interacting with water outdoors or indoors can help limit the exposure and spread of waterborne infections. For example, when swimming or playing in [recreational facilities](#), it is important to keep water out of the mouth and ears. Likewise, individuals who have been sick with diarrhea in the past 2 weeks should stay out of the water. [Disinfection with chlorine or bromine](#) and properly regulated pH is the first defense against the germs that cause recreational water illnesses in pools, hot tubs/spas and water playgrounds. If one suspects something is not right with the body of water they are about to enter, such as it smells bad or is cloudy, they should stop and report it to someone who can inspect the facility.

People should not participate in outdoor water sports in lakes, rivers and streams without proper eye and nasal protection to protect against the rare, but deadly, parasite *N. fowleri*. Also, the importance of careful hand hygiene when caring for aquariums cannot be stressed enough. Aquarium filters, filter floss, biofilm, charcoal and gravel might have exceptionally high concentrations of bacteria. Gloves are critical when cleaning aquariums, and immunocompromised individuals should avoid this task altogether. Finally, one should never drink water from an unknown source or if there is uncertainty about whether the water has been [properly treated](#).



Priest Lake, Idaho is a popular spot for water sport activities during the summer. [Source](#)

Individuals who acquire waterborne illnesses usually have primarily gastrointestinal symptoms, but upper respiratory symptoms and skin manifestations may also appear. Gastrointestinal symptoms are usually self-limited, and supportive treatment may be all that is necessary. However, some infections can cause significant morbidity and mortality.

In some infections, [specific antimicrobial agents may be required to limit the pathology](#). For example, *Vibrio vulnificus* can cause a dangerous infection known as cellulitis, which can lead to amputation or death. With Legionnaires' disease, if it is unrecognized and progresses, the mortality rate can be as high as 10%. Lastly, there could occasionally be medical and public health reasons to utilize [prophylaxis](#) to prevent the spread of disease in outbreak situations (e.g., long-term care facilities, daycare centers and among immunocompromised populations).

[In 2012, the WHO estimated that 12.6 million deaths \(23% of global mortality\) were attributable to modifiable environmental factors](#), many of which could be influenced by climate change or are related to the driving forces of climate change. While this article does not provide a comprehensive look at climate change, one can conclude that environmental conditions do influence pathogens via [direct impacts of climate change](#) that result in higher temperatures (e.g., heatwaves) and increases in the frequency of complex, extreme weather events (e.g., windstorms, floods, droughts).

# Is Your Water Safe to Drink? Ask the Microbes

BY LEAH POTTER

Source

While invisible to the naked eye, seemingly clear water can be full of microorganisms. Some microbes, like *Escherichia coli* (*E. coli*), can cause adverse health effects. But others, like *Dokdonella kunshanensis*, could actually be [used to purify wastewater](#). Thus, the presence of microbes in water poses the question: are they friends or foes?

## A CALL FOR TESTING

Every year, there are millions of reported cases of waterborne diseases, including typhoid fever and cholera. There are [11 million cases of typhoid fever globally, resulting in 129,000 deaths. Additionally, there are 3 million cases of cholera globally and 95,000 related deaths.](#) According to the [Lancet 2015 Global Burden of Disease study](#), more than 6.5 million cases of acute diarrhea occur globally each day. These cases are primarily the result of waterborne diseases that inconspicuously lurk beneath the water's surface, like rotavirus (caused by rotavirus gastroenteritis), cholera (caused by *Vibrio cholerae*) and dysentery (caused by *Shigella*), and are often contracted through drinking water. According to the [World Health Organization](#) (WHO), more than 2 billion people do not have access to safe drinking water.

"You don't see the microorganisms that cause the disease when they could be present in the water," said Dr. Kehinde Eniola, dean of the College of Postgraduate Studies at Joseph Ayo Babalola University in Osun State, Nigeria. This is why testing for pathogens in drinking water is incredibly important. Even the clearest water could have contaminants from fecal matter. Testing drinking water can identify both the presence of harmful microbes and the level of risk associated with the water source, which could help a community mitigate the spread of waterborne diseases. "It's like if someone says, 'I don't have COVID-19,' you would say, 'I'm going to test you first,'" Eniola continued. "So only a negative PCR test will tell you if you are free of COVID-19."



## EVIDENCE-BASED MICROBIOLOGY AT THE COMMUNITY LEVEL

Water-testing kits can serve as educational tools that demonstrate best practices for identifying safe drinking water. In Lower Nyakach, Kisumu County, Kenya, its population of 70,000 has little to no access to safe drinking water. "They've got a shallow well; they are getting water from their river," said Dr. Robert Metcalf, a professor emeritus of biological sciences at California State University, Sacramento and co-founder and president of the International Water and Health Alliances. "And the most important public health piece of information you've got to have is whether the water that you're drinking is safe to drink or not."

Metcalf has worked with the [Friends of the Old](#) (FOTO) leadership team since before the project's inception. FOTO [aims to eliminate](#)

[cases of waterborne diseases](#) completely in Lower Nyakach. As a result of community-based education, community members in Lower Nyakach are more aware of water quality and disease-detection practices. "Instead of trying to get 5 more vaccines for pathogens found in drinking water that are going to be hard to distribute and pay for, let's empower community-based organizations, like FOTO," Metcalf said. He explained that between the 2 types of prevention strategies—removing the threat (e.g., water sanitation) and conditioning one's immune system to recognize pathogens upon exposure (e.g., vaccines)—the former is more cost-effective and practical at the community level. "Let's ensure [community-based organizations] have enough funds to get chlorine, distribute it to people and provide education about how to use it," he said.

According to Dinah Chienjo, the project director of FOTO, 95% of community members in Lower Nyakach are treating their drinking water by boiling the water or sterilizing it with chlorine. She explained that this is a result of water-testing and purification education that uses the local language, which community members easily understand, and is delivered by local women who they know well and can easily identify with. "There are many communities that drink water that isn't safe," Chienjo said. "They need to know that they can do something about it to make it safe—so they can change their health, so they can be healthier, and their children can be healthier."

Chienjo added that FOTO also educates children in schools, demonstrating how to test and treat their water, practices that can be easily replicated in many parts of the world to eliminate waterborne diseases. While neighboring districts saw a high number of cases of cholera between February and May 2015, Lower Nyakach did not see any cases of cholera and recorded a decrease in cases of diarrhea. Between 2012 and 2016, 4,000 families in Lower Nyakach were provided with [solar Cookits](#), which use solar energy to heat and pasteurize water. Since 2012, the community also has been provided with chlorine for water treatment every month, including 62 primary and 17 secondary schools in the project area. "Numerous programs that focus on water purification require large, expensive pieces of equipment

that a community might not have access to, even in a large city," explained Dr. Musau WaKabongo, the founder and CEO of Dr. Musau WaKabongo Science Education, Inc. and co-founder of the African Initiative Group. "There are many programs and projects focused on water purification. But are they practical at the village level?" For WaKabongo, education is key. Equipment cannot simply be dropped off at a location without explanation or demonstrated results. "To teach people water treatment," she said, "you really need to go to them. You need to talk to them in their language."

WaKabongo said building relationships and credibility in a community is integral to implementing and sustaining water-testing and purification practices. "The only way to win the community's trust is whatever you are doing must be done in their presence, they must be witnesses," she said. "The moment you take anything away from their presence, they doubt you. And that's where portable microbiology laboratories (PMLs) seem to give an edge, because now you do it yourself."

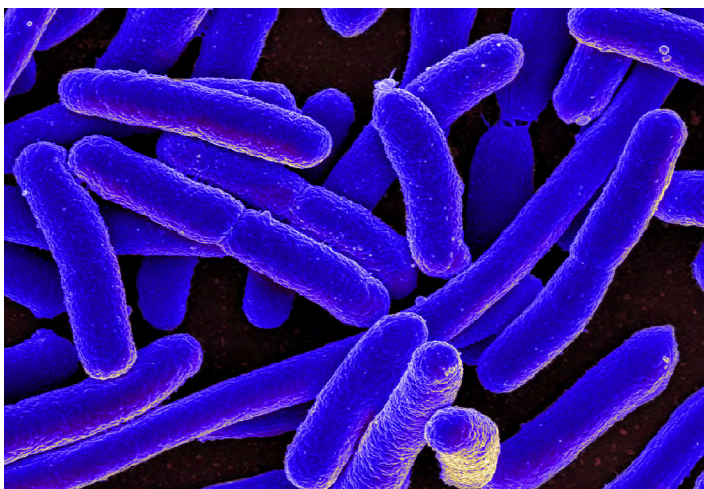
Last year, in the Democratic Republic of Congo, WaKabongo helped train adults and children on how to test water safety using PMLs in the village of Mpoyi and Tshibombo, a city of displaced people in the Kasai-Oriental Province. The hope is that providing testing materials will enable the community to have safe water and lower the risk of disease.

### "YOU CAN BE A WATER MICROBIOLOGIST"

Some standard water-testing practices are not affordable or accessible in under-resourced communities. However, PMLs, which require less equipment and cost around \$1 per test, can check for the presence of *E. coli*, using just 10 milliliter of water. Many waterborne diseases, like cholera and typhoid, are often [associated with fecal contamination](#). When German physician and microbiologist [Robert Koch's methods in the 1880s led to the discovery that bacteria from feces cause diseases](#), testing for all



Water samples in 5 Whirl Packs are ready to be tested with Colilert 10 ml and Petrifilm tests in Kisii, Kenya. Source: Robert Metcalf



*E. coli* bacteria. Source

diseases associated with fecal contamination was understood to be impractical, so scientists quested after a microbe that would survive as long as most types of bacterial pathogens: *E. coli*. Additionally, the presence or absence of *E. coli* is typically used as an indicator of water safety because it is found in a large number of fecal samples and bacterial colonies are easy to count, if sampled.

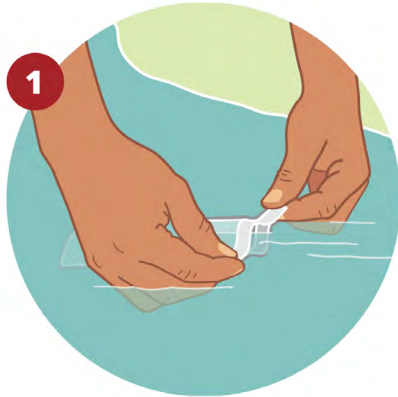
PMLs include a Colilert 10 milliliter presence/absence test, an *E. coli* count Petrifilm, a plastic spreader, a sterile collecting bag, a sterile plastic pipette and a long-wavelength UV light. Water samples are collected and first tested for the presence of *E. coli*. Then, using the Petrifilm—a quantitative test to estimate the number of *E. coli* present per milliliter—bacteria from the sample is grown to help assess the risk level of the water. "Much of what we do in microbiology labs is a little mysterious," said Metcalf. "But with the right tools, you can be a water microbiologist."



The components of a PML include a Colilert 10 milliliter presence/absence test, an *E. coli* count Petrifilm, a plastic spreader, a sterile collecting bag, a sterile plastic pipette and a long-wavelength UV light. Source: Robert Metcalf

[UNICEF recommends testing 100 milliliters \(a more expensive option at about \\$4 per test\) of water and utilizing membrane filtration](#), which can cost upwards of \$2,000, to detect the presence of *E. coli*. But Metcalf says collecting such a large water sample is not always necessary. If there's no presence of *E. coli* in a 10 milliliter sample, he says, the risk of disease is low. "High" risk is defined as 1-10 *E. coli* per milliliter, and "very high" risk is defined as more than 10 *E. coli* per milliliter, according to the [WHO Guidelines for Drinking Water Quality](#).

## HOW TO CONDUCT A COLILERT TEST



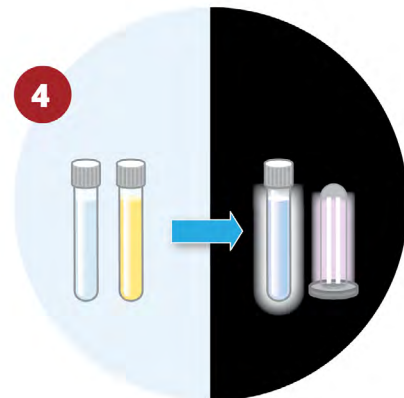
Collect a water sample of about 100 milliliters from the source.



Pipette 10 milliliters of the sample into the presence/absence test.

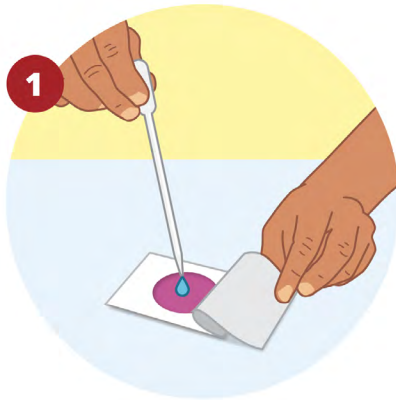


Incubate the sample for 12-18 hours. The average body temperature of about 37 degrees Celsius happens to provide an ideal environment for bacterial cell growth, so sealed test tubes can even be carried in one's pocket to incubate the sample.



If the sample turns yellow, determine whether *E. coli* is present by using the UV light. If the sample produces fluorescence, *E. coli* is present in the sample. Colilert tests rely on 2 food sources to grow *E. coli* and environmental bacteria: *Ortho-Nitrophenyl-β-galactoside* (ONPG) and *4-methylumbelliferyl-β-D-glucuronide* (MUG). The latter is only used by *E. coli*, and when the bacteria metabolizes MUG, it produces fluorescence.

## HOW TO USE A PETRIFILM

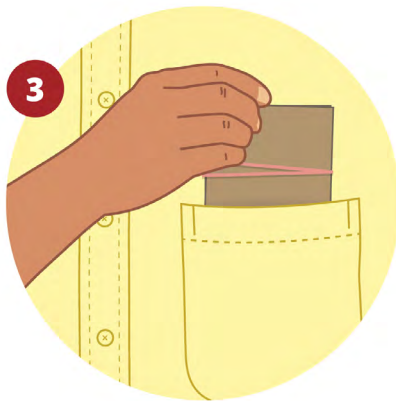


Pipette 1 milliliter of the water sample onto the Petrifilm. Unlike other tests, utilizing a Petrifilm forgoes the need for autoclaves, petri dishes and agar media.

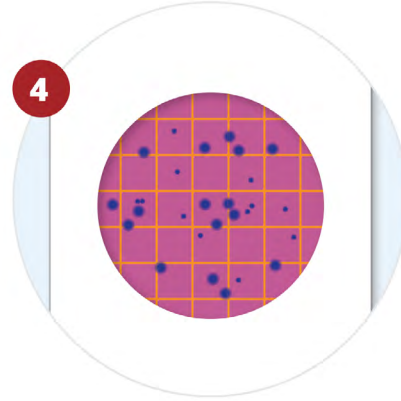
The Petrifilm test uses 2 substrates—lactose and 5-bromo-4-chloro-3-indoxyl- $\beta$ -D-glucuronide (BCIG)—which are metabolized by *E. coli*.



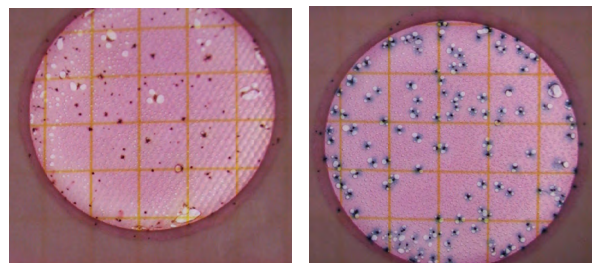
Place the plastic spreader on top of the Petrifilm to distribute the water sample evenly. Let the sample sit for 1 minute to allow the gelling agent to bind to the nutrients on the film.



Incubate the sample for 8-10 hours at body temperature (37 degrees Celsius). This allows for the sample to grow to about 1 million cells, making the bacteria colonies visible on the Petrifilm.



Count the bacteria colonies on the Petrifilm. If no bacteria are visible on the Petrifilm, the risk level is classified as "low" or "moderate." If 1-10 bacteria colonies are present, the risk is "high," and finding more than 10 bacteria colonies is considered "very high" risk, according to the [WHO Guidelines for Drinking Water Quality](#).



Two Petrifilm tests after incubation show *E. coli* growth (right) and coliform bacteria growth (left). Source: Robert Metcalf



## DISINFECT BEFORE YOU DRINK

You've identified bacteria in your water source, which means it's not safe to drink. Now what? Metcalf recommends 2 options: Boil the water, or use chlorine.

If you heat water to 65 degrees Celsius, pathogenic organisms, including *Giardia*, *Entamoeba*, *Cryptosporidium*, *E. coli*, Shigella, cholera, typhoid, rotaviruses, polioviruses and hepatitis A viruses, will be killed. In place of a thermometer, a device the size of your thumb called a Water Pasteurization Indicator (WAPI) can tell you when the water temperature arrives at 65 degrees Celsius. The WAPI consists of a sealed tube filled with wax, which is attached to a fishline with a metal washer at the end that can be dipped into the boiling water. As the water's temperature increases, the wax inside the WAPI will start to move toward the bottom of the interior.

If you opt to use chlorine, about 20 liters of water can be purified using a bottle cap's worth of the chemical element. After adding the chlorine and waiting about 30-45 minutes, the water is safe to drink. However, after 4 days, chlorine weakens as a purifying agent, and the water should be repurposed for something other than drinking.



A Water Pasteurization Indicator (WAPI) indicates when the water temperature arrives at 65 degrees Celsius. Source: Robert Metcalf

# What's Hot in the Microbial Sciences

BY ASHLEY HAGEN, M.S. & STANLEY MALOY, PH.D.

In this issue, "What's Hot" takes a look at ... groundbreaking research and cutting-edge science pertaining to the impact of climate change on plants and pathogens, how shipwrecks contribute to microbial diversity on the ocean floor, new insights in antimicrobial resistance, how tumor resident microbes contribute to metastasis and so much more!

## AS THE PLANET GETS HOTTER... MICROBES CAN INCREASE DROUGHT TOLERANCE IN PLANTS

Drought is a significant source of abiotic stress that impacts plant growth and crop production. The threat of warming soil temperatures and a changing climate have only intensified these concerns, but microbes can help! A number of studies have implicated bacteria in the drought resistance of plants, and a couple of recent papers provide thought-provoking data that should catalyze future experimentation on the subject. Scientists are exploring ways to enhance the natural ability of plant-growth-promoting bacteria (PGPB) to alleviate drought stress in plants through the solubilization of mineral nutrients in the soil; production of important chemicals that help regulate plant growth, called phytohormones; and the production of osmolytes, molecules that maintain cell integrity by regulating the viscosity, melting point and ionic strength of biological fluids.

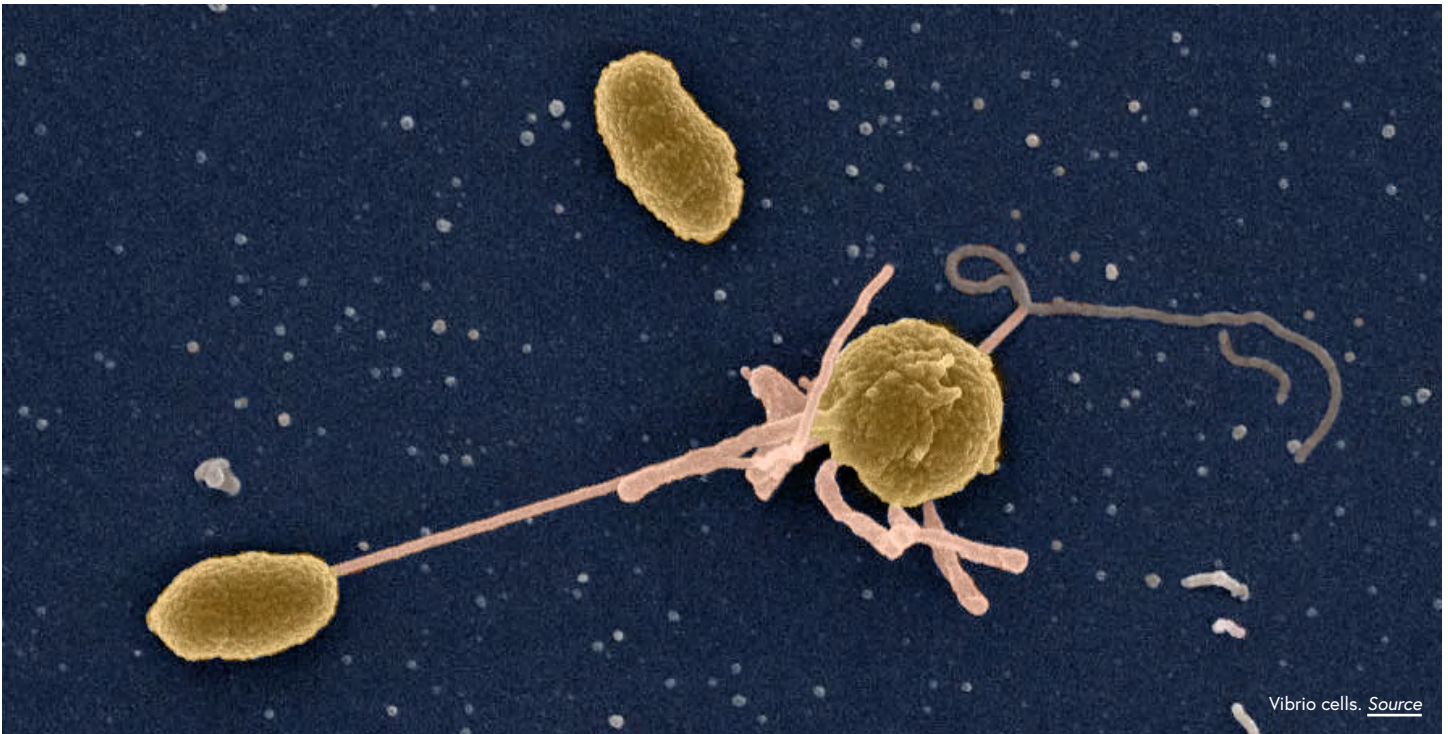
One study, published in the [Chemical Engineering Journal](#), demonstrated that an engineered strain of obligate methanotroph, *Methylovimicrobium*

*alcaliphilum* 20Z, could overproduce the phytohormone indole 3-acetic acid (IAA) from methane. The new strain was able to convert a dangerous greenhouse gas to a usable plant-growth-promoting hormone at high titers, and researchers further proved that microbial treatment of wheat seeds with the IAA-producing strain under saline-alkaline conditions caused markedly increased germination and elongation of the shoots and roots of the plant. The authors proposed a promising approach to simultaneously reducing greenhouse gas emissions and facilitating sustainable crop production, suggesting that the strategy should be expanded as a seed treatment biofertilizer.

Other studies have been looking at strategies to assess drought tolerance in agricultural lands. A [study published in mSystems](#) used stable isotopes and Raman spectroscopy to quantify microbial phenotypes of drought-tolerant bacteria in soil samples and found that (depending on the sample) 0%-52.2% of all measured single cells exhibited drought-tolerant properties. Furthermore, scientists were able to relate the phenotypic properties of the soil microbiome to the behavior of surrounding plants under drought conditions. In other words, soil samples that were rich in drought-tolerant bacteria were correlated with heartier plants. Metagenomic analysis helped link these phenotypic properties to genes that encode phytohormone production.



Drought in Morocco. [Source](#)



## AND THE PLANET'S WATER GETS HOTTER... MICROBES ARE MIGRATING TO NEW ENVIRONMENTS AS A RESULT OF WARMING TEMPERATURES

Many pathogenic microbes have adapted a thermotolerance that allows them to thrive at host body temperatures, and studies indicate that warming climates may select for, and allow, pathogenic microbes to inhabit new environments that were previously considered less-than hospitable. *Nigleria fowleri*, also known as the brain-eating amoeba, has been detected as far north as the upper-Midwest in the U.S., and scientists think warming temperatures may be to blame. *N. fowleri* is free-living and thermophilic in nature. The amoeba is commonly found in warm fresh water and soil, preferring high temperatures of up to 45°C. Although infection is rare, it is a particularly dangerous pathogen due to its ability to cross the blood/brain barrier and cause acute brain infection upon nasal passage entry, known as primary amoebic meningoencephalitis (PAM), which is typically fatal within 3-7 days of symptom onset. A Centers for Disease Control and Prevention [Emerging Infectious Diseases](#) dispatch analyzed trends in recreational water exposure associated with PAM cases in the U.S. from 1978-2018, and noted air temperature increases (compared with 20-year historic averages) in the 2 weeks prior to PAM exposures in the Midwest. The observation led researchers to suggest that warming temperatures, along with increased recreational water usage, may be contributing to the observed northward expansion of PAM cases.

Other dangerous pathogens belonging to the *Vibrio* genus have been increasing in prevalence in marine environments. It has previously been determined that [slight increases in water temperature can have a dramatic effect on \*Vibrio\*](#) colonization, prompting scientists to recently study sea-surface temperature data around the English and Welsh coastlines in an attempt to identify locations where conditions for the presence and growth of *Vibrio* species is favorable. The [research is published in Water Research](#). Researchers collected shellfish from 3 separate locations that were experiencing sea-surface temperature increases and detected *Vibrio* species in samples from all 3 locations. Some of the specific species that were identified (i.e., *Vibrio rotiferianus* and *Vibrio jasicida*) had not previously been reported in U.K. waters. The study suggests that the increased biodiversity and prevalence of *Vibrio* species may be tied to warming water temperatures and decreased salinity caused by heavy summer rainfalls.



## WHY MICROBES CAUSE EARTHY ODORS AFTER A RAINSTORM

You might be surprised to learn that it is microbes, not earthworms, that produce the earthy smell that fills the air after a good rainstorm. The source, geosmin, is a naturally produced chemical compound that is heavily conserved in actinobacteria, myxobacteria, cyanobacteria and some fungi. Although the odorous terpene is detectable at picomolar concentrations by humans, its biological function remained elusive until the recent publication of [a study in \*Applied and Environmental Microbiology\*](#). In this study, researchers hypothesized, and were able to demonstrate, that geosmin is used as a warning signal by toxin-producing microbes to ward off eukaryotic predators. The addition of the chemical to agar plates containing *Caenorhabditis elegans*, a free-living nematode that feeds on bacterial species, repelled the worms, even in the absence of bacteria. Furthermore, a predation assay revealed that geosmin reduced *C. elegans* grazing on *Streptomyces coelicolor* bacteria. While geosmin itself was not harmful to the worms, the bacteria produced a number of toxic metabolites that were up-regulated when bacterial sporulation was triggered by grazing. Geosmin was therefore shown to benefit both predator and prey.

## HISTORIC SHIPWRECKS FORM ISLANDS OF MICROBIAL BIODIVERSITY ON THE SEABED

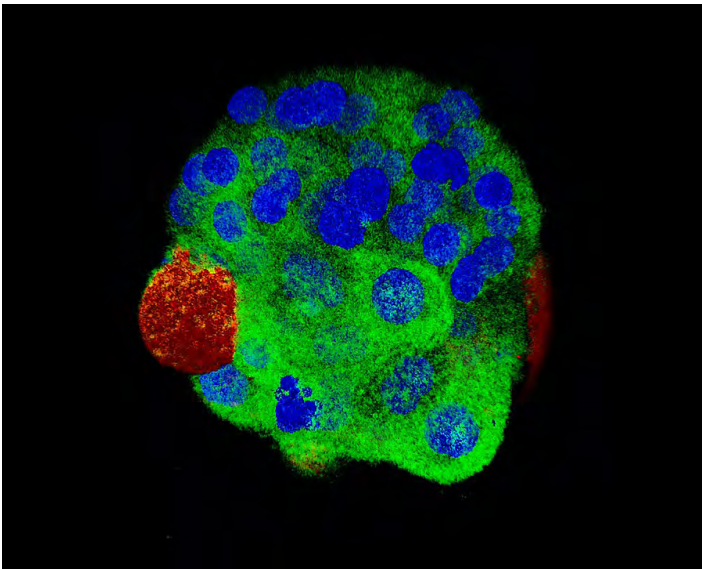
Sunken ships introduce man-made structures and materials that are unnatural to marine environments. However, over time, and with significant help from microbes, these abandoned vessels become artificial reefs teeming with life. Now researchers are investigating how wooden shipwrecks influence the biodiversity and ecology of deep-sea sediment microbiomes. In [a study published in \*Limnology and Oceanography\*](#), scientists used 16S rRNA gene amplicon sequencing to identify microbes in samples taken from 2 different deep-sea wooden shipwreck sites. The wreckage was located at water depths of 525 m and 1800 m, respectively, and samples were collected along 60 m transects extending in 4 directions from the hull of each boat. This particular experimental design allowed researchers to consider distance from the shipwreck and depth of the sediment as potential factors contributing to microbial diversity and abundance.

Sequencing revealed that the composition of the sediment microbiome was distinct and consistent across shipwreck sites, and researchers were able to identify unique habitat patches on the ocean floor that resembled those that have been associated with organic tree falls. Increased diversity of anaerobic archaea, including *Bathyarchaeia* and *Lokiarchaeia*, was detected at the deeper shipwreck site, as was a higher abundance of *Bacteroidetes*, *Chloroflexi*, *Desulfocarcinales* and *Desulfobacteriales* bacteria. Samples taken nearer the shipwreck revealed more unique taxa, including microbes that can break down complex organic matter, metabolize cellulose and fix sulfur. It is still unknown whether these microbes were introduced to the sea floor by the shipwreck or they were already present and enabled to thrive when new raw materials were introduced to the ecosystem. However, scientists have concluded that shipwrecks may serve as especially significant sources of organic matter in oligotrophic areas that are far from shore and otherwise offer low levels of nutrients.



## CONSERVED BACTERIA FOUND IN HUMAN BREAST CANCER CELLS MAY CONTRIBUTE TO METASTASIS

Microbes have been found living inside the tumor cells of a variety of cancer types. However, until recently, little was known about the functional significance of resident microbiota. A [recent study published in \*Cell\*](#) used a murine spontaneous breast-tumor model MMTV-PyMT to better understand the role of microbes in tumor growth and/or metastasis. While depletion of intracellular bacteria did not affect primary growth of the tumor, it did significantly reduce the development of secondary malignant growths. Scientists further determined that the bacteria actually promoted the survival of tumor cells during metastatic colonization by reorganizing the actin cytoskeleton to be more resistant to fluid shear stress. Introduction of select bacteria strains in two murine tumor models generated similar results, suggesting that tumor-resident bacteria stimulate metastasis and are therefore an important consideration in cancer treatment and care.



A 3D model of a heterogeneous breast tumor. [Source](#)

## MICROBES ARE KEY DRIVERS OF HONEYBEE AND VULTURE BEE HEALTH

The gut microbiome is comprised of microbial communities that reside in the digestive tract and are critical to the health and development of host species. Although the gut microbiota of adult bees is highly conserved, diet, stress, physical activity, exposure to pathogens and age have all been shown to influence microbiome function and composition. While the vast majority of bees feed on pollen and nectar, vulture bees, a small, stingless group that inhabits tropical rainforests, demonstrate a necrophagous lifestyle. These bees collect and feed on carrion, the decaying flesh of dead animals. The role of microbes in this extreme dietary shift remains undefined and of great interest. Therefore, [an \*mBio\* study](#) sought to better understand the microbial implications of a necrophagous diet. Scientists used deep sequencing, 16S rRNA genomic sequencing and community analyses to compare the microbiomes of vulture bees and closely related species that are consumers of both meat and pollen (facultatively necrophagous), as well as those that consume only pollen (obligate pollinivorous). Data revealed that the vulture bees had lost some core microbes, retained others and had notably gained an abundance of novel acidophiles, which were present in both the environment and on carrion food sources



[Source](#)

of the bees. Because acidophilic bacteria have also been found in other animals that feed on carrion, scientists hypothesized that an acidic gut is likely important for digestion and nutrition under necrophagous diets.

The gut microbiome of adult honeybees, one of the most well-characterized of all animals, is similarly conserved and adapted to dietary requirements. For example, honeybees have been shown to harbor bacteria that facilitate the digestion of toxic sugars found in the nectar of some plants. However, less is known about bacteriophage communities in bee digestive systems. In a [recently published \*mSystems\* paper](#), scientists sequenced the virome of western honeybees obtained in Austin, Texas. Sequencing results were compared to the viromes of 2 European bee populations, which were previously characterized.

Thirteen phage clusters were shared amongst European and U.S. bees. However, most of the bacteriophages associated with the bees collected in the U.S. were novel, leading researchers to suggest that the phages coevolved with their bacterial hosts. Neither study quantified bacterial abundance, and hosts for 25-50% of phages, including 1 of the 13 conserved species, could not be identified. Therefore, much about these host-microbe dynamics remains to be explored. This was the first time widespread geographic variation in bacteriophage composition was considered for honeybee populations.

## NEW INSIGHTS INTO ANTIMICROBIAL RESISTANCE

What do we know about the afterlife of dead bacteria? A recent [Applied and Environmental Microbiology](#) study found that dead cells can transfer antibiotic resistance genes to live bacteria via horizontal gene transfer. Researchers tracked the movement of extracellular antibiotic resistance genes (eARGs) from dead antibiotic-resistant *Pseudomonas stutzeri* cells to live *P. stutzeri* in antibiotic-free soil, even at low concentrations, and demonstrated that natural transformation from dead bacteria can contribute to the spread of antibiotic resistance, even in the absence of antibiotic selection.

Researchers are further investigating examples of indirect AMR selection, including the long-term use of atypical antipsychotic (AAP) medication. AAPs target dopamine and serotonin receptors and are used to treat symptoms of psychiatric disorders. However, demonstration of intrinsic antimicrobial activity has prompted concerns about the development of antimicrobial-resistant bacteria as an unintended consequence of long-term AAP treatment. Scientists recently published a study in the [Journal of Bacteriology](#) that demonstrated that *Escherichia coli* isolates demonstrated increased minimal inhibitory concentrations for ampicillin, tetracycline, ceftriaxone and levofloxacin after 6 weeks of exposure to an AAP called quetiapine at gut concentrations. Whole genome sequencing identified a number of mutations in genes that are linked to AMR, revealing an important connection between AAP use and AMR in *E. coli*.

For more about AMR and countering misconceptions in the field, check out research published in the [Journal of Microbiology and Biology Education](#), in which researchers developed a set of reading interventions and an assessment tool to confront intuitive misconceptions leading students to misconceive antibiotic resistance as a goal-oriented process of evolution, as opposed to the accrual of random genetic variations that allow some bacteria to survive, reproduce and form resilient populations. The study provides insights to the understanding of AMR by undergraduates and has clear implications for promoting a better understanding of AMR by the general public.



*Pseudomonas stutzeri* from an eye grown on agar for 2 days. [Source](#)



[Source](#)

## DOG BEHAVIOR IS A KEY FACTOR IN THE TRANSMISSION OF RABIES INFECTIONS

Scientists sought to characterize the transmission dynamics allowing rabies virus to continue circulating at low levels, despite control efforts, in Tanzanian dog populations. From 2002-2016, researchers traced rabies transmission in a population of 50,000 dogs in Tanzania. The [data, published in Science](#), was spatially resolved, allowing modeling to be applied to individual dog behaviors. It was discovered that while some dogs simply bite other animals in their surrounding environments, others travel long distances and introduce new virus lineages to neighboring populations. This dispersal can lead to cocirculation of lineages and metapopulation persistence. Overall, this study indicated that individual dog behavior is a key determinant of rabies transmission dynamics, and the authors argue that the findings may have important implications for other pathogens that circulate in spatially structured populations.