CHAPTER ONE

Extremophiles

Julie Huber is a marine oceanographer working at the Marine Biological Laboratory in Woods Hole, Massachusetts. She is thirty-four but has an easy laugh that makes her seem even younger, and if you saw her jogging on a beach you might take her for a professional volleyball player. Her accomplishments in the field of oceanography and marine biology are many; she has logged nearly a year on oceangoing research vessels, and made several descents in the most famous of all research submarines, *Alvin*. Her main research focuses on the microbes that live in and beneath the crust of the seafloor. They are organisms that sequester carbon, cycle chemicals, and affect the circulation of ocean water—all of which are activities crucial to the oceans' overall health. Yet these same organisms happen to be, in the language favored by field biologists, "vastly undersampled." Consequently, they have been little studied and so are little understood. They are also in places so difficult to reach that if Huber hopes to study them she must use techniques from various disciplines, among them geology, genetics, and molecular chemistry.

Dr. Huber is possessed of a passionate intellect that can catch you by surprise. She will look you straight in the eye and affirm that a certain development in microbiology "really excites the marine sediment community."[1] While you register the fact that there *is* a marine sediment community and consider that its members could perhaps benefit from some excitement, she is already explaining, in language that is an unself-conscious mash-up of technical and colloquial, the particular challenges of trying to detect organisms by measuring the chemical components of seawater, noting by the way that certain recent work on methanogen diversity is very cool.[2]

Lately Huber has been, in her words, "chasing seafloor eruptions." She is particularly interested in organisms that live in the Pacific, near three "seamounts" (oceanographers' term for submarine mountains). All three seamounts are geologically active, and Huber makes periodic visits to each—or, more often, to the place on the ocean's surface several kilometers above them. The visits usually hold surprises. In May 2009 she was on a research vessel in the western Pacific, only twelve hours out of port in Samoa, when the remote operating vehicle *Jason*, from 2 kilometers beneath them, began transmitting live, full-color video images of the West Mata volcano erupting. It was 3:00 a.m. on the ship, but everyone—scientists and off-duty crew alike—was crowded into one room watching the televised images of lava flowing, sometimes explosively, from the deepest erupting volcano anyone had ever seen. It was, Huber said, "definitely the coolest thing I've seen on the seafloor," adding wryly, "and I've seen a lot of seafloor."[3]

The water in the samples that *Jason* retrieved from the vicinity of the volcano was as acidic as battery acid, yet it contained living bacteria. There were fewer kinds than at similar sites, and so there was a less diverse *microbial community*—this the phrase used to describe many populations of microbes living together, sharing resources, and in various ways making life better for one another. Whether the relative paucity of kinds means the environment is too harsh for certain organisms, or whether West Mata's young age means that things are just getting started there, is an interesting and open question—one that Huber hopes to answer in the months ahead.

At the moment, Huber is managing several projects simultaneously. Late on a Friday morning in March 2010, she has just received an e-mail from the postdoctoral student studying under her and doing fieldwork on a research vessel near Guam. It seems that they had set markers and moorings on the seafloor and now couldn't find them. Huber gives a "this sort of thing happens all the time" shrug and suspects the culprit is what geologists, rather unromantically, term a "slumping event." In all likelihood a part of the seafloor slid sideways, taking the markers and moorings with it. Just another reminder, not that Huber needed reminding, that the seafloor is not the grave-quiet place that only half a century ago, many scientists believed it to be.

Huber's research may trace its origins, quite directly, to the discovery in 1977 of the so-called hydrothermal vent communities, and less directly to questions that arose in the first decades of the twentieth century—as to why the continents are shaped as they are.

A SCIENTIFIC MYSTERY

Anyone who sees a world map centered on the Atlantic Ocean cannot help but notice that the east coast of South America seems made to fit, jigsaw puzzle–like, into the inward-bending coast of Africa. In 1922 a German geologist and meteorologist named Alfred Wegner went several steps further. Assembling the evidence of fossils, mineral deposits, and scars left by glaciers, he proposed that the comparison was apt. The continents *were* pieces of a puzzle, pieces that happened to be slowly drifting apart. In the decades that followed, others developed Wegner's hypothesis into a theory of plate tectonics, which proposed that the Earth's crust is composed of plates—perhaps ten "major" ones and as many as thirty "minor" ones. It was thought that their upper parts were brittle, their lower parts warmer and more malleable, and that some might be as much as 80 kilometers thick. Geologists found evidence that molten rock was pushing into the seams where the plates had pulled apart.

If this were the case, it might help to answer a question that was surprisingly long-standing and surprisingly straightforward: Why is the chemistry of seawater what it is? Lakes like the Dead Sea—lakes with no outlet other than evaporation—are called "closed basins." They are alkaline in the extreme, and they grow more alkaline over time. Logically, since the world's oceans have no outlet, like very large closed-basin lakes, they should be very, very alkaline. Yet their pH, the measure of the acidity or alkalinity of a solution, was between 7.5 and 8—very near the middle of the scale—and this was the case for foaming breakers in the Florida Keys; for dark, dense water in the Mariana Trench; and for the frigid water lapping Antarctic icebergs. It seemed clear that

some process was at work, filtering the water and maintaining the pH, and doing it everywhere. A few scientists began to suspect undersea hot springs, and they had ideas as to their whereabouts. Hot springs on the Earth's surface were heated by the molten rock in nearby volcanoes; it seemed reasonable to expect that hot springs on the seafloor would also be near molten rock. And many thought there was molten rock in the seams between tectonic plates. Find the seams, many suspected, and you'd find the hot springs.

But no one knew for sure. Even by the early 1970s, textbooks in oceanography introduced their subject with the startling fact that we knew less about the ocean floor than we knew about the near side of the Moon. If anything, this appraisal was generous. Sonar for mapping the floor was crude, and equipment used to measure temperatures and pressures was towed on cables behind ships. Sooner or later a cable would snag on an undersea rise, and the ship would idle its engines while a dispirited crew pulled the equipment aboard. It usually came back wrecked—unless the cable just broke, in which case it didn't come back at all.

The United States Navy, however, had developed sophisticated techniques for mapping the ocean, and by the mid-1970s the navy had begun to share them with researchers. Using these techniques, scientists at Woods Hole Oceanographic Institution (WHOI) implemented a three-stage method to explore a swath of seafloor. First, the research ship *Knorr* would drop transponders. Because the seafloor is uneven, they would settle at different depths. Then their positions were measured with great precision by sonar, allowing researchers to derive a low-resolution map of the terrain. Finally, a camera vehicle—a 1.5-ton "gorilla cage" mounted with cameras, strobe lights, and power supplies—would be towed over the terrain at a cautious 4 kilometers an hour, 20 meters above the seafloor. Every few hours the crew would haul the vehicle aboard, pull the film, and develop it.

In the spring of 1977, Woods Hole researchers on the *Knorr* were mapping the seafloor in the eastern Pacific about 280 kilometers northeast of the Galápagos Islands. After one run over terrain 2 kilometers deep, the film showed white clams. It was obvious they were alive.

At such moments the research submarine *Alvin* (owned by the United States Navy and operated by WHOI) would be called into play. By the 1970s she was already something of a workhorse. In 1968 she had been lost once and recovered. Two years earlier, when an air force B-52 bomber collided with a tanker over the Mediterranean Sea and (accidentally) dropped an undetonated hydrogen bomb, *Alvin* was given its moment in Cold War history and summoned to search the ocean floor off Spain. On March 17, 1966, *Alvin*'s pilots found the bomb resting on the seafloor nearly 910 meters deep. It was raised intact. By 1977 *Alvin* had had several upgrades, but its fastest speed was a modest 4 kilometers an hour, and its lights penetrated only 15 meters. Not that any of this mattered. What *Alvin* was especially good for, and what she is still good for, is close observation. And so, when the *Knorr* found live clams 2 kilometers beneath the surface, *Alvin* was towed to the site.

Alvin's crew compartment, a hollow titanium sphere 3 meters across, holds three—a pilot and two researchers. The researchers for this particular investigation were geologists—John Corliss of Oregon State University and John Edmond of MIT. They spent most of the 2,000-meter descent peering out the Plexiglas portholes. There wasn't much to see, and even when they were a few

meters above a sloping seafloor, *Alvin*'s lights illuminated nothing but the hardened molten rock that geologists call pillow basalt. It covered the sloping floor in all directions, making for a scene that, even to a geologist, was not particularly remarkable. Then they noticed something about the water itself. It was shimmering like the air over a hot grill. Hurriedly, Corliss and Edmond took measurements and found that the water was warmer than water at this depth should be, by about 4 degrees.

The pilot took *Alvin* up the slope, and when they neared the crest of a ridge they were astonished to see, lit by the searchlight and through the shimmering water, reefs of mussels, giant clams, crabs, anemones, and fish. It was a fantastic undersea garden, an oasis vibrant with life. Corliss and Edmond did not know how the riotous island *ecosystem* around them was possible. They did know, however, that *Alvin* had only five hours of power remaining, and they spent that time, Edmond would later write, "in something close to a frenzy," measuring water temperature, conductivity, pH, and oxygen content, and taking samples of everything that *Alvin*'s mechanical arm could grab.[4] That evening, back aboard the *Knorr*, there was a small celebration. Someone had a camera and snapped photos of Corliss and Edmond—young men, bleary-eyed and smiling.

In 1830, British naturalist Edward Forbes claimed that because sunlight could not penetrate deeper than 600 meters, phytoplankton could not survive below that depth. Without phytoplankton, there was no base for a food chain. It followed, reasonably enough, that the deep ocean must be sterile.[5] By the mid-twentieth century, the processes by which oceanic life sustains itself were well understood. Sunlight supplies the energy. Nutrients in the form of nitrogen and phosphorus are brought in by rivers and streams and stirred up from the seafloor by upwelling currents. The floating single-celled plants called phytoplankton use the sunlight, nutrients, and carbon dioxide dissolved in the water. They are eaten by the tiny invertebrates called zooplankton that float freely throughout the seas and other bodies of water, and the zooplankton are eaten by shrimp and other crustaceans, all the way up the food chain to the braised tuna with lemon on your dinner plate. Obviously, such processes could operate only near the ocean surface.

In the decades that followed, however, scientists came to realize that there *was* life at great depths. Fish, crabs and other organisms lived in total darkness at enormous pressures, and survived by feeding on dead and decaying matter that sank slowly from the waters above. By the mid-twentieth century, advances in nautical engineering allowed biologists to see this life firsthand. In the mid-1940s the Swiss scientist Auguste Piccard designed a vessel he called a "bathyscaphe." Unlike its predecessor the bathysphere, a simple spherical pressure chamber lowered and raised by a cable, Piccard's new design featured a float chamber for buoyancy and a separate pressure sphere for the crew. The third bathyscaphe Piccard built was called the *Trieste*. It was sold to the United States Navy in 1957, and three years later it took Jacques Piccard (Auguste's son) and navy Lieutenant Don Walsh to the bottom of an undersea canyon called the Mariana Trench. It was there that they noticed, more than 11 kilometers beneath the surface, the deepest place in any ocean on Earth, a flatfish.[6]

Still, even in 1977, most marine biologists expected such organisms would be few and solitary. And since recycling of decaying matter in the ocean's upper levels is fairly efficient and allows very little to sink much lower before it is consumed, they expected those organisms to be quite hungry. So in 1977, when the Woods Hole expedition's chief scientist called a marine biologist named Holger Jannasch to give him the news of a thriving community of life 2 kilometers deep, Jannasch simply didn't believe him. "He was," Jannasch explained, "a geologist, after all."[7]

The expedition would conduct fourteen more descents to the site. It became apparent that Corliss and Edmond had happened upon a hot-spring field. Warm water was flowing up through every crack and fissure in a roughly circular patch of seafloor about 100 meters across. While *Alvin* investigated the newfound life below, scientists aboard the *Knorr* studied the water samples already returned, and found that all had a high concentration of hydrogen sulfide. That turned out to be a thread that wove together an entire *ecology*.

On land, some bacteria were known to derive energy from hydrogen sulfide through a process called chemosynthesis. They were rare, and most organisms took their energy, directly or indirectly, through photosynthesis. But in the dark 2 kilometers deep, chemosynthesis might be the only synthesis possible. Soon, researchers at Woods Hole developed a model to describe the process. It was this: Deep within the Earth, naturally radioactive materials produce heat that melts rock into the substance called magma. Magma is pushed up through the seams between the midocean ridges, where it cools and spreads outward to become new oceanic crust. Meanwhile, seawater continually percolates down through the crust, where the sulfate it carries combines with iron in the crust to produce hydrogen sulfide and iron oxides. When the same seawater, now heated, is pushed back up through cracks and fissures in the crust and returned to the deep ocean, it carries hydrogen sulfide that certain bacteria find quite tasty. The same bacteria absorb oxygen dissolved in the water, and some of that oxygen combines with sulfite to become sulfate.[8]

We would seem to have come full circle, returning to the chemistry we began with. But the story is not quite over. As you may recall from chemistry class, some reactions absorb energy, while others release it. The chemical reaction that yields sulfate releases energy—which the bacteria, in lieu of sunlight and in a model of efficiency—use to drive their metabolism. From here on up, the food chain of what would come to be called hydrothermal vent communities was thought to be, roughly speaking, like that in the sunlit waters 2 kilometers above.

Corliss and Edmond understood that the water issuing from the vents was probably much diluted as it rose tens of meters through the crust, and that the real action, geochemically speaking, must be in the crust, a kilometer or two deeper down. But they would never see or study that chemistry as it was happening. Or so they thought.

Two years later, researchers who were using *Alvin* to investigate warm upwelling on the Pacific Ocean floor near the Gulf of California happened upon its source: natural chimneys of sulfide minerals, 2–3 meters high, furiously pumping water black with iron sulfide and very, very hot. Soon Corliss and Edmond arrived on-site, took their turn in *Alvin*, and measured the temperature of the water released by the chimneys. It was a nearly incredible 300°C. Under an atmospheric pressure at sea level, if you try to heat water gradually, it will boil away long before it reaches that temperature; and if you heat it rapidly to that temperature, it will boil explosively like (in fact, exactly like) a geyser. It is the pressure of 2 kilometers of water that keeps the chimney's water well behaved.

For Woods Hole scientists, the heat presented some challenges. They had to design and build water samplers that would work at high temperatures, and they had to be careful to keep *Alvin* a safe distance from the chimneys, as the heat might soften

its Plexiglas portholes enough to implode them. But the work was exciting and welcome. In the months and years that followed, scientists from many different institutes and universities found more vents and more chimneys (they would come to be called "black smokers") along other midocean ridges, and near all of them, a great many living organisms.*

The theory of plate tectonics had predicted hot springs in the seams between tectonic plates. In the most dramatic fashion, Corliss and Edmond's discovery of the hydrothermal vents went a long way to support that theory, and thus closed a chapter in geology. At the same time, their discovery of life that was fed and energized by hydrothermal vents opened a new chapter in biology. Like all good chapters, it provoked questions. Exactly what sorts of organisms live in these places, and in what numbers? How did they adapt to the pressure, the dark, the heat? And how exactly did they get there to begin with?

THERMOPHILES AND HYPERTHERMOPHILES

Some of these questions had been answered several years earlier by a microbiologist named Thomas Dale Brock. Brock was an assistant professor at Indiana University, developing an inter-

* A few years after Corliss and Edmond discovered the community near the Galápagos, scientists returning to the site and conducting a more thorough reconnaissance discovered rows of slender white tubes with red filaments emerging from their tips—organisms now called tube worms, or *Riftia pachyptila*. In 2002 another expedition visited the site—informally termed the "Rose Garden"—and found that it had been covered with hardened lava. The midocean ridge giveth, and the midocean ridge taketh away. And, it seems, giveth again. The same expedition found tiny tube worms and mussels the size of a fingernail—in a place they called, naturally enough, "Rosebud." (Nevalla, "On the Seafloor")

est in microbial ecology—the study of the relationship of microorganisms with one another and with their environment. In the summer of 1964, on a brief sabbatical, he was among the thousands of tourists visiting Yellowstone National Park. Brock was captivated not by the bison and grizzly bears so much as by somewhat smaller organisms. He noticed distinct colors in the outflow channels of the hot springs, and when he took a closer look he was astonished to see what he later described as "pink gelatinous masses of material, obviously biological."[9]

The water was decidedly hot. In, fact it was nearly boiling. People had seen the pink stuff before, of course, but they did not know what Brock knew: that no microbiologist expected any microbe could live in water this hot. Microbes that live in water at temperatures between 60°C and 80°C are called "thermophiles," and microbes that live in water with a temperature of 80°C or higher are called "hyperthermophiles." But that is now. In 1964, few would have believed hyperthermophiles possible, and a standard textbook recommended that researchers incubate thermophilic bacteria at a temperature of 55°C or 60°C.[10] The temperature Brock measured in the outflow channels was 90°C. For years, Brock had suspected that research limited to lab-grown bacteria would lead to a blinkered view, and here was his vindication. Because no one had thought bacteria could survive at temperatures much higher than 60°C, no one had bothered to look for them.

Exactly how something in plain sight might pass unnoticed by researchers was a very good question. One answer, offered by historian of science Thomas Kuhn, is that people (scientists included) see what they expect to see, and may not see what they don't expect to see. By way of example, Kuhn described an experiment in which subjects were asked to identify the color and suit

of playing cards presented to them quickly and in sequence. The experiment used a trick deck. Most of its cards would be found in any deck, but a few were special, with combinations of color and suit, like a red six of spades, that do not appear in a normal deck. The test subjects, shown the cards quickly and in sequence, did not register the special cards as special, and mistakenly assigned them normal combinations of suit and color. When shown the red six of spades, for instance, many saw a red six of hearts. On the second or third run-through, some subjects began to hesitate before answering. On still more run-throughs, several became hopelessly confused, with one nearly unraveling altogether. "It didn't even look like a card that time," he said. "I don't know what color it is or whether it's a spade or a heart. I'm not even sure now what a spade looks like."[11] Only a few recognized the red six of spades *as* a red six of spades. But as soon as they did, they began to look for other special cards. Kuhn made the point that something similar happens in science. When a scientist recognizes something no one else has recognized, Kuhn wrote, there follows "a period in which conceptual categories are adjusted until the initially anomalous becomes the anticipated."[12]

In the fall of 1964, with his own conceptual categories properly adjusted, Brock anticipated the anomalous—and sought it out. He set up a laboratory in West Yellowstone and began to spend his summers exploring the boiling and superheated pools, doing a sort of microbial fishing. At particularly interesting places he would attach one end of a long string to a tree branch and the other end to a glass microscope slide, and drop the slide into the water. A few days later he would retrieve the slide and examine it. On almost every slide he found heavy bacterial growth.

Initially, other microbiologists thought Brock's discoveries too specialized and esoteric to be of wider interest. There were, after all, only so many hot springs in the world, and so there could be only so many species of thermophilic (or hyperthermophilic) bacteria living in them. Then Corliss and Edmond and their many successors found ecosystems whose basis was organisms that thrived in hot water. And the environments that they preferred, while difficult for species like *Homo sapiens* to reach, were not rare. Far from it. Midocean ridges snake along the oceanic crust for tens of thousands of kilometers. By the late 1970s, microbiologists were poring over Brock's published works for ideas on how thermophilic bacteria adapted and how thermophilic ecosystems might work. And since it was easier and a lot cheaper than mounting an expedition to a midocean ridge, quite a few began to visit his old haunts at Yellowstone.

Meanwhile, word of life on midocean ridges was reaching all corners of academia. The life sciences department at most universities and colleges has a large bulletin board mounted outside the department's main office. On that bulletin board one is likely to find a notice of a forthcoming department meeting, announcements of conferences and calls for papers, as well as more personal ephemera, like a scribbled note about lost car keys. Occasionally there is a page torn from a journal on a subject that someone thought might be of general interest. Such was the case in the fall of 1979, when it seemed that every bulletin board outside every main office of every life sciences department at every university and college—and high school too—had an article about the deep-water sulfide chimneys and the life around them.

Soon enough, biologists began to wonder whether there were other "special" cards in the deck, and many began looking. Through the 1980s and 1990s, to anyone reading the science section of a newspaper, it seemed that every other week someone had found life where (one would have thought) it had no reason

to be. There were heat lovers, cold lovers, pressure lovers, acid lovers, alkaline lovers, salt lovers, and even radiation lovers.* As a group they became known as *extremophiles*, a term that had been coined by R. D. MacElroy in 1974.[13]

Through much of the twentieth century, biologists classified organisms within a taxonomic system whose largest and most fundamental categories were Animalia and Plantae. Single-celled organisms like bacteria were included among the Plantae, it seemed to some, as an afterthought. In the 1960s, biologists began to regard the system as inadequate, especially with respect to microorganisms, and they developed a new taxonomy in which the most fundamental divisions were five "kingdoms": Animalia, Plantae, Fungi, Bacteria, and Protista. The kingdom of Protista, its boundaries particularly ill defined, included many organisms simply because they fit nowhere else. Certain microbiologists (among them evolutionary biologist Ernst Mayr) proposed a more fundamental division into two "empires." Bacteria, whose cells were relatively small and lacked a nucleus, were classified as *prokaryotes* (*pro* meaning "before" and *karyote* meaning "kernel" or "nucleus"); and the other four kingdoms, whose organisms were composed of larger, nucleated cells, were classified as *eukaryotes* (*eu* meaning "true").

In the 1960s, microbiologist Carl Woese and his colleagues began to sequence ribosomal *RNA* and realized that many microorganisms that had been classified as bacteria (under a light microscope they *looked like* bacteria) were in fact fundamentally different. The categories were redrawn yet again, this time as three "domains." The eukaryotes were called *Eukarya*, and the

* Or, thermophiles and hyperthermophiles, psychrophiles or cryophiles, barophiles or piezophiles, acidophiles, alkaliphiles, halophiles, and radiophiles.

prokaryotes were split into the domain *Bacteria* and the newly discovered domain *Archaea*. Woese's taxonomy is especially pertinent to our interests here. While extremophiles include members of all three domains, most are archaea.

Of course, since they are as unlike each other as they are unlike other life, extremophiles are a group only in the sense that "all composers not Beethoven" or "all painters not Monet" are a group. Any given extremophile can be represented by a different outlying point on a bell curve, and there are bell curves for temperature, pressure, and pH. Many, like a certain species of *Acidianus* that thrives at high temperatures and low pH levels, can be represented by outlying points on *two* bell curves.[14] What counts as extreme, of course, depends on who is ringing the bell. R. D. MacElroy, presumably, had a body temperature of 98.6°F and most probably a distaste for strong acids. If the *Acidianus* species were to categorize him, it would call him a "psychrophile" and an "alkaliphile"—a cold lover and an alkaline lover.[15]

In any case, by the 1990s the search for extremophiles had accelerated. NASA, interested in learning how organisms might adapt to harsh environments like the subsurface of Mars, funded numerous research programs—some independently, some with the National Science Foundation. In 1996 a group of biologists convened the first International Conference on Extremophiles. Within a few years, researchers in the new field had established a journal and a professional society, and had published thousands of papers.

One point of agreement in all this research was that if there is a limit, an outer boundary beyond which the most extreme of extremophiles cannot pass, it was probably set by the swish, gurgle, and drip of liquid water. It so happens that every place scientists have found life, they have also found liquid water or

evidence of its presence. And almost every place they have found liquid water, they have found life.[16]

WATER

In a list of chemicals arranged by their molecular weight, you would expect water, with a lower weight than oxygen or carbon dioxide, to be a gas at room temperature. In fact, the only reason water is a liquid hereabouts is that its molecule is polarized—the two hydrogen atoms on one side of the oxygen atom holding a slight positive charge, the oxygen atom itself holding a modest negative charge. It is an arrangement that allows water molecules to form bonds that are gentle, yet strong enough to make water bead on glass, to let it be pulled upward through a plant stem, and to endow it with surface tension—that intriguing quality by which molecules on the liquid's surface are attracted to each other more powerfully than they are to the air molecules above them or the water molecules below them.

The charged poles that pull water molecules together are the very feature that enables them to pull other molecules apart. Chemists speak of water as an unusually versatile solvent. Like the perfect dinner party host, water gently breaks apart couples (like sodium chloride) and large groups (like sugars and amino acids). Chemists also speak of water as a very good medium for diffusion. Again like that perfect host, water provides its guests an environment in which their parts can move and mingle freely. This environment, it should be said, happens to be particularly congenial to life. Water offers protection from DNA-damaging ultraviolet radiation, and it holds heat so well that temperatures near the ocean floor are unchanging year-round. And because, by comparison with other chemicals, water stays liquid at a very wide range of temperatures (in fact, a range of 100 degrees on the scale that is based on that very liquidity), life can operate at that same wide range.

One of water's properties is at once so peculiar and so conducive to life's presumed beginnings and long-term well-being that some nineteenth-century naturalists pointed to it as evidence of intelligent design.[17] If water were like most liquids, it would become denser and heavier when it froze. Ice would sink, and bodies of water in colder climes would radiate away heat and freeze solid from the bottom up. Life in those places—especially aquatic life—would have a very hard go of it. In fact, though, ice expands when it's frozen, becoming more voluminous by about 10 percent and forming a surface layer on lakes and oceans that insulates the water and organisms beneath.

As if all this congeniality weren't enough, water also uses dissolved compounds to make "microenvironments" within itself. The charged poles of water molecules lead other molecules to orient themselves side by side and facing in the same direction, some forming whole choreographed chorus lines, row after row of them, until they are best regarded as membranes. Some of these membranes develop into the microscopic bubbles that molecular chemists call *vesicles*, and whose interiors, some 4.6 billion years ago, may have sheltered the first self-replicating molecules and over time developed into cells.

Given all the virtues of water, we should not be surprised if organisms no one would call extreme go to astonishing lengths and employ ingenious strategies to get it. And they do. Spanish moss (*Tillandsia usneoides*) pulls water directly from the air; a species of kangaroo rat (*Dipodomys merriami*) draws it from metabolized food; and California redwood trees (*Sequoiadendron giganteum* and *Sequoia sempervirens*), by a means only

imperfectly understood, pump it to their highest branches 100 meters above the forest floor. And once they have water, organisms no one would call extreme go to great lengths to hold it, to keep it from freezing or evaporating, to distribute it within themselves, and, where possible, to recycle it.

As for extremophiles? To acquire and retain water, they go to lengths that are, well, extreme.

FIRE AND ICE

The Celsius temperature scale uses the range at which water is liquid as its central scaffolding, but that range may be extended upward into hotter temperatures if, as we've seen, the water is kept under pressure. It may be extended downward into colder temperatures if the water is mixed with something else. Extremophiles are quite willing to exploit this wider range of temperature, and biologists are interested in the strategies they use to do it.

To appreciate the ingenuity of those strategies requires a brief refresher in biology. The *cell* is the smallest structural unit of an organism that can function independently. The cells in you and me, and in any other multicellular being, have a nucleus that contains their DNA. In the rather simpler cells of bacteria and archaea (groups to which all microbial extremophiles belong), the DNA floats freely in the semiliquid *cytoplasm*. In the cytoplasm are large molecules called *proteins* that initiate and accelerate chemical reactions in the cell and (in some cases) act as a supporting structure. The DNA, cytoplasm, and proteins are held inside a plasma membrane covered by a cell wall. The membrane protects what is inside the cell from the harsh environment outside it, and the wall prevents the cell, in certain situations, from expanding and bursting. The membrane and the proteins in the cytoplasm inside happen to be especially vulnerable to high temperatures. In water approaching boiling, the cell membrane grows more and more watery, eventually becoming too porous to do its job, while the proteins inside it are twisted, bent, or just plain broken (or as microbiologists put it, "denatured"), and so made useless.

To stay healthy in hot water, some thermophiles substitute the weaker parts of proteins with parts that are more durable and heat resistant. This is probably the method used by the hot-water record holder at present, a bacterium retrieved from a hydrothermal vent off Puget Sound. In 2003, University of Massachusetts microbiologists Derek Lovley and Kazem Kashefi had cultured the bacterium successfully and were curious about how much heat it could tolerate. They increased the temperature to 100°C, and the bacterium kept growing. The only means to still-higher temperatures that they had on hand was an autoclave, the pressurized steam–heated vessel commonly used to sterilize medical equipment—an instrument, one can't help but note, designed not to culture microorganisms, but to kill them. They left the bacterium cooking in the autoclave for ten hours. The bacterium reproduced at 121°C and survived for two hours at 130°C. "We were," Lovley said, "truly amazed."[18]

There are reports of microbes, also living near hydrothermal vents, that survive at still-higher temperatures, but collecting samples in the vicinity of hydrothermal vents is difficult, and the samples in question may have been contaminated. Still, since scientists can imagine substituting parts that would allow a cell to hold up under even higher temperatures, a confirmed finding would not be particularly surprising. As the NRC's *Limits of*

Organic Life report observes, "the upper temperature limit for life is yet to be determined."[19]

As to the lower temperature limit? Ice threatens an organism by an act of omission, denying the organism the solvent it needs to work its chemistry. It also threatens with an act of commission: ice crystals can easily tear a cell membrane. When water inside a cell freezes, the result is, in the ominous language of one paper, "almost invariably lethal."[20]

If water didn't mix well and life insisted on taking its drinks straight up, the coldest temperature at which an unprotected cell could survive would be 0°C, and we could learn all there was to learn about the chemistry of water in an afternoon. But as it happens, water will mix readily with any number of solutes. Stir in the right salts and you can keep water liquid at –30°C. Add some organic solvents and the temperature can go lower still. Where organisms can supply these salts and organic solvents, they will.* Some keep the juices flowing by increasing the concentration of solutes between cells; others, by modifying lipids and proteins in cell membranes. Mix in an amino acid like methionine and an organic compound like ethylene glycol and you can expect that enzymes, the proteins that act as biochemical *catalysts*, speeding up reaction rates, will still do their catalyzing at a chilly –100°C.

Ideas of how organisms might adapt to mixes of water and ammonia or water and liquid methane are what get biologists (and especially *astrobiologists*, who hypothesize about extraterrestrial life) excited about places like Saturn's moon Titan, where the warmest midday temperature might be –179°C, water

* It's worth noting that *organic* does not mean "living or once living"; it means "denoting or relating to chemical compounds containing carbon."

ice is hard as granite, and methane, a gas in our atmosphere, is cooled to a liquid. Exactly how low, under the limbo stick of temperature, can life go? The NRC report proffers that, given the right solvent, "it is possible that there is no low temperature limit for enzyme activity or cell growth."[21]

THE CHALLENGE OF SALT

It is not quite accurate to say that all extremophiles were identified after the discoveries of Corliss and Edmond. Some, including members of a group called "halophiles" (salt lovers), were found decades earlier. In the late 1930s, a graduate student named Benjamin Volcani, then studying at Hebrew University in Jerusalem, began to look for microorganisms in the Dead Sea. It was, to many, a curious pursuit. Hydrologically speaking, the Dead Sea is a closed-basin lake. In recent years, with the diversion of water from the Jordan River, its only substantial inflow, the Dead Sea has grown saltier and more alkaline. But even in the 1930s, its waters could be five times as salty as seawater, and often reached the point of saturation.[22]

The threat of salt water to a cell derives from the tendency of water molecules to balance the concentration of solutes on either side of a cell membrane. Salt water outside a cell will pull water from the cytoplasm inside through the membrane, and the cytoplasm will dry up.

In the 1930s, one would have had good reason to suppose that the water in the Dead Sea was lifeless, and many did. And so it came as no small surprise when Volcani found not merely a few organisms, but a thriving microbial community.[23] They had solved the salt problem, as do many archaea and bacteria in brine lakes everywhere, with an "if you can't beat 'em, join 'em" strat-

egy, keeping high concentrations of salt in their cytoplasm, and so balancing the concentration inside against the concentration outside. But enough salt inside a cell can cause other problems. It will, for instance, bond with the water molecules that normally coat proteins, stripping them of that protection and making them vulnerable to denaturing. It turns out that the proteins in the cells of salt-loving archaea and bacteria have defenses—like, for instance, charged amino acids on their surfaces that hold on to the watery coating.

THE TEST OF ACID

On a shelf in her tidy, book-lined office in Woods Hole, biologist Linda Amaral Zettler keeps a small glass vial that she purchased in a tourist shop. It contains a few milliliters of what, in another setting, you might suppose to be red wine—perhaps a cabernet. In fact though, it is not quaffable, at least not by *mesophiles* like us. The liquid in the vial is a dilute acid laced with heavy metals, and it is from the Rio Tinto, a river in southwestern Spain.

The Rio Tinto's source is an iron ore deposit—or rather, what's left of one. The site has been mined, literally, since Paleolithic times, and what remains is a crater filled with water more acidic than vinegar. It is this acidity that dissolves iron, and it is the iron, oxidized by bacteria and exposed to air, that gives the water its reddish color—an indication of the high concentration of metals that the river maintains for all its 600-mile length, as it winds through rust-colored hills and scrub pines to empty, finally, into the Atlantic.

For years many had assumed the river was lifeless. As Dr. Amaral Zettler will tell you, they did not look very closely.[24] Even without a field microscope, anyone can see films of algae on seeping walls along the river's edges and, attached to rocks beneath the surface, green filaments of algae and whitish filaments of fungi waving in the current. But perhaps more surprising is what is living in and among the films and filaments. There are amoebas, ciliates, euglenoids, and flagellates—a thriving microbial community—not as diverse as that in a freshwater pond, but far more diverse than anyone expected.

Amaral Zettler is interested in many aspects of these organisms—one of which, quite naturally, is exactly how they manage to survive. Some set up defenses at the cell membrane, mostly with added proteins, that keep the inside at a more neutral pH and mostly free of metals. Others accumulate metals inside the cell, evidently without doing themselves serious harm. But research on the subject has barely begun, and it is probable—in fact, likely—that the microbial life in the Rio Tinto is protecting itself by other means as well.[25]

GOING WITHOUT

Readers of a certain age will recall an advertisement found in the back pages of many comic books, alongside the X-ray glasses and hovercraft plans, for "sea monkeys." An illustration promised an underwater city bustling with miniature creatures that looked a bit like chimpanzees, if chimpanzees had spiny dorsal fins and webbed fingers and toes. It was, so we readers were led to believe, a completely self-contained alien civilization we could keep on the dresser in our bedroom. What actually arrived in the mail was less miraculous, but only slightly. It was a small foil packet containing what looked like coarse-grained paprika. If you poured it into a glass of warm salt water and held a magnifying lens to the side of the glass, you would see tiny creatures uncurl, wriggle, and swim. In fact, they were brine shrimp (*Artemia salina*).

The shrimp had survived without water through a trick shared by many organisms—including bacteria, yeasts, fungi, plants, and insects. It is a process called "anhydrobiosis," by which cells shut down their whole metabolism and simply wait, as it were, for a rainy day. Some can wait a very long time. In the 1960s, archaeologists excavating Masada, the fortress in the Judean desert built around 35 BCE, found date seeds. Radiocarbon testing dated the seeds' shell fragments to the same period, and someone thought it might be interesting to see what would happen if the seeds were planted. Of the three, one germinated and soon grew into a healthy meter-tall plant.[26]

These remarkable examples notwithstanding, the undisputed champions of longevity are not any particular organism, but the dormant stage in the life cycles of many bacteria, plants, algae, and fungi. They are the small, lightweight, stripped-down versions of seeds known as *spores*. As a group, spores are profligate (a single mushroom may release millions), but as individuals they are downright spartan. They keep within them little, if any, stored food, and evidently they don't need much. In 1995, scientists resuscitated *Bacillus* spores that had been trapped in amber at least 25 million years.[27] And spores are inventive, making salt (a cell's enemy) into a shield. When salt water evaporates, it may leave deposits that have, trapped within them, tiny pockets of water called "brine inclusions," microenvironments in which spores can survive. A *Bacillus* spore has been reported revived from brine inclusions thought to be 250 million years old—older, that is, than the first mammals.[28]

To many scientists in the late nineteenth century, spores seemed overengineered, far tougher than they needed to be; and some wondered whether they might have evolved in an environment much harsher than any on Earth. If they did, then they might explain life's origin.

In the early nineteenth century, many natural philosophers held that organisms arise by spontaneous generation from organic matter. In 1860, French chemist and microbiologist Louis Pasteur conducted a series of experiments involving much care and many flasks and filters, and demonstrated that such could not be the case. Two possibilities for life's origin on Earth remained: either life had arisen in the distant past, in the form of organisms far simpler than any in existence in 1860; or it had come from somewhere else. The second hypothesis, now termed *panspermia*, was put forth a few years after Pasteur's work by Lord William Thomson Kelvin, who suggested that life originating on another world may have arrived on Earth via "seed-bearing meteoric stones."[29]

Such a trip would not be easy. Suppose it were from Mars to Earth. An organism, actively metabolizing or dormant inside a fragment of Martian rock, would have to be well positioned—not so near a meteor strike that it would be vaporized, but near enough that it could ride the blast's shock waves (and withstand tremendous g-forces and heat) up through the atmosphere and into interplanetary space. Once in space, it would have to survive vacuum, radiation, and extremes of temperature, and it would have to do so for years, decades, or perhaps centuries. Finally, it would have to withstand a fiery entry, along with more g-forces, into Earth's atmosphere, ending its journey with an arrival violent enough to leave a crater.

Given spores' well-known feats of endurance, many astrobiologists have wondered whether they might be up for the trip, and a few have devised experiments to simulate one. If you were

a spore, you might regard astrobiologists as the sum of all fears. Astrobiologists have baked spores, frozen them, irradiated them, fired them from guns, and slammed them between quartz plates with explosives. And in case such simulations fell short of the rigors of actual space travel, they placed them aboard NASA's orbiting Long Duration Exposure Facility and left them outside the spacecraft, unprotected except for a thin aluminum cover, for six years. At present, despite its advocacy by several respected scientists, panspermia lacks widespread support. Nonetheless, the upshot of all these experiments is that spores can withstand a violent launch and reentry, and that as long as they are shielded from ultraviolet radiation with a few centimeters of soil or rock, they are quite capable of surviving in space for decades—long enough for travel among planets within the Solar System. If life on Earth did come from elsewhere, it could have made the journey as a spore.

NOTHING LIKE THE SUN

Microbes collectively called "intraterrestrials" have been found several kilometers beneath Earth's surface, making for a kind of subterranean *biosphere*.[30] Bacteria have been found in rock samples taken several hundred meters below the seafloor, even in places where the seafloor itself is several kilometers below sea level. No one knows how many organisms are living in this environment, but the number may be large. One recent study found between a million and a billion bacteria per gram of rock. It may be that a large proportion of all bacteria on Earth live below the floor of the sea, where their metabolisms are driven by energy from various sources (like natural radioactivity) that are utterly independent of the Sun. But even extremophiles on Earth's surface have been discovered exploiting unusual energy sources. One fungus was found in the water core of the Chernobyl nuclear reactor, ingeniously and fearlessly converting nuclear radiation into usable energy and managing radiation damage by keeping copies of the same chromosome in every cell.[31]

THE PRESENT

A list of extremophilic world record holders that elicits a "wow" also risks a dismissal—an assumption that they are freaks in a biological sideshow, having little to do with biologists' larger interests. In fact, though, there are real, baseline reasons to count extremophiles as important players in the epic of life on Earth. Until the late twentieth century, many biologists supposed that all life on Earth began in the "warm little pond" that Darwin's contemporaries favored or its more sophisticated successor, the "*prebiotic* soup" that Stanley Miller and Harold Urey tried to replicate in their famous experiments in the 1950s.* These suppositions and many others, along with a century or so of thought, were challenged when, three years after Corliss and Edmond discovered hydrothermal vent communities, Corliss and a group of colleagues published a paper arguing that life might have begun in or near a hydrothermal vent.[32] Recent evidence suggests that thermophiles much like those now living near the vents may have been the ancestors of all life on Earth.[33]

These discoveries come at a time when many mesophiles are being discovered and catalogued for the first time. Especially in a moment when species are being made extinct at a terrifying

* Experiments enshrined in textbooks but whose presuppositions about the chemistry of the early Earth's atmosphere are now largely discredited.

rate—exceeding that of the five great extinctions in the last half-billion years, and at least a hundred times faster than the normal background rate—it may come as a surprise to learn that since 2005, about 400 species of mammals have been newly identified. But this is not necessarily good news. Many were discovered precisely because, with their habitats destroyed by logging, human settlement, climate change, pesticides, invasive species, and so on, they were disturbed, made suddenly visible—and vulnerable. We are, as it were, burning down the forest and watching to see who runs out.

It is here that extremophiles bring cause for a kind of big-picture optimism. If individual organisms and whole species are fragile, then life in general is resilient, tenacious, and, in its willingness to exploit any and all environments, downright aggressive. It is also inventive. When a suitable environment does not exist, life may create one. The most extreme extremophiles are of the domain called Archaea—the domain whose members were the first life on Earth, and a billion or so years from now, when our ever-warming Sun will have baked the ground and boiled away oceans, are likely to be the last. Even now, if the worst happened and a nearby star exploded, roasting Earth with gamma rays and exterminating all life on the surface and in the upper layers of the oceans, those assemblages of bacteria and fungi living a kilometer deep would go on as if nothing had happened. In time they would colonize the surface, probably learn the trick of synthesizing sunlight, and start things all over again.

Certainly the resulting scenery wouldn't satisfy the aesthetic of, say, nineteenth-century American landscape painters. But then again, the assemblages of bacteria and fungi probably wouldn't much care for the aesthetic of nineteenth-century American landscape painters—or ours, for that matter. And yet they and we are distant relatives. In fact, all life we know shares certain basic features. If you could take a cell from any organism—an alga, a giant sequoia, a condor, or your second cousin—and dive through its cell membrane and into its cytoplasm, you would find precisely the same nucleic acids and proteins doing precisely the same things in precisely the same ways.[34]

In fact, these shared features are what lead evolutionary biologists to suspect that everything that lives and has ever lived is descended from a single common ancestor, a microbe that metabolized some 3.5–3.8 billion years ago and (luckily for us) reproduced.[35] You might expect that, given its role as the very origin of life on Earth, this microbe would have been granted a name evocative of grandeur and myth. But, perhaps doing an end run around cultural politics, or realizing that any name would have to be borrowed from one of the microbe's descendants, biologists call it, somewhat prosaically, the *last universal common ancestor*, or *LUCA*.[36]

What interests a great many biologists is that many of the features shared by all known life seem to have no "selective advantage." In other words, it didn't have to be this way. There were, and are, alternatives. Chemists can imagine billions of organic compounds, but life uses only about 1,500. Those working in the new field of *synthetic biology* can imagine other amino acids, other proteins, and other metabolisms (or at least parts of metabolisms) that use other processes and would work just as well, perhaps better.

Quite naturally, a question arises. Was LUCA truly universal? Might there have been, in the 4.6 billion years of Earth's history, a second genesis—a moment when complex molecules gave rise to another living organism, independent of and unrelated to LUCA? Might this organism have then reproduced? Might

it even have established a line of descent that has endured as microscopic, single-celled Sasquatches into our own time? And if such organisms exist, since they arose from a chemistry different from that which produced LUCA, might they survive and even flourish beyond the limits of the most extreme of extremophiles?

These are profound and haunting questions, and they much occupy the thoughts of the several scientists (and one philosopher) we will meet in the next chapter.