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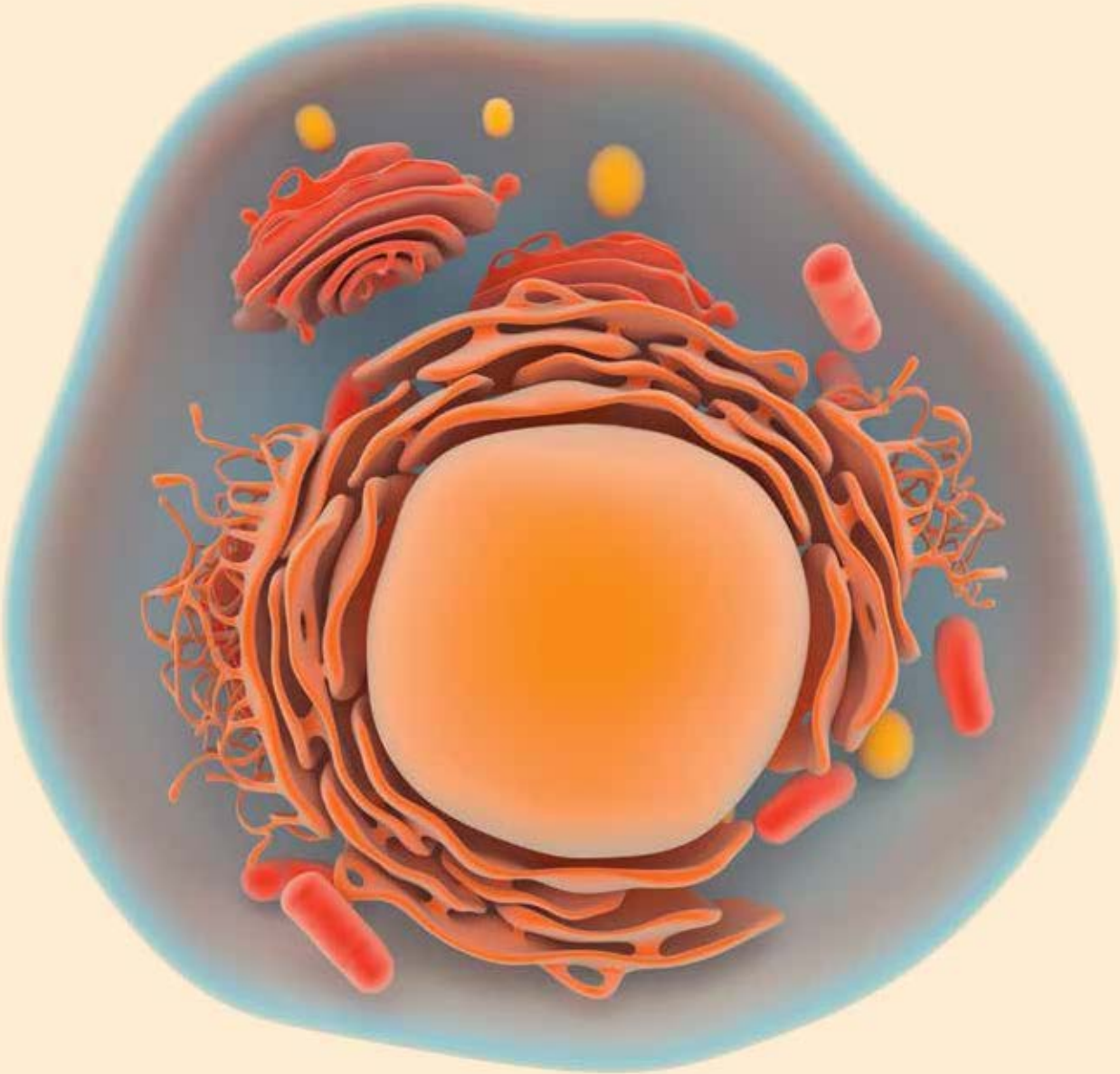
TheScientist

EXPLORING LIFE, INSPIRING INNOVATION

CELL MATTERS

FROM EUKARYOTE EVOLUTION TO BACTERIAL HIJACKING, RESEARCHERS PEER EVER FURTHER INTO THE BUILDING BLOCKS OF LIFE



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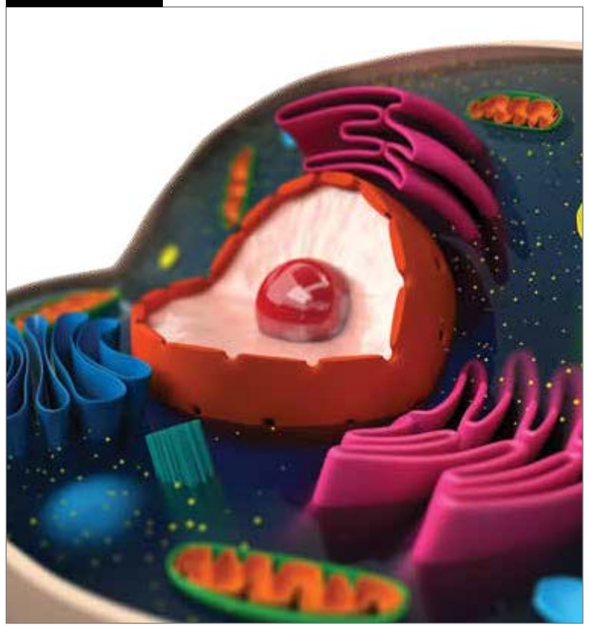

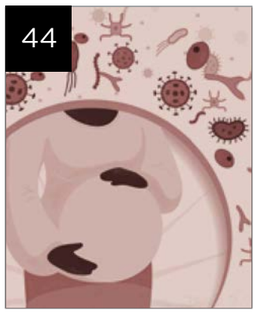

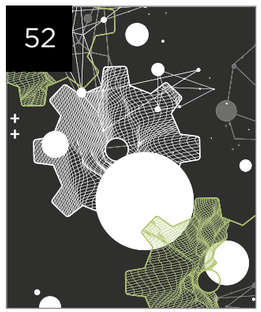

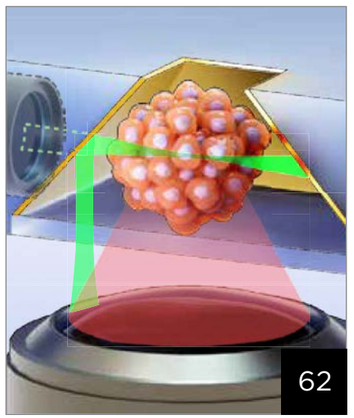

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Insights Abound Even at Journey's End

Although my time at *The Scientist* has drawn to a close, I am consistently surprised by science.

BY BOB GRANT

For more than 15 years, I have had the pleasure of helping *The Scientist* cover everything from breaking news on an emerging pandemic to the latest research breakthroughs that chip away at long-standing mysteries. Depending on the day, this job is a mixture of challenging, delightful, and enlightening. Some days are more challenging. Others contain more delights. But one thing has remained constant throughout the entirety of my tenure at this publication: Science and the world it probes never cease to amaze me.

Recently, I have been reminded of this fact in a beautifully mundane way. During an early-November news meeting—the type we hold weekly to explore the life science research goings-on—members of the editorial staff discussed a study that purported to uncover the neural circuitry responsible for controlling vomiting. The research, published in a November issue of *Cell*, used mice as experimental models. The authors of the paper exposed the animals to bacterial toxins and a chemotherapeutic drug to induce a behavioral reaction, and then traced the signaling cascade that caused the response.

ANDRZEJ KRAUZE

But here's the catch: Mice can't vomit. It's not that they won't vomit or that it's exceedingly difficult to elicit that particular behavior in *Mus musculus*. Mice, by dint of their physiology, cannot vomit. Now, anyone who writes about or conducts science, especially biomedical science, is fully aware that the humble mouse is one of, if not *the*, most common experimental organisms. We at *The Scientist* are virtually awash in mice, as our remit is to cover basic biological research, for which mice have long served as subjects. And yet, this simple fact of mouse biology had escaped my appreciation for all these years.

That physiological reality stopped me in my tracks. How did modern mice, not to mention their evolutionary ancestors, persist without the capacity to forcefully eject toxins or otherwise unpalatable substances from their digestive tracts? What, indeed, does a mouse do when it finds itself in the precarious position of having ingested something that might cause many other mammals to lose their lunch, perhaps saving their lives in the process?

These questions pestered me enough to do some cursory reconnaissance, which taught me that mice aren't the only species in that particular boat. Rodents writ large—squirrels, rats, gophers, etc.—can't vomit. Horses, too, are part of the puke-proof club. Fascinating. And it was only recently that researchers began to explore why it is that rodents lack what would seem to be an advantageous reflex. (Spoiler alert: The answer lies deep within the brain stem, where the neurological components necessary to initiate vomiting appear to be missing.)

With regard to the recent study, deftly covered for our website by intern Katherine Irving, the authors creatively circumnavigated the curious murine deficiency by using a proxy for nausea involving contorted facial expressions and contracted abdominal muscles. The rest is living history, with a neatly described pathway from toxin to serotonin to a specific region of the mouse brain, which houses neurons that fire to initiate retching.

It's an exciting and oddly comforting feeling to know that we don't need to peer into the darkest reaches of space to feel afloat in a sea of potential knowledge.

This particular example involves me, a nonscientist, being caught off guard by a scientific fact that is surely known by a great many scientists. Nevertheless, learning something so fundamental about the biology of a ubiquitous laboratory animal reminded me that, for all that humanity has learned about the natural world, surprises still abound. If we extend the perimeters of our inquiry beyond Earth and into the universe, there are even more massive unknowns. Dark matter, dark energy, the nature and lifespan of black holes, just to name a few.

It's an exciting and oddly comforting feeling to know that we don't need to peer into the darkest reaches of space to feel afloat in a sea of potential knowledge. We can find pockets of discovery and astonishment much closer to home. In mice, for example. And deep within the cells and molecules of our own bodies—indeed, within the very brains we use for all this puzzling—lie mysteries untapped, insights waiting to be unearthed.

I am moving on from *The Scientist*, and I leave behind a body of work that I hope captures my wonder and amazement with the natural world and of the concerted human effort to understand its intricacies. But I sincerely hope I never move past the giddy fascination that I feel whenever I brush against individuals driven by a similar impulse, be they science journalists covering emerging concepts or researchers on the front lines of discovery. ■

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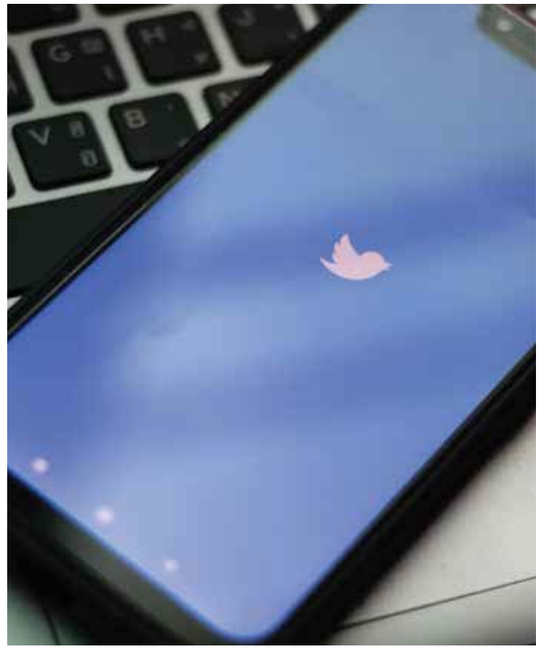
Speaking of Science

"At some level, I made a choice that I don't want to support, personally, his ecosystem."

—Astronomer **Mark McCaughrean** of the European Space Agency, speaking to *Science* about his decision to start using a different social media platform after Elon Musk purchased Twitter in October (November 4)



"[This period in science is] by no means just a return to business as usual. There is much more interest in academe in training researchers in public engagement, very much including training in communication."

—**Mary Woolley**, president of Research! America, speaking with *Inside Higher Ed* about an Elsevier report she contributed to on the pandemic-era research environment (November 8)

"Currently, avian flu outbreaks have been limited in humans because the virus doesn't spread easily between us. But this is a ticking timebomb. A mutation that makes this virus circulate more easily between humans is possible."

—**Devi Sridhar**, chair of global public health at the University of Edinburgh, in a recent story in *The Guardian* about the ongoing bird flu epidemic (November 9)

"I think, inside baseball, many doctors of academic centers are aware that he's on the Columbia faculty and feel angry about it."

—New York University bioethicist **Arthur Caplan**, speaking with *The Eye* about cardiothoracic surgeon Mehmet Oz. Oz, who last month lost a bid for the US Senate, has drawn fire for years for promoting what critics say is pseudoscience. (November 2)

"There should be a unified response, where everybody realizes that the enemy is the virus, not each other. We need to do everything we can to protect ourselves and protect each other."

—**Anthony Fauci**, director of the National Institute of Allergy and Infectious Diseases (NIAID), speaking with *Wired* about his experiences during the COVID-19 pandemic, in advance of his retirement at the end of the year (November 8)

"I was looking forward to a robust discussion on the topic of backlash against public health officials. Unfortunately, there are some people who have made their wishes known that they oppose such crucial civil discourse."

—Public health expert **Leana Wen** in a statement explaining why she would not participate in a panel at the American Public Health Association after threats were made on her safety (November 8)

Eating Our Way Out of Trouble

The key to averting cataclysmic events such as pandemics, climate change, and mass extinctions, lies partly in what's on our plates.

BY GIULIA WEGNER AND KRIS A. MURRAY

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The world may be at greater risk of infectious diseases that originate in wildlife because people are increasingly encroaching on natural habitats in the tropics to graze livestock and hunt wild animals. Devastating pandemics such as HIV/AIDS, Ebola, and COVID-19, all of which likely originated in wildlife, are reminders of how environmental destruction and infectious disease are intertwined. Tropical deforestation and overhunting are also at the root of global warming and mass species extinction.

All of these phenomena suggest that what we choose to eat has a fundamental impact on our health and that of the planet.

We recently conducted a review of the scientific literature to explore how wildlife-origin diseases, global warming, and mass species extinction are linked to the global food system. Our second objective was to explore reparative actions that governments, NGOs, and each one of us can undertake.

From the perspective of individual consumers, the global population needs to shift to diets low in livestock-sourced foods to stem human encroachment on tropical areas of wilderness. Second, there is a need to curb wildmeat demand in tropical cities.

Eating less foods from livestock

Closer to the equator, biodiversity becomes richer. These tropical regions have historically seen less development and are typically teeming with wildlife and carbon stored in the form of abundant vegetation. But in recent decades, agricultural frontiers have expanded rapidly into tropical forests. This unprecedented expansion of farmland for grazing and feed production may be increasing contact between wildlife, people, and livestock, which may enhance the likelihood of pathogens jumping from one to the other.

Such habitat destruction also has a negative impact on large herbivores and predators, as they lose sources of food and breeding grounds. This can lead to an increase in generalist species of rodents, bats, birds, and primates that are better adapted to thriving in human-modified landscapes. Some of these species are known reservoirs for infectious diseases of livestock and humans. For example, the white-footed mouse (*Peromyscus leucopus*) is a reservoir host for the bacterium *Borrelia burgdorferi*, which causes Lyme disease, while some fruit bats (family Pteropodidae) are reservoir hosts for Nipah virus and probably Ebola virus. Intensive livestock farms further increase the likelihood that domesticated animals can serve as intermediate hosts for wildlife-origin diseases, thereby amplifying the risk of human contagion. (See illustration on page 13.)

In addition, if the human population continues to grow and adopt diets rich in livestock-sourced foods, it's unlikely that global warming can be kept well below 2°C and that the rate of species extinction can be slowed. This is because livestock production has the largest environmental footprint of all food production systems in terms of land and water use, greenhouse gas emissions, and pollution of terrestrial and aquatic systems.

Asking everyone to become vegan is not realistic or even desirable. But flexitarian diets could feed the growing world population without further expanding farmland into tropical wildlands and with reductions in greenhouse gas emissions. These diets consist of large amounts of plant-based foods, including vegetable proteins like pulses, nuts, and seeds; modest amounts of fish, poultry, eggs, and dairy; and small quantities of red meat and processed animal proteins.

Paired with conversion to environmentally friendly or organic farming and reductions in food losses and wastage, diets low in livestock-sourced foods are therefore a key component of a sustainable global food system. Such a dietary shift would have other public health benefits too, such as reducing overweight and obesity, diabetes, heart diseases, and colorectal cancer.

Measures available to governments, civil society, and businesses to promote healthier and more sustainable levels of consumption of livestock-sourced foods include education in schools, training of physicians and pediatricians, eco-labels on food packaging, taxation of meat and dairy products, a statutory duty for retail and hospitality sectors, and food procurement for workplaces, schools, and hospitals.

Governments tend to dodge such interventions for fear of public backlash. But the public tends to expect government leadership in tackling such a complex challenge.

Curbing wildmeat demand in tropical cities

In the tropical forests of Africa, Asia, and South America, hunting pressure to supply nearby cities has dramatically increased over the past 30 years. In addition to imperiling vulnerable animal populations, a vigorous wildmeat trade may increase the risk of zoonotic disease transmission.

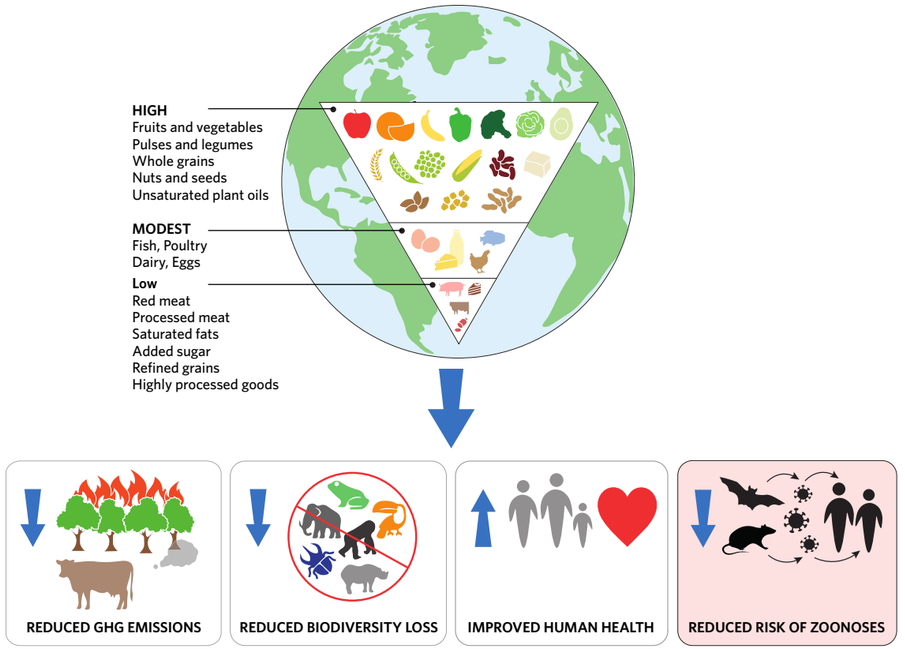
But in the absence of effective state law enforcement and sustained campaigns to reduce consumer demand, bans do not work. In fact, consumers' strong preferences for wildmeat mean that they may continue to purchase it despite price increases induced by a ban, boosting black markets. In the case of "luxury meat," increased price and rarity may even drive higher demand. Bans could also shift the wildmeat trade to illegal, unregulated channels where less attention is paid to biosecurity measures necessary to prevent contagion from wildlife-borne diseases.

Outright bans can have other undesired effects. While in most large cities, legume, fish, and livestock-sourced proteins are easily available at affordable prices, there are Indigenous people and rural communities who rely on hunted meat for vital nutrition and income. Their rights to sustainably provision themselves within their customary territories should be safeguarded.

The ideal course of action would be to contain tropical wildmeat hunting and trade by curbing demand in urban areas and extractive outposts, while supporting hunting rights and biosecurity measures among communities in remote subsistence areas.

Avoiding biohazards from animal-sourced food

Interventions in rural communities should provide wildmeat hunters, traders, and butchers with training in inexpensive biosecurity measures they can easily adopt to avoid infection from contact with wild animals. Biosecurity measures should also be extended to livestock and wildlife farms, abattoirs, food markets, and restaurants. These measures include wearing protective clothing when handling wild animals, wrapping carcasses to prevent blood from contacting cuts in people's skin, and cooking wildmeat thoroughly before eating.



ILLUSTRATOR: EMILY WRIGHT. TAKEN FROM WEGNER ET AL., eCLINICALMEDICINE, 2022

Flexitarian diets could feed the growing world population without further expanding farmland into tropical wildlands and with reductions in greenhouse gas emissions.

Other physical distancing measures should be taken in farms, pastures, and live-animal markets. These include fencing and reducing livestock densities to minimize contact with wild herbivores, planting fruit trees visited by bats at a sufficient distance from livestock sites, and limiting the number of animals on sale in live-bird markets.

Different strategies across different regions

Levels of consumption of livestock-source foods, and the degree of reliance of human communities on animal-source proteins, vary dramatically. Efforts to reduce livestock production should focus on curbing excessive consumption in wealthier countries and expanding metro-polises in less developed and emerging economies. In the poorer rural areas of resource-limited countries, home gardening as well as smallholder livestock development programs can help decrease malnutrition with limited environmental and public health impacts.

Pastoralist communities in arid rangelands and hunter-gatherer communities in tropical rainforests and arctic locales that are inhospitable to crop cultivation would instead continue to rely on animals for nutrition. Nonetheless, the minor environmental impacts of their subsistence way of living are not comparable to those of dense and better-off urban populations.

Our future depends on urgent change

The incidence of infectious diseases originating in wild animals is high and may be increasing. This may be yet another warning signal that our degradation of ecosystems is undermining the capacity of planet Earth to sustain human health and well-being.

Dietary shifts away from livestock-sourced foods and reductions in tropical urban wildmeat demand are crucial to simultaneously protect the environment, safeguard resource-limited vulnerable communities, and reduce the risk of further disease outbreaks and pandemics. We all share the responsibility to act now to prevent pollution, floods, drought, famine, and epidemics from becoming increasingly prevalent. ■

Giulia Wegner is a socioenvironmental researcher at the Wildlife Conservation Research Unit (WildCRU) of the University of Oxford in the UK. Kris Murray is an associate professor in Environment and Health at the MRC Unit The Gambia and the MRC Centre for Global Infectious Disease Analysis at Imperial College London.

Gollum in the Ivory Tower

Resource hoarding is an unfortunate reality of the research enterprise. The time has come to discuss it in an open way.

BY JOSE VALDEZ AND SANDEEP SHARMA

Humans have likely been hoarding and fiercely guarding resources since the start of recorded history. Those with access to resources and the means to defend them typically have a higher chance of survival. This tactic and the hostility it engenders are still part of the human psyche, and currently extend into scientific spheres. Just like greedy Gollum tirelessly seeking and defending the One Ring in J.R.R. Tolkien's *The Lord of the Rings*, some researchers possessively guard precious study sites, model organisms, research topics, and even entire scientific fields. These Gollums of the academic ivory tower are willing to defend their "rings" at all costs, preventing other competitors from getting too close to their research arena and severely hindering scientific progress along the way.

In one of the best-known cases of academic rivalry, paleontologists Othniel Charles Marsh and Edward Drinker Cope took this Gollum effect to the extreme with turf battles that became known as the "Bone Wars." Across the second half of the 19th century, these researchers resorted to bullying, sabotage, theft, and even the destruction of rare fossils and precious research sites to hinder access and prevent each other from making scientific discoveries in newly unearthed and fossil-rich bone beds in the American West. While the Bone Wars represent a somewhat severe example, cases of bullying, harassment, gatekeeping, and threats are all too common experiences for many in today's research community. Although this issue is rarely discussed, a recent opinion piece coauthored by one of us (J.V.) hopes to bring the Gollum Effect into the light and encourage victims of such behaviors to discuss their past experiences so we can come together to find solutions.

The published article was originally written after the authors and their colleagues, in their academic studies and early careers, had run-ins with several Gollums. This included supervisors using their positions of power to claim authorship for work they were not involved in or to discourage colleagues and other scientists from conducting experiments that they believed only they had the right to conduct. Things sometimes turned more acutely abusive, as with a particular mentor who had no direct connections with a research project, but went out of their way to thwart the research plans. In our view, this person did this because they were already working on the target species—a particularly charismatic species that was garnering a lot of grant money. In another situation, a senior scientist with a more defined territory in a particular field of research successfully plagiarized the ideas and data of a colleague, attempting to claim them as their own. In both cases, the junior researchers involved decided to keep their heads down and voices low, deeply anxious that any noise would jeopardize the projects they had planned and prepared for years and maybe even compromise their careers.

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In another example, a young scientist submitted a research paper, the culmination of years of hard work and long nights, to a highly regarded journal. Months later they received an email containing reviews and eagerly began reading. However, instead of the expected constructive feedback, they found that one of the reviewers levied an extremely harsh and disparaging review. The reviewer not only attacked the work but lobbed personal criticisms at the author as well. While journal editors often ignore such ad hominem attacks and let the review stand, this particular editor realized the negative review represented the resource guarding of a Gollum and did not reflect the quality of the work. The editor-in-chief of the journal got personally involved and contacted the reviewer, letting them know that their tone and accusations were inappropriate, that their attitude ran contrary to the advancement of science, and that they would not be invited to serve as a reviewer again. In this case, the journal's editorial team was extremely helpful and supportive. Nev-

Gollums of the academic ivory tower are willing to defend their "rings" at all costs, preventing other competitors from getting too close to their research arena and severely hindering scientific progress along the way.

ertheless, for someone just starting their professional career, such reviews can be demoralizing and can lead young authors to question their value as researchers. Situations like these have the potential to lead individuals not only to give up their specific research aspirations, but to leave science entirely.

Sometimes the damage of a Gollum's behavior can extend beyond individuals. For example, our colleagues in Asia have observed that whole research institutions, local and international NGOs, government agencies, and even foreign aid/development organizations can often behave like Gollums. This is common for popular and well-known study areas, such as UNESCO World Heritage Sites, or universally charismatic species such as tigers. This means that resources, mainly funding and financial incentives, become scarce and often controlled by a few individuals or groups, further creating power imbalances. We have found that corruption, nepotism, and steep hierarchies, in both academic and government systems, only serve to feed the Gollum.

These dynamics affect not only future generations of researchers but also the future of scientific pursuit. As we are all part of the scientific community, combating this issue requires systemic change and collaborative action by all of us, regardless of position or power. By encouraging a culture of ethical research etiquette and removing unjustifiable roadblocks, we can keep researchers excited about science and foster an environment where anyone can freely study the subject they are passionate about. However, to fully bring about a paradigm shift and increase scientific openness, we must first be open and comfortable talking about our own experiences. Sparking an honest conversation about this issue can eventually lead to a new era in which science is practiced fairly and the pursuit of discovery is accessible to all. ■

Jose Valdez *and* Sandeep Sharma *are postdoctoral researchers at the German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig.*

MIGUEL ÁNGEL GÓMEZ-SERRANO

TRUTH OR DARE: Birds such as this Kentish plover (*Charadrius alexandrinus*) use deceptive tactics to protect their eggs from predators.

The Lies Birds Tell

About five years ago, Clinton Francis and a gaggle of ornithology students were walking toward the ocean at San Simeon Beach State Park in California when they noticed a type of plover called a killdeer about 60 feet away, calling *Dee! Dee! Dee!* They hadn't seen the shorebird flush from its ground nest, but Francis, an ecologist at California Polytechnic State University, says that it would have crept slyly away from its eggs after registering the herd of humans as potential predators.

Now that it had an audience, the killdeer (*Charadrius vociferus*) began to lie through its beak. It contorted its wings in what's called the broken-wing display, feigning an injury that would make it seem unable to fly. Francis was already familiar with this sort of bird theater, typically performed for earthbound diurnal predators in the hopes that, instead of finding the killdeer's nest, they'd see a plump, apparently wounded parent as an easy meal. When this deceptive behavior works, the predator charges the killdeer, which then launches skyward to escape.

While watching the bird, Francis recalls, one of his students asked: "What other species do that display?" Francis knew the behavior was commonly associated with shorebirds, but beyond that, he wasn't sure. In response, he told the student, Wren Thompson, "You should really look into that for your research question for this class." Taking the advice and diving in, Thompson found sporadic examples across avian species through an exhaustive literature search, followed by surveys sent to ornithologists, avian ecologists, and experienced birders around the world. In the end, she and her colleagues uncovered evidence that 285 avian species perform the broken-wing display (*Proc R Soc B*, 289:20220058, 2022).

Mapping those behaviors onto the avian phylogenetic tree revealed that the trait spans from some of the most basal

bird families, including pheasants and ducks, to more recently evolved taxa such as songbirds. “It’s pretty amazing,” Francis says, adding that he was surprised how “particular clades on the avian tree of life really just light up,” including blackbirds, warblers, and sparrows. The frequent and disjointed appearance of the behavior across the tree suggests it evolved independently several times, he adds.

The analysis, published earlier this year, also indicates that predation risk has driven the trait’s evolution. “Birds that experience higher levels of predation, by visual predators in particular, tend to use the display more than those that do not,” Francis says. The team found that the farther the birds’ breeding zones were from the equator, the more likely the animals were to use the broken-wing display. One possible explanation for this relationship, Francis says, is that the portfolio of predators becomes increasingly diurnal—and more visual—towards the Earth’s poles.

“It is certainly surprising to see that broken-wing display is so widespread in phylogenetically distant groups of birds,” Miguel Ángel Gómez-Serrano, a conservation ecologist at the University of Valencia who studies deceptive nest defense behaviors but was not involved in this research, tells *The Scientist* by email. Plovers have a lot of tactics to distract predators beyond feigning broken wings, he adds. They may begin by calling to catch a predator’s eye.

TOP ACT: A female Kentish plover performs a broken-wing display.

If this doesn’t work, they may escalate to so-called false brooding: lying down to simulate incubating their eggs—something that could trick a predator into looking for the nest away from its true location. Or a plover may begin what’s known as a rodent run, mimicking a flightless mousy snack to entice the predator into chasing an apparently easy meal away from the nest. “The bird runs crouched forward with its chest close to the ground,” says Gómez-Serrano. “Often plovers place the tail folded towards the legs to [better] resemble the shape of a mouse.”

The degree of predation risk seems to dictate the form that a bird’s dishonesty assumes. When the risk is lower, a plover may fake an injury while running, giving the deceiver momentum to fly off and escape. “If nothing seems to work, or the risk of losing their offspring seems obvious—for example a predator that is right next to the nest,” Gómez-Serrano says, “the birds take even more risk by [enacting] the broken-wing display statically near the predator,” imperiling themselves by proximity as well as by losing their running start. Previous work by Gómez-Serrano’s group has found that when Kentish plovers (*Charadrius alexandrinus*) engaged more in risky stationary displays, their nests survived longer, providing evidence that the potential cost pays off (*Behav Ecol*, 28:260–69, 2016).

Sometimes, though, the price for lying is death. In 2008, on a beach in Spain, Gómez-Serrano saw a Kentish plover enacting an in-motion broken-wing display for a small predator hidden in the surrounding vegetation. While trying to lure that foe away from a nest containing days-old chicks, the plover was itself too distracted to notice a different predator: a kestrel that swooped out of the sky, snatched it up, and flew inland, likely to feed its own chicks.

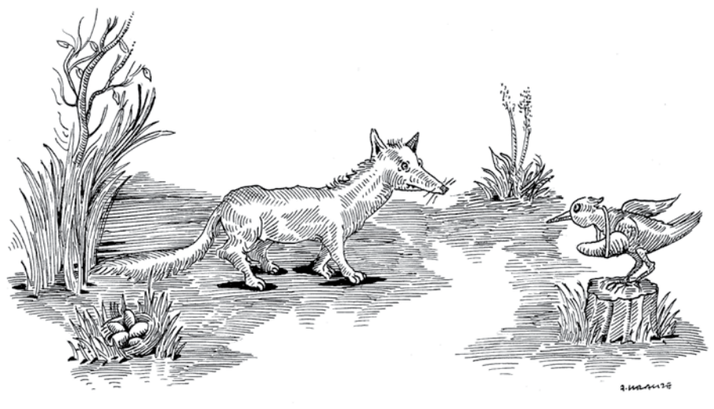
In addition to broken-wing, false brooding, and the rodent run, other documented dishonest behaviors include playing dead, feigned exhaustion, false feeding, and pseudo-sleeping. Gómez-Serrano says some birds fake eating, pecking at nothing on the ground—perhaps giving predators the impression they’re distracted and easy to sneak up on. Some birds vocalize their lies. Burrowing owls (*Athene cunicularia*) hiss like rattlesnakes to protect against ground squirrels, and fork-tailed drongos (*Dicrurus adsimilis*) mimic meerkat alarm calls to scare the mammals into abandoning food. “I think there’s some other really interesting deceptive tactics out there that are worth exploring,” and we may be unaware of many, Francis says.

Filipe Cunha, a behavioral ecologist at Wageningen University & Research in the Netherlands, happened upon a particularly unusual case of avian deception while studying Siberian jays (*Perisoreus infaustus*). “They’re definitely liars,” he says, explaining how the territorial birds fake an alarm call that’s typically reserved for alerting group members to the presence of predators such as sparrowhawks. Cunha determined that the jays deceive neighboring groups of Siberian jays to scare them into fleeing, after which the liars steal caches of scavenged meat that the tricked birds had hidden to survive the Arctic winter (*Sci Adv*, 7:eaba2862, 2021). He says that he hopes studying within-species dishonesty will shed light on how trust evolved in our own species.

A plover may fake an injury while running.

Research on avian deception highlights the importance and diversity of these behaviors as survival tools, Francis says. Consider a familiar example of a bird without known deception or indeed any other predation-avoidance behaviors: the extinct dodo, “which [people] were able to just walk up to and club because they had no evolutionary response to approaching humans or any other type of predator,” Francis says. “It’s worth keeping this quiver of tactics because otherwise reproductive success is zero.”

—Andy Carstens



MIGUEL ÁNGEL GÓMEZ-SERRANO; ANDRZEJ KRAUZE

The Shape of Whales

Whales are weird. The Cetacea clade contains the largest animal to ever live—the blue whale—as well as other gigantic baleen whales and a diverse array of toothed whales, including dolphins, porpoises, narwhals, sperm whales, and more. The group contains some of the only fully aquatic mammals that give birth to live young in saltwater. Whales’ nostrils are on the tops of their heads. The list of bizarre characteristics goes on.

Whales’ skulls are one of their crowning oddities. Skulls in general are a paleontological treasure trove, explains paleontologist and macroevolutionary ecologist Ellen Coombs, a postdoc at the National Museum of Natural History. They host the brain, the sensory organs, and the teeth, all of which can tell researchers about the animals’ behavior and diet. For several years now, Coombs has been studying whale skull peculiarities—such as the structure’s unusual asymmetry in some species—and their implications for cetacean evolution.

She first started collecting 3D scans of whale skulls in 2018, when she was a PhD student at the Natural History Museum in London. She aimed to collate data on skulls that came from not only different periods across whales’ evolutionary history, but also various geographies, from Europe and North America to Peru and New Zealand. The basic procedure was straightforward. Using a 3D scanner,

SKULL SCAN: Ellen Coombs uses a 3D scanner on skulls at the Los Angeles County Museum of Natural History.

VANESSA RHUE, LOS ANGELES COUNTY MUSEUM

she'd take several images, then process them, clean them up, and merge them into a single, coherent 3D model.

That's easier said than done when you're scanning hundreds of skulls, especially when they're from whales. The skull of a vaquita (*Phocoena sinus*), the world's smallest cetacean, is pretty manageable, as it's around the size of a melon. But the skull of a blue whale (*Balaenoptera musculus*) is more on the scale of a family sedan. Scanning a single skull could take Coombs anywhere from 30 minutes to an entire day.

Computer model of an orca (*Orcinus orca*) skull

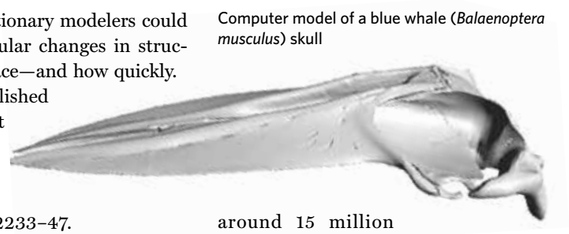
ELLEN COOMBS

Once she'd assembled the images, Coombs could place digital markers on them to note the position of particular structures. For each of the 200 or so skulls she studied, she placed more than 2,000 markers, for more than 400,000 markers in total. "It took me the good part of a year to finish," she says. Coombs adds that she also listened to a lot of podcasts during that time.

Some findings jumped out right away, she notes. For example, the odontocetes, or toothed whales, "have very asymmetrical skulls." In 2020, Coombs coauthored a paper on some of her initial findings, which revealed that skulls of the toothed whales had evolved to accommodate the melon, a mass of fatty tissue that amplifies the high-pitched calls that these whales produce for echolocation (*BMC Biol*, 18:86).

Looking deeper by mapping the skull markers onto the cetacean phylogenetic tree, she and a team of fellow whale experts and evolutionary modelers could infer when particular changes in structure had taken place—and how quickly. The findings, published this year, show that whales' cranial evolution came in three waves (*Curr Biol*, 32:P2233–47.E4, 2022). The first was right at the beginning of whale evolution, just shy of 50 million years ago, when the archaeocetes—the ancestors of modern cetaceans, which emerged in the Eocene Epoch—were first entering the water. "Within eight to twelve million years," she says, "they went from being fully terrestrial to fully aquatic."

Computer model of a blue whale (*Balaenoptera musculus*) skull



ELLEN COOMBS

The next major shift came roughly 39 million years ago when the two suborders of whales—the mysticetes (baleen whales) and the odontocetes—went their separate ways. The baleen whales began developing the long keratin sheets that enable filter feeding. That carried on until about 23 million years ago, when their rate of skull evolution slowed to a crawl. The toothed whales, meanwhile, developed echolocation, which enabled them to hunt in low-light conditions and in difficult terrain, such as murky rivers clogged with rocks or ice.

In the final wave of evolution, from roughly 18 million to 10 million years ago, there was an explosion in diversity among the toothed whales. Echolocation, unlike baleen, was a tool that could help the animals exploit many different niches, Coombs explains, thereby encouraging new adaptations and creating new species, from the river dolphins to the deep-diving sperm whales.

Anatomical scientist Paul Manger of the University of the Witwatersrand, Johannesburg, who was not involved in the work, says that one of the study's most interesting findings is that these three occurrences of rapid changes in the anatomy of the skull "correlate with changes in brain [size during] evolution." He notes, for example, the there was a significant increase in brain size right at the origin of odontocetes, and again around 15 million years ago at the emergence of Delphinoidea, the largest group of toothed whales and the taxon containing dolphins (*Anat Rec*, 281A:1247–55.E4, 2004). "So this study does support and extend previous studies and does so very nicely," Manger says.

Abdullah Gohar, a cetacean paleobiologist at Mansoura University in Egypt who was also not involved in the research, writes in an email that the paper was "a fabulous work. It's fantastic to see this monumental effort come to fruition!" He says that the extreme specialization of cetacean skulls makes them an excellent target for study, as they "capture many of the extreme shifts in feeding, respiration, and sensation."

Whales' cranial evolution came in three waves.

The sheer scale of the database is worth noting too, he says. "Large databases take a long time to compile, but they allow scientists to throw light on larger trends rather than making qualitative findings based on a single fossil." He adds that he hopes future studies "incorporate these findings to better understand cetacean evolution."

Coombs says that she's already had several researchers, including other evolutionary biologists, climate scientists, and behavioral ecologists, asking to use the dataset for further research, and adds that she's not finished with it either. "There's so many questions we can ask using this dataset alone, but there's more we can add to it too. I'm super proud."

—Connor Lynch

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A MOTH'S METHOD: *Lymantria* species make ultrasonic, mechanical rasping noises when they hear bats nearby.

Bat Repellent

Spending months in hostels in far-flung locales is more often the province of graduate students than of established researchers. But it's been the side project of a lifetime for Jesse Barber and Akito Kawahara, two scientists who have spent more than a decade criss-crossing continents to catch moths, play bat sounds at them, and see if—and how—they squeak back.

Like other nocturnal insects, moths need to contend with bats. Unlike grasshoppers or beetles, they have soft bodies without spines or hard cuticles to protect them. Yet bats' reliance on echolocation has given moths a way to avoid ending up as food: by tapping into their predators' acoustic signals. Many have evolved ears that can hear the calls of bats. Some moths make ultrasonic squeaks, chirps, or clicks to warn their predators (honestly or not) that they are poisonous. Others generate near-constant, ultrasonic buzzes capable of jamming bat sonar.

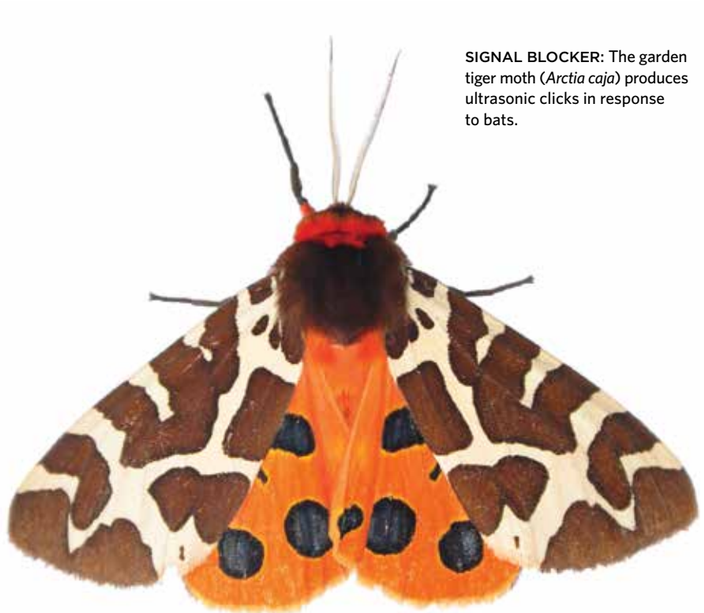
While such abilities have been documented in a range of moths, it's less clear whether these behaviors are rare evolutionary quirks or common strategies across the 160,000 or so moth species worldwide. Barber, a sensory ecologist at Boise State University, and Kawahara, an entomologist at the Florida Museum of Natural History, have been thinking about this mystery since they first crossed paths about 17 years ago at a Lepidopterists' Society meeting in Sierra Vista, Arizona—a gathering that Barber describes as "a raging good time." During the course of the doctoral work he'd just completed, Barber had stumbled across a quirk of moth behavior: Some hawkmoths (family Sphingidae) respond to recordings of bat echolocation by squeaking back at them, though it wasn't immediately clear why.

To find out more, "we got a grant to study that system and started traveling around the world, just the two of us, staying in hostels, catching moths and playing bat sounds back at them," Barber says. The pair would attract moths with ultraviolet

light, gently hold smaller moths down with locking forceps or use fishing line to tether larger moths so that they could fly but not escape, and play prerecorded bat sonar through an ultrasonic speaker. Moths responded with all sorts of sounds of their own, which the pair recorded. The large *Gonodonta bidens*, for example, made a short, light buzz that, when slowed down, sounds like a small handful of rice being poured into a plastic container, while the mechanical rasps of *Lymantria* species sounded more like the noise made by a rattlesnake.

Over roughly the next decade, Barber says, "we went to French Guiana at least five times, Borneo at least five times; we went to Mozambique a couple of times, Ecuador a couple of times," among other places, roping in more and more graduate students as they went. "We were just doing it as a side project as other grants and projects carried us around the world," he explains, "so it was kind of pieced together over time."

After testing 252 genera across most of the 28 large-bodied moth families—those that are big enough to be capable of making a sound bats can hear from a distance—the researchers documented anti-bat ultrasound production as a form of warning signal of poisonous qualities in 52 genera (*PNAS*, 119:e2117485119, 2022). They also found evidence that sonar-jamming strategies had evolved independently multiple times—at least twice in hawkmoths and four times in the Erebidae family—and noted overlap between species that signal and species that jam. Interestingly, many of the moths appeared to be converging on similar sounds, Barber says.



SIGNAL BLOCKER: The garden tiger moth (*Arctia caja*) produces ultrasonic clicks in response to bats.

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Bats' reliance on echolocation has given moths a way to avoid ending up as food.

Investigating a community of moths in Ecuador, Barber and Kawahara also found evidence of complex acoustic mimicry, featuring various combinations of moths—some jammers, some warners, some that do both, and some that make warning noises even though they aren't poisonous—all copying one another's signals. "[It] is likely that ultrasonically signaling moths comprise one of the largest mimicry complexes on earth," the authors write in their paper.

University of Bristol behavioral and sensory ecologist Marc Holderied, who was not involved in the work, says that while researchers knew that bats and moths were in an "acoustic arms race," this paper provides new information on the distribution of these strategies across moth species. "This is a very nice piece of research, and it's well executed. . . . It's something that needed to be done and it's been done very well," he says, adding that "the chapter on these acoustic signals has just increased in volume multifold." The findings on multiple convergent acoustic mimicry strategies is particularly interesting, he says. "What in the bats' brain makes this particular signal so efficient in protecting the prey?"

Now-retired behavioral ecologist Michael Greenfield, formerly of the University of Kansas, compliments the collaborative nature of the project. "When I started in ecology and evolution, it was the era of the rugged individual" and single-author publications, he says. But in this paper the authors "made use of having a team: people [were] testing on different continents, on many different species," which made a paper of this scale possible. Greenfield also notes that while Barber, Kawahara, and colleagues uncovered many novel examples of sound-producing moths, most were found in four superfamilies where they had already been documented.

Athanasios Ntelezos, a graduate student in zoology and electrophysiology at the University of Cambridge, writes in an email to *The Scientist* that while the paper adds to scientists' understanding of "how widespread this strategy is," he would have liked to see data on the effects of moths' signals. "Ideally one would want to test the function of the sounds produced by moths by pitting them against bats and comparing the effectiveness of the sound-producing group to that of a control group," he says, but "the new study is large-scale and [it] would be very hard indeed to test each moth species against bats."

The project has certainly been a massive undertaking, Barber says. "A lot of the reason it took us so long to publish it [was that] we felt the story was so incomplete that, what could we say?" After all, "to sample enough animals in a phylogeny this diverse, with this many species, is a lifetime task." Yet the result is a testament not only to a legion of collaborators the world over, but to a long-standing friendship and partnership, and the passion of everyone involved, he adds. "Something I can say about the entire list of authors: we all love moths. And, you know, that's sort of the type of scientist and person you have to be to delve this deeply into this question without getting paid to do it."

—Connor Lynch

Data Savers

Just after midnight on March 24, 1989, the Exxon supertanker Valdez slammed into Bligh Reef in Prince William Sound, Alaska. The resulting oil spill was an unprecedented disaster for the region, its fish and rich wildlife, and the people and industries who depended on them. In the aftermath, more than $150 million of civil suit settlement money was allocated to ecological research and monitoring efforts to help scientists understand and mitigate the long-term effects of the spill.

Three decades later, most of the data collected in the wake of the disaster have gone missing. A five-year project that began in 2012 to recover the original data turned up just 30 percent—the rest were never digitized, never shared, or kept in a format inaccessible to outside researchers. In purely financial terms, a new study estimates that more than $100 million was spent to collect data that, effectively, no longer exist (*Proc R Soc B*, 289:20220938, 2022).

"Truly wild" is how University of Arizona community ecologist and study coauthor Ellen Bledsoe describes the scale of the Valdez data loss. Tallying it up "was definitely eye-opening, just as a way of quantifying monetarily how much data is lost." Bledsoe and colleagues at the Canadian Institute of Ecology and Evolution (CIEE) published their estimate earlier this year alongside guidelines for the recovery and archiving of important ecological data. As part of CIEE's Living Data Project, their goal is to identify datasets in danger of loss and take steps to preserve them before they disappear into the ether. Data rescue is the official term, but Bledsoe says she likes to think of it as "data necromancy"—bringing data back from the dead.

The project tackles a common contradiction in science. Without data, there is nothing to analyze and no way to test any hypothesis. Yet once they have produced results and publications, data are sometimes treated as tools that have outlived their usefulness, rather than the valuable, and often irreplaceable, records that they are. "Data have been seen as not exciting. They're not science, they're not proper idea generation," says CIEE board member Alison Specht. "They're a means to an end, and the curation, the management, and sharing of data was a time-consuming and rather low-grade task, and not usually funded."

The Living Data Project, which got its funding from the Natural Sciences and Engineering Research Council of Canada (NSERC), aims to address both the immediate problem of data loss and the underlying cultural causes. The project trains graduate students on data management, then matches them with data owners such as research organizations or retiring academics. Students help clean and process aging datasets, eventually sharing them in an accessible repository.

"There are no courses in most biology curricula that teach people how to manage their data," says Dominique Roche, a postdoctoral fellow at the University of Neuchâtel in Switzerland and coauthor on the Living Data Project paper. "It seems like such an essential skill. I guess it's assumed that people who do research know how to work with data, but that's the biggest fallacy ever."

TREASURE SECURED: Ecology professor George H. La Roi's data—collected over 35 years of studying North American boreal forests and stored in notebooks, CD-ROMs, and slides—are now preserved by the Living Data Project.

THE LIVING DATA PROJECT

IN STORAGE: The Living Data Project helped to archive these boxes of files from the basement of the Atlantic Forestry Centre in Fredericton, New Brunswick.

DIANE SRIVASTAVA

The number of older projects in need of rescue can be daunting—sharing data was rare in ecology before journals and funding agencies started to require it in the early 2010s. So the team's new paper gives guidelines for prioritizing certain projects over others, including studies that cover a long period of time, a large geographical area, or multiple species. These are likely to be the ones that are most useful to future researchers, says Bledsoe, although she acknowledges that there are exceptions. If a biologist studies lions, a small but detailed dataset of lion behavior could be more useful than a continental-scale, long-term ecological dataset that doesn't include lions. "It really is one man's trash is another man's treasure."

Once they have produced results and publications, data are sometimes treated as tools that have outlived their usefulness.

Another factor in setting priorities for rescue is the risk of permanent loss. Information stored only on paper or on outdated media like floppy disks is especially vulnerable. Sometimes data are stored in official university department space, but just as often, they can end up in researchers' garages or handed down to their children. In their paper, Bledsoe and colleagues describe the example of University of Alberta forest ecology professor George H. La Roi. Upon La Roi's death, his children bequeathed his collected notebooks, CD-ROMs, and slide images from 35 years of studying North American boreal forests to one of his colleagues. The Living Data Project was able to match the new owner with trained students to restore and preserve this irreplaceable ecological record.

Technical advances are making data preservation easier and more reliable than ever before. Repositories are much more common than they were even a few years ago, and programs such as CoreTrustSeal, established in 2017 through an international collaboration of organizations focused on data archiving and transparency, now grant certification to repositories that are sustainably maintained and updated.

Still, technological developments don't address a lack of incentives to maintain datasets in a usable state, says Mark Westoby, a professor emeritus at Macquarie University who is not involved in the Living Data Project. "Academic careers run on publication," he says. "It's by far the most important incentive for how academics—probably government scientists as well—decide how to spend their time." Westoby recently coauthored a paper calling for a new career currency for data providers, apart from publications and journal impact scores—but such cultural changes take time, he says. While funding agencies and scientific journals are increasingly implementing data-sharing requirements, these can lead to a letter-of-the-law approach, he adds, where some data are shared to meet requirements, but not necessarily in complete datasets or in a particularly legible format.

Westoby is supportive of the Living Data Project's efforts to rescue old datasets, but notes that the group's paper sidesteps the costs of doing so as well as the motivation issue. "Having guidelines to revive, resurrect, rescue data that otherwise might be lost is all good advice. It didn't really tackle the question of how many person-hours and person-years are we talking about, and is it worth it?"

Ultimately, everyone who spoke to *The Scientist* agreed that the ideal system is one where rescue isn't necessary at all. "Data rescue is a great concept," says Roche, "but ideally what we want to do is get rid of data rescue. It would be a lot less work for people if they thought of data management and sharing from the very onset of a project, so that data are not at risk of being lost."

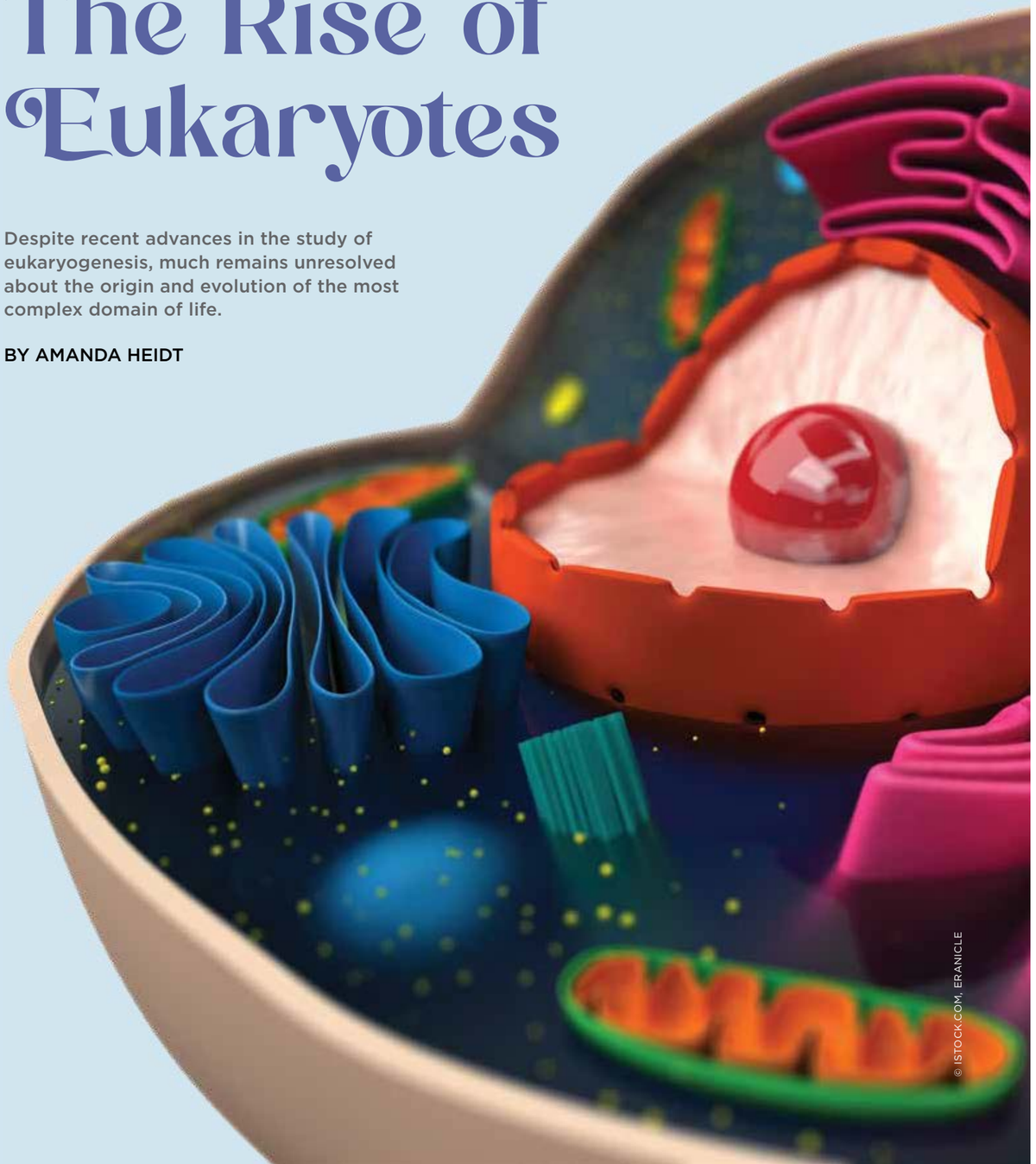
—Ian Rose

ANDRZEJ KRAUZE

The Rise of Eukaryotes

Despite recent advances in the study of eukaryogenesis, much remains unresolved about the origin and evolution of the most complex domain of life.

BY AMANDA HEIDT



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This year, University of Paris-Saclay biologist Purificación López-García embarked with colleagues on a journey into life's ancient past. The researchers traveled to the altiplanos of the northern Atacama Desert, high-altitude stretches of rocky soil and shrubbery in South America that are among the driest places in the world. Despite their inhospitable reputation, these plateaus may hold clues about the very origins of complex life. Amidst the dunes and barren mountains, there are pockets of life—warm, briny pools crusted over with colorful microbial mats of cyanobacteria and archaea stacked atop one another like crepes. Long before Earth resembled its current state, López-García says, these microbial mats "were the forests of the past," adding that scientists now use these clumps of microscopic life "as analogs of past ecosystems that certainly occurred at the time when eukaryotes first appear[ed]."

Each layer of these living mats is composed of different types of microbes that rely upon one another. At the surface, where light and oxygen are plentiful, photosynthesizing cyanobacteria dominate, while just below, heterotrophs that can persist in low-oxygen environments feed on their byproducts. Deeper down, the mats become dark and smelly, the result of the sulfate reducers and methanogens that populate these oxygen-bereft zones. Here, these partnerships become even more essential, with the castoffs of one group serving as fuel for another.

These close metabolic associations between organisms, a type of symbiosis known as syntrophy, may have prefaced the evolution of complex life by creating alliances that turned permanent over time, López-García says. In this way, individuals of different microbial species could have nested within one another to create a host with one or even several symbionts. This is exactly what scientists suspect happened to form a whole new type of cell, the eukaryote, which thrived and subsequently diversified into the macroscopic array of life we see today, including humans. So-called eukaryogenesis is not defined the same way by all researchers, but broadly, the term describes an evolutionary surge toward increasing cellular complexity between 1 and 2 billion years ago.

During this time, some of the defining characteristics of modern eukaryotic cells—the nucleus, mitochondria, cytoskeleton, cell membrane, and chloroplasts, among others—made their debut. These occurred between the first and last common ancestors of all living eukaryotes, known by their acronyms, FECA and LECA, respectively. Most of the details of these evolutionary leaps, however, remain unsettled. Researchers do not uniformly agree on which branch of life eukaryotes sprang from, which microbial players might have contributed to the process, or on the order of specific evolutionary milestones along the way. But the recent identification of the Asgard archaea, thought to be the closest living relatives to modern eukaryotes, has enlivened discussions about eukaryogenesis.

Today, at the microbial mats in the Atacama Desert and other sites throughout the world, scientists are investigating what the earliest eukaryotic cells may have looked

> [Eukaryogenesis is] arguably one of the most important events in the history of life, after the origin of life itself.
>
> —Daniel Mills
> Ludwig-Maximilians-Universität München

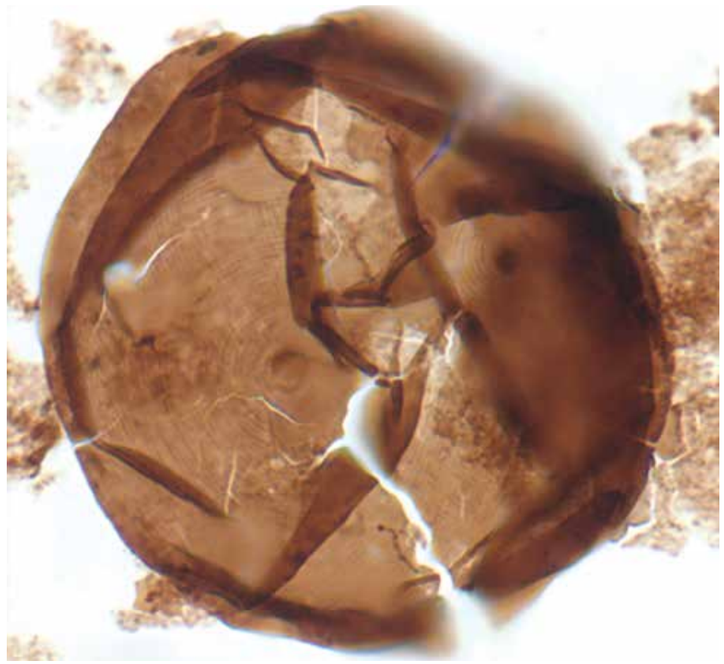
EARLY INCUBATORS: Microbial mats such as these taken from the altiplanos of South America's Atacama Desert may mimic the conditions on early Earth that gave rise to eukaryotic life.

like, the partnerships they may have struck up with other organisms, and how their molecular machinery might have functioned and evolved. Already, the discovery of the Asgards has solidified certain aspects of eukaryogenesis while raising new questions about others. "I think this is the most exciting development in biology right now. So much is being discovered and so many predictions are being met," says Daniel Mills, a geobiologist and postdoctoral researcher at Ludwig-Maximilians-Universität München who recently coauthored a paper suggesting that eukaryotes likely evolved in the absence of oxygen. Eukaryogenesis, he adds, is "arguably one of the most important events in the history of life, after the origin of life itself."

Arrival of the Asgards

After receiving her PhD in 2013, evolutionary microbiologist Anja Spang was shopping around for a postdoc. For her dissertation, Spang had studied a group of archaea called the *Thaumarchaeota* (now *Nitrososphaerota*), and during that work, she'd picked up hints that the genomes of these and other archaea contained code for genes that produce what are known as eukaryotic signature proteins, or ESPs. These proteins should not have had recognizable counterparts in archaea, and yet, there they were. Wanting to understand just what was going on, Spang joined the lab of Thijs Ettema, an evolutionary microbiologist then at Uppsala University in Sweden, and set out in search of new data.

The team extracted genomes from sediments collected during a research cruise to a deep-sea vent site called Loki's Castle located more than 2,300 meters below the surface of the Arctic Ocean, between Greenland and Norway. Ettema told *The New York Times* that the initial sample amounted to less than a teaspoonful of deep-sea muck. But almost immediately, software responsible for annotating and analyzing the genetic material began to return odd results—it flagged ESP homologs for actin, a distinctly eukaryotic protein that gives cells their shape, in a genome that was otherwise clearly archaeal.[1] The microbes turned out to be members of a new group that Spang and the team named the *Lokiarchaeota* when they published their findings in *Nature* in 2015.[2] In the years that followed, the team continued to flesh out this branch of the archaeal family tree, leading to the establishment of the Asgard superphylum,[3] which in addition to *Lokiar-*

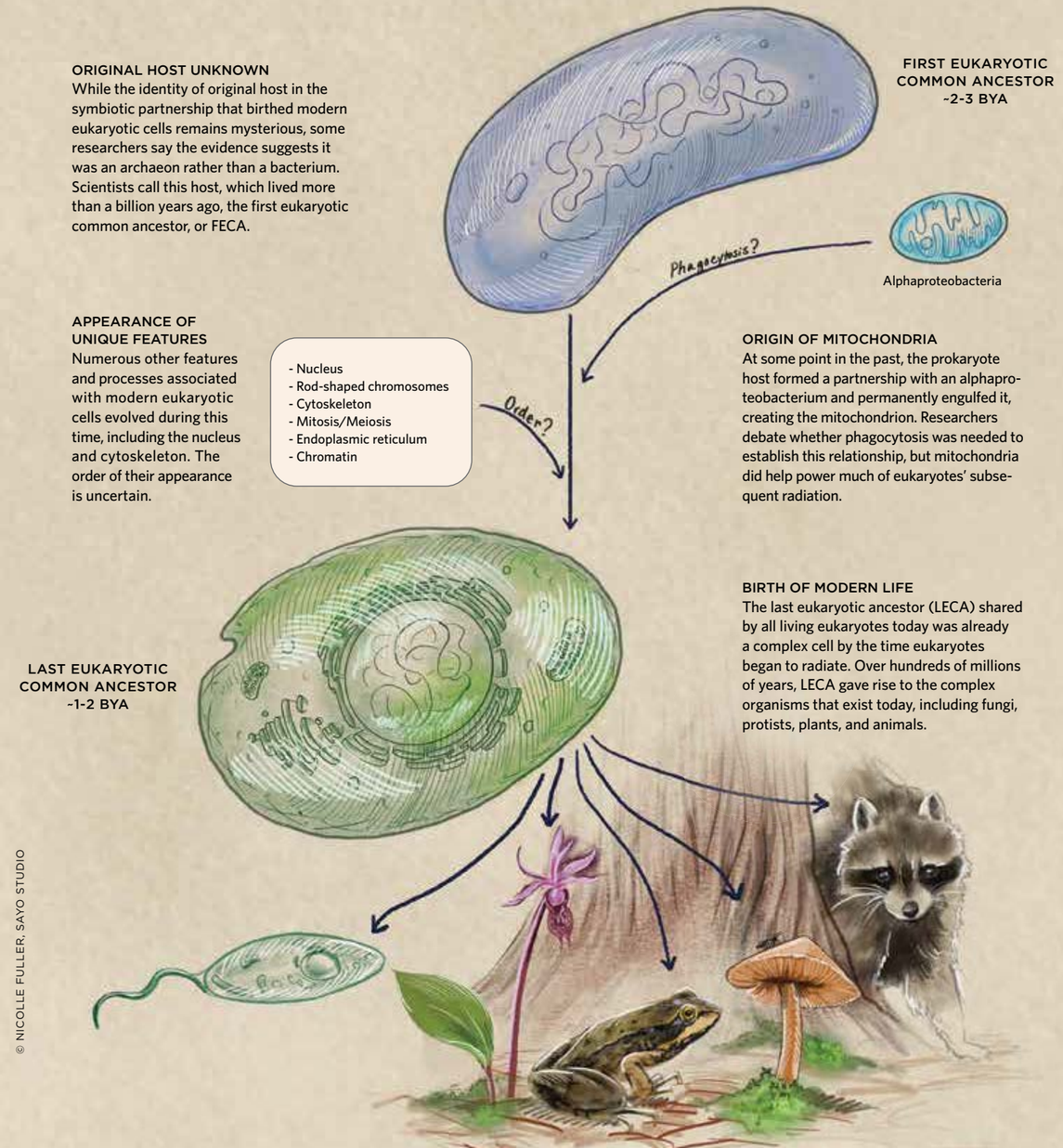


MINISCULE FOSSILS: While metagenomics have rapidly advanced the study of eukaryogenesis, the study of microfossils such as this 750-million-year-old *Valeria lophostriata* may also help shed light on when certain eukaryotic features first appeared.

DIVERSITY, ECOLOGY AND EVOLUTION OF MICROBES (DEEM)/PURIFICACIÓN LÓPEZ-GARCÍA; COURTESY OF SUSSANAH PORTER

THE PATH TO COMPLEXITY

Eukaryogenesis is broadly defined as the evolutionary path taken by increasingly complex lifeforms as they diverged from the simpler prokaryotes that dominated the early part of Earth's biological history. The functional period of eukaryogenesis started just prior to the symbiosis between two prokaryotes and ended when the last common ancestor of modern eukaryotes arose. During this time, many of the most recognizable eukaryotic features appeared, including organelles such as mitochondria, nuclei, and chloroplasts, as well as cellular processes such as phagocytosis. The ordering of these events in time remains unclear.



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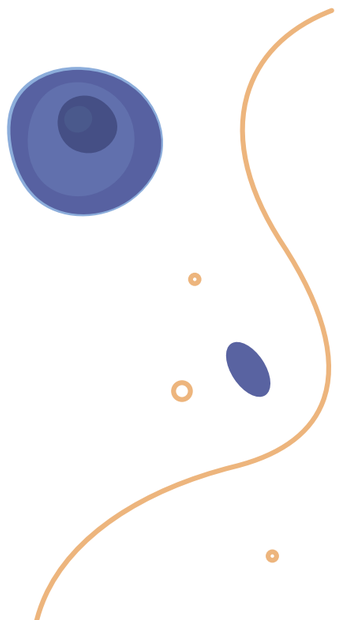
People were already arguing for a two-domain system before the Asgards were discovered, but then once the Asgards were described, it gave even more evidence.

—Andrew Roger, Dalhousie University



chaeota includes nods to other Norse gods, including the *Thor*-, *Odin*-, and *Heimdallarchaeota*.

Researchers have since identified other ESPs in these groups, including homologs of proteins involved in everything from ubiquitin signaling to gamete fusion. That ESPs are so common among Asgards suggests that these microbes represent the closest living prokaryotic relatives to modern eukaryotes and that modern eukaryotes may well have inherited aspects of their molecular machinery from archaea. Indeed, most scientists now argue that an ancient Asgard or another archaeon, and not a bacterium or proto-eukaryote as many previously assumed, likely served as the first host in the evolutionary process that ultimately resulted in a new type of cell.

In 2019, researchers successfully cultured an Asgard archaeon for the first time, allowing scientists to dive deeper into their biology.[4] Using microscopy, Hiroyuki Imachi of the Japan Agency for Marine-Earth Science and Technology and colleagues found that the cultured species, for which they proposed the name *Candidatus Prometheoarchaeum syntrophicum*, is small and extremely slow-growing, dividing only every two to three weeks; some microbes can double in as little as a few minutes or hours. In addition, they found that *Ca. P. syntrophicum* lives in close association with another archaeon called *Methanogenium*. *Ca. P. syntrophicum* gets its energy by digesting amino acids and peptides for their nitrogen, and in turn, *Methanogenium* uses the hydrogen produced during that process to create its own fuel and at the same time reduce environmental hydrogen, which can induce cellular stress. This partnership confirms that Asgards engage in the type of relationships that researchers suspect gave rise to eukaryotes.

Hints of such a syntrophic relationship had been gleaned from other archaeal genomes, says Spang, who now oversees her own research group at the Royal Netherlands Institute for Sea Research, but *Ca. P. syntrophicum* provides tangible evidence. "I was really happy when I heard" of the preprint that first described the organism and its syntrophic lifestyle, she says. "[It] verified that at least the metabolic predictions for the Asgards were making sense with actual experimental work."

Lots of hypotheses, few answers

These early observations precipitated a flood of new research, with hundreds of papers published as preprints on *bioRxiv* touching on Asgards and eukaryogenesis in the last several years. The most immediate effect of the discovery of Asgards was a shift in support from a three-domain tree of life that included eukaryotes, prokaryotes, and archaea to a two-domain model, often called the eocyte hypothesis, that lumps archaea and eukaryotes together. (See illustration on page 34.)

In the three-domain model, eukaryotes belong to a separate branch that shares a common ancestor with archaea. But phylogenetic analyses suggest that complex cells emerged from within the archaea. This results in two primary domains—bacteria and archaea—with eukaryotes being nested within archaea. "People were already arguing for a two-domain system before the Asgards were discovered, but then once the Asgards were described, it gave even more evidence," says Andrew Roger, a molecular biologist at Dalhousie University in Nova Scotia. He adds that the two-domain hypothesis also "supports that the host during eukaryogenesis was an archaeon" and not a type of proto-eukaryote that formed a distinct lineage.

Researchers who spoke to *The Scientist* say that many scientists have rallied behind the idea that the first eukaryotes evolved out of a syntrophy between an archaeal host and bacteria that somehow found their way inside to become the organelles, such as nuclei and mitochondria, that distinguish eukaryotes. The details of these relationships remain murky, but mitochondria provide the most tantalizing clues to their origin story. "There's DNA in mitochondria that we can somewhat clearly connect or trace back to alphaproteobacteria," says Laura Eme, an evolutionary microbiologist at France's National Centre for Scientific Research (CNRS). "Even if we don't know exactly which lineage, we have a smoking gun."

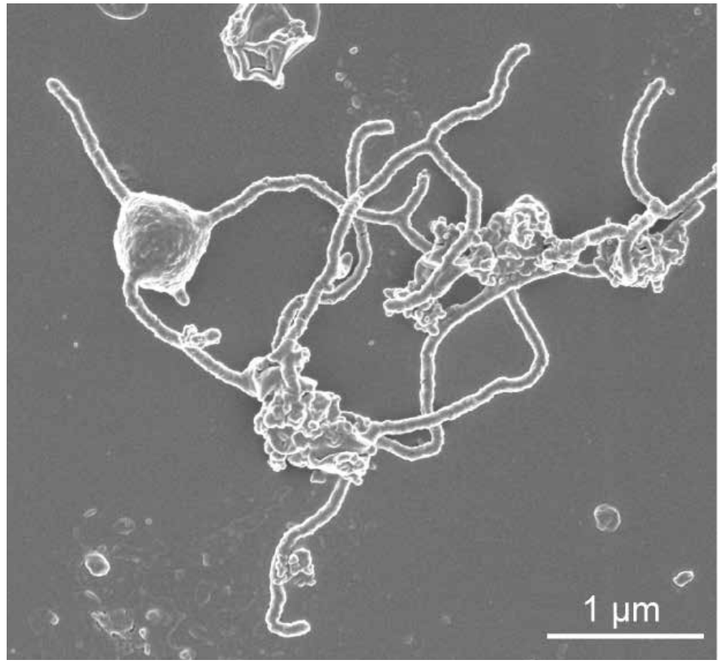
There are contrasting hypotheses as to how the alphaproteobacterium would have gotten inside an archaeal host, however. In the eukaryogenesis version of the chicken-and-egg conundrum, scientists go back and forth on whether mitochondria would have been necessary to power the energetically expensive process of phagocytosis, or whether phagocytosis would have had to arise first as the means of ingesting the symbiotic partner. An oscillation between "mito-early" and "mito-late" hypotheses appears frequently in the literature, but intriguingly, there were no known examples of phagocytosis in prokaryotes until very recently, when researchers identified a phagocytosis-like process of engulfment in a bacterium. "[M]any people were saying it is impossible to have the ancestor of mitochondria incorporated in any cell because phagocytosis is not known in the prokaryotic world," says Eme. "Well, now we know that phagocytosis exists in bacteria, at least."

Moreover, initial observations of the Asgards point to other mechanisms of engulfment. When scientists first cultured *Ca. P. syntrophicum*, they immediately noticed a series of thin projections coming off of the microbes—extensions of their membrane system called blebs. This observation suggested that these blebs might be able to surround an external entity—perhaps with the help of those actin homologs—and fuse together, trapping the foreign body inside. The phagocytosis conundrum "is much less of a problem now," Eme tells *The Scientist*.

When it comes to the nucleus, what López-García calls "the typical diagnostic eukaryotic feature," the picture is much less clear. Hypotheses of its origin run the gamut from a bacterial endosymbiont within an amoeboid host to the remnants of a giant virus. (See "Sidebar" on page 35.) In the 1990s, López-García proposed the Syntrophy hypothesis for the origin of eukaryotes, which posited a three-party metabolic symbiosis between two bacteria and an archaeon. She maintains that this hypothesis is the only one that explains not only the origins of the nucleus, but also the so-called lipid divide, another unsettled aspect of eukaryogenesis in which the lipids that make up the cell membranes of eukaryotes are more similar to those in bacteria than to those in archaea.[5]

A couple of years ago, López-García and her Paris-Saclay colleague David Moreira, also affiliated with the CNRS, updated the hypothesis to reflect the discovery of Asgards, but rather than place an archaeon as the original host, they propose than an archaeon—specifically a hydrogen-producing, Asgard-like archaeon—was the original nucleus.[6] The host, they suggest, was likely a deltaproteobacterium, and the ancestor of mitochondria an alphaproteobacterium. This idea is supported, they say, by the fact that most genes in modern eukaryotes are actually bacterial, and not archaeal, in origin, and that eukaryotic membranes are made up of phospholipids that more closely resemble bacterial ones. "Our model is one potential model—it may be wrong, [or it] may be right—but the others don't explain these discrepancies," López-García says. "And at some point, I think they should."

Michelle Leger, a postdoctoral researcher and evolutionary microbiologist at the Institute of Evolutionary Biology in Barcelona, is currently scouring the genomes of extant archaeal species to support or refute the many hypotheses floating around. With respect to the Syntrophy hypothesis, for example, "if I were to imagine that there was the deltaproteobacteria in that relationship as well, I would expect a similarly clear [genomic] signal" to that of the alphaproteobacteria in the mitochondrial genome, Leger tells *The Scientist*. She hasn't found such a signal yet, but she says she thinks the evidence does support an archaeal origin for the nucleus. Although archaeal genes make up a small fraction of the nuclear genome, the genes that play roles in highly conserved processes within the nucleus itself, such

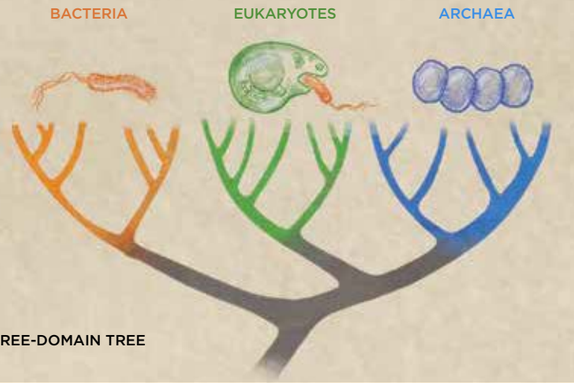


HIROYUKI IMACHI, MASARU K. NOBU, AND JAMSTEC

HELLO COUSIN: Researchers first identified Asgard archaea, thought to be the closest living prokaryotic relatives to modern eukaryotes, from metagenomic data in 2015. A few years later, the first Asgard was cultured, revealing unique aspects of its biology.

FROM THREE DOMAINS TO TWO

The question of where exactly eukaryotes branch on the tree of life has been debated by scientists for decades. But the discovery of the Asgard archaea—the closest prokaryotic relatives to modern eukaryotes—has shifted most researchers away from a three-domain tree in which eukaryotes are a distinct lineage and toward a two-domain tree, in which eukaryotes emerged from within the archaea as a secondary domain.



THREE-DOMAIN TREE

TWO-DOMAIN TREE

as DNA replication and transcription, are largely archaeal. So "it makes sense" that the nucleus developed from an archaeon, Leger says. "But it's not very clear what other partners might have been involved."

The next big frontier

Even as the number of sequenced archaeal and bacterial genomes continues to increase, offering new clues about the relationship between these microbes and the rise of early eukaryotic cells, many researchers tell *The Scientist* it's entirely possible that some questions will never be fully answered. Too much time has passed since eukaryotes first appeared on the evolutionary scene, and too much DNA has been scrambled between too many groups, for scientists to piece everything together. But that hasn't stopped them from trying.

Eme tells *The Scientist* that the "next big frontier" will be functional studies in modern eukaryotes to yield clues about how individual genes and proteins may have behaved in their early ancestors. While there was only a single Asgard genome a few years ago, today there are hundreds, and researchers are mining them for details. "Now we have a clear idea of which genes in eukaryotes have been inherited from Asgard archaea, and there's a lot of novelty here," Eme says. "But what we don't know, and that's really important, is what these genes did or are doing in Asgard currently."

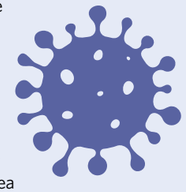
In 2020, researchers synthesized suspected homologs of eukaryotic actin proteins encoded in Asgard genomes. Injected into rabbit cells, these proteins bound to eukaryotic actins and performed similar functions, including aiding the flow of calcium across cell membranes.[7] The findings suggest that a calcium-controlled actin cytoskeleton likely existed in Asgards prior to the emergence of eukaryotes. In another study, researchers attempted to resolve the lipid debate by expressing archaeal phospholipids in *E. coli*, and found that the bacteria were able to successfully incorporate as much as 30 percent of the archaeal lipids into their cell membranes.[8] The study doesn't fully reconcile whether eukaryotes would have been able to transition their membranes from bacterial to archaeal lipids—López-García notes that bacteria with membranes composed of more than 30 percent archaeal lipids begin to die—"but it does lay the groundwork for future research," Eme says.

Additional clues could come from the study of microfossils, microscopic impressions of early cells embedded in rock, says University of California, Santa Barbara, paleontologist Susannah Porter. When metagenomic sequencing came to the fore, it seemed as though fossils fell out of favor, she says, but many phylogenetic trees rely on a methodology called a molecular clock that uses fossils to anchor analyses in time. In addition, the fossils themselves can be useful, allowing scientists to determine when certain external features first appeared, adds Porter, who is currently interrogating such specimens to order certain events of early eukaryote evolution. "We do have a fossil record back 2 billion to 1 billion years, but I don't think it's been taken advantage of or leveraged to its full extent," she says. "Maybe we could actually use these characteristics of the fossil record to be able to piece together eukaryogenesis."

Meanwhile, other researchers are devising alternate methods for timing the events of eukaryogenesis to complement that fossil evidence. For example, Berend Snel, a computational biologist at Utrecht University in the Netherlands, recently used gene duplications to correlate the lengths of branches on phylogenetic trees with time—the assumption being that the number of duplication events increases with time.[9] That assumption was challenged by some, and even Snel admits that "it may not be perfect," but breaking the story of eukaryogenesis into more manageable chunks may help resolve many of these unanswered questions, he says. "What I'm arguing for is that it's a lot of little, small stories, but if people would integrate these small stories in the right way, there should be a tapestry that ultimately weaves a real story."

Leger agrees that our understanding of eukaryogenesis is likely to advance with baby steps. "Part of the nature of these deep evolutionary questions is that we will never know, we will never have a clear proof of some of the hypotheses that we're trying to develop," she says. "But we can keep refining our ideas." ■

VIRUSES OF THE ASGARD



While much about the origin of the nucleus is speculative, one hypothesis suggests that the nucleus of modern eukaryotes may have resulted from a partnership between a prokaryotic host and a virus. This idea was first suggested in a pair of papers published back-to-back in 2001 after two researchers independently arrived at the same conclusion, and both groups recently published updates to their viral origin hypotheses following the field-rocking discovery of the Asgard archaea (*Front Microbiol*, 11:571831, 2020; *Virus Res*, 289:198168, 2020).

At the turn of the 21st century, Masaharu Takemura, then a molecular biologist at the Nagoya University School of Medicine in Japan, noticed that one group of viruses, the poxviruses, had DNA polymerases that were extremely similar to those found in eukaryotes, and that poxviruses replicate inside their hosts by creating self-contained compartments. Meanwhile, Philip Bell, the head of research for the biotechnology company MicroBioGen, was similarly puzzled by the differences between eukaryotes and the bacteria that led to organelles such as mitochondria. Eukaryotic chromosomes are linear, for example, while bacterial ones are circular. Many features of the nucleus just didn't support a bacterial origin.

Since that time, researchers have identified the so-called giant viruses, first described in 2003. These viruses are much larger than most, with fittingly massive genomes, and they've since been found to harbor genes associated with various metabolic processes. Now, Takemura, Bell, and others say that a giant virus could have been the original nucleus. Giant viruses replicate within complex compartments that look very similar to modern nuclei—they're large and include both inner and outer membranes—and also carry versions of genes that produce proteins involved in essential host cell processes.

The idea that the nucleus could have been a virus has been a tough sell, however. According to Purificación López-García, a biologist at the University of Paris-Saclay, "there is no structural evidence" to support it. Michelle Leger, an evolutionary microbiologist at the Institute of Evolutionary Biology in Barcelona, agrees that the hypothesis is not supported by existing data, which she argues more clearly point to an archaeon as the organism that became the eukaryotic nucleus.

But Valerie De Anda, a microbiologist at the University of Texas at Austin Marine Science Institute who studies early prokaryote metabolism, isn't dissuaded by the current lack of evidence from the idea that a virus may well be the source of the eukaryotic nucleus. She and her colleagues are currently looking for mRNA-capping genes involved in transcription and translation that were suggested by Bell to have been derived from a long-ago "first eukaryotic nuclear ancestor" (*Nat Microbiol*, 7:953–61, 2022).

"People don't take seriously great ideas right at the beginning . . . and then it turns out to be true," De Anda says.

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Micro Manipulators

Biologists are learning how intracellular bacteria hijack host cells—and they're unveiling secrets of human cell biology in the process.

BY CATHERINE OFFORD

As a grad student in cell biology, Shaeri Mukherjee was always on the lookout for new ways to fiddle with cells' internal structures. It was the early 2000s, and Mukherjee was working in Dennis Shields's lab at Albert Einstein College of Medicine, studying how cells organize the internal transport of proteins and other cargo. She was particularly interested in the Golgi apparatus, a cluster of membrane-bound compartments that help coordinate this trafficking, and spent much of her time manipulating the organelle's activity to try to better understand how it works. Genetics methods could slow down or alter the organelle's structure in days; certain pharmacological agents made it disintegrate in less than half an hour. But in 2008, Mukherjee stumbled across a new and much faster way to cause intracellular mayhem.

The technique came from a paper by Craig Roy at Yale University School of Medicine. Roy's team had found that "this protein called AnkX, when microinjected into cells—even at picomole levels—could cause the entire Golgi to fragment in, like, five minutes," Mukherjee says. Remarkably, AnkX hadn't been made by cell biologists or a pharmaceutical company. Rather, it was produced by a tiny intracellular bacterium known as *Legionella pneumophila*, the pathogen behind a serious lung infection called Legionnaire's disease.[1] For Mukherjee, the paper was a revelation: not only did it identify the fastest way yet to target Golgi biology, it suggested that scientists could use intracellular bacteria "as a lens to understand basic processes inside the cell."

Intrigued by this powerful little microbe, Mukherjee applied for a postdoc in Roy's lab, where she would study in detail how *Legionella* attacks human cells from the inside. She learned that researchers had identified more than 300 *Legionella* peptides that mimic host proteins or otherwise hijack existing cellular pathways to the bacterium's advantage. And she gained a new appreciation for the myriad other types of intracellular bacteria, a diverse group that includes many medically significant pathogens such as *Salmonella*, *Listeria,* and *Chlamydia*, as well as the causative agents of tuberculosis and leprosy. (See Bug Box on page 42).

These microbes all enjoy at least part of their lives shielded from the onslaught of white blood cells, antibodies, and other immune defenses that the body launches against pathogens that live outside of host cells. As a trade-off, they have had to come up with ways to bypass a cell's internal immune system, navigate the complicated, busy environment of the cell cytoplasm, and ultimately escape that environment to infect other cells—becoming tiny maestros of manipulation in the process.

Research on how intracellular bacteria take control of their hosts is not only informing scientists about how these microbes cause disease, but revealing secrets of mammalian biology, says Mukherjee, who now heads up a lab at the University of California (UC), San Francisco. These bugs have a knack for pinpointing critical cell functions, she says, adding fondly that *Legionella* is continuing to help her explore how eukaryotic cells work. "It's an excellent cell biologist."

Get the gear

In the late 1980s, bacteriologist Daniel Portnoy visited actin expert Lewis Tilney at the University of Pennsylvania to discuss a new research idea. Or as Tilney later relayed the experience to the *Journal of Cell Biology:* "Portnoy crashed a department picnic and insisted I look at his damn *Listeria*."

By then it was known that *Listeria* infects cells such as macrophages—motile human immune cells that engulf pathogens and cellular debris—by being taken up into vacuoles and breaking out of those vacuoles into the cytoplasm. Researchers had also described the bacteria quickly spreading among neighboring cells. But Portnoy, who joined UPenn's medical school in 1988, had found he could block this cell-to-cell spread by chemically inhibiting the polymerization of actin, a protein that cells use to build an intracellular cytoskeleton to aid cell motility, division, and other important processes. Intrigued, Tilney took on the project, and together the pair showed that after breaking out of the vacuole, *Listeria* managed, somehow, to construct its own tail-like structure out of actin filaments.

> Studying bacterial interactions with actin is still throwing up new mysteries.

The resulting little *Listeria* "comet," as the pair called it, used its new actin motility to whiz around the cytoplasm and eventually hurl itself at the plasma membrane, causing a protrusion that extended into a neighboring macrophage. This protrusion got engulfed by the recipient cell to form a vacuole, and *Listeria* then simply had to muscle out of its new compartment to complete infection.[2] (See illustration on opposite page.) Portnoy and Tilney described the whole process in a 1989 paper, observing that their findings "should be important to those concerned with stages in the cell biology of infection by parasites and . . . exciting to cell biologists who want to know how actin filaments become organized in cells."

Many other scientists have since joined in the study of *Listeria*'s actin co-option. Pascale Cossart, a microbiologist who was studying *Listeria* virulence at the Pasteur Institute in the 1990s, soon identified a bacterial protein, ActA, that the microbes require to build their tails.3 Cell biologist Matthew Welch, then at UC San Francisco, and colleagues next isolated a host cell protein complex, Arp2/3, that is also necessary for *Listeria* to become motile. The team found that *Listeria*'s ActA was recruiting the Arp2/3 complex to the bacterial cell surface, and this was what was initiating actin polymerization—confirming that the bacterium was co-opting the cell's own machinery and raw materials for personal use.[4] The work hinted that eukaryotes might possess their own Arp2/3-activating proteins, and sure enough, researchers have since described a whole family of host actin nucleation–promoting factors, which *Listeria*'s ActA successfully mimics.

A number of other intracellular bacterial taxa—including *Shigella*, *Rickettsia*, *Mycobacterium*, and *Burkholderia*—have been observed constructing their own actin tails, often by hijacking Arp2/3. And while many take a *Listeria*-like approach to infecting new hosts (shoving into neighbors and being taken into vacuoles), it's not the only way. Welch, now at UC Berkeley, and grad student Nora Kostow recently used live cell imaging and other technologies to study *Burkholderia thailandensis*, which spreads by causing neighboring cells to fuse. The bacteria essentially expand "the available environment for them to grow," says Welch. "They can do that repeatedly, so you can get hundreds of cells fusing together in some cases." He and Kostow showed that actin-powered *B. thailandensis* achieves this spread by pushing on the plasma membrane to create protrusions that, rather than create vacuoles in a neighboring cell, cause those two cells to become one.[5] This melding appears to be dependent on specific proteins secreted by the bacterium as it forms the protrusions—an insight that could help cell biologists understand cell fusion more generally, the authors write in their paper.

Studying variations on these bacterial interactions with actin is still throwing up new mysteries. While investigating *Mycobacterium marinum*, a close relative of *M. tuberculosis*, Welch and postdoc Norbert Hill recently found that microbial proteins could confer actin motility not just on the bacterium, but also on another sort of intracellular object: lipid= droplets.[6] It's not yet clear how this lipid movement relates to *Mycobacterium*'s presence, "but it's tempting to speculate that it could happen during infection," either to the bacterium's benefit or as some cellular response to infection, Welch says. Several lines of research suggest that *Mycobacterium* species might use lipid droplets as a source of chemical energy, among other things, he adds, so perhaps it's in the bacterium's interest to have those droplets whizzing around too.

Master the membrane

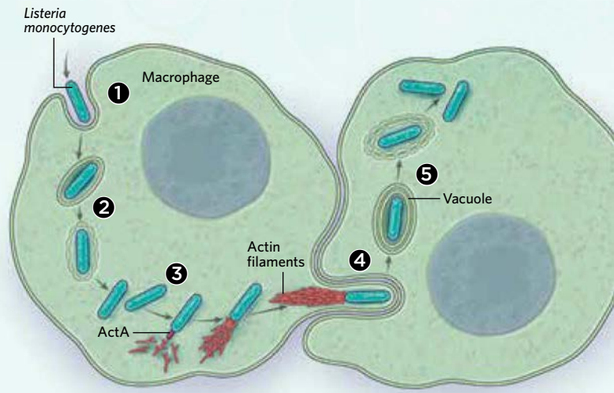
The cytoplasm isn't for everyone. While bugs such as *Listeria* gain access to building materials like actin, they also have to dodge the cell's immune defenses and survive a chemical environment they have little control over. Some bacteria avoid these inconveniences by instead occupying organelle-like compartments that separate them from the rest of the cell. These microbes can and do still interact with actin—some secrete proteins that cause rearrangements in the cell cytoskeleton to help take up the bacterium from outside, or to form highway-like protrusions into other cells, for example. But many have also mastered a different sort of target that allows them to wield control over the rest of the cell without venturing into the cytoplasm: namely, lipid membranes.

It was *Legionella*'s ability to take over and even mimic intracellular membranes that occupied Mukherjee during her postdoc at Yale. It turned out that AnkX, the same microbial protein that had caused the Golgi apparatus to disintegrate, was part of a larger scheme to coerce the host into preparing a bacteria-friendly compartment. *Legionella* was using AnkX, among other peptides, to target host enzymes known as Rab GTPases, which sit on the surface of organelles such as the endoplasmic reticulum and the Golgi apparatus and regulate the trafficking of protein cargo around the cell. (See illustration on page 40.)

Specifically, Mukherjee, Roy, and colleagues showed that by making an unusual type of modification known as phos-

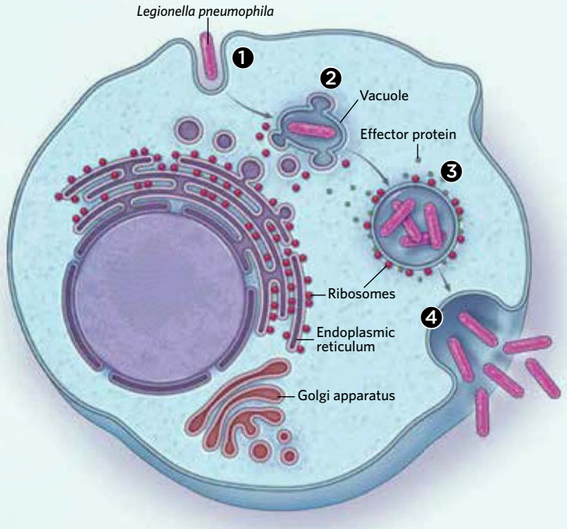
ACTIN TAILS

Some intracellular bacteria use the host cell's actin supplies to build their own transport system. The foodborne pathogen *Listeria monocytogenes* infects immune cells called macrophages by being taken up into a vacuole ❶ before entering the cytoplasm where it lives and replicates ❷. There, it uses a protein called ActA to recruit the host cell's actin polymerization machinery to construct a tail of actin filaments behind it ❸. This process gives the bacterium a means to propel itself around and lets it push on the host cell membrane, forming protrusions into neighboring cells ❹. Those neighbors take up these protrusions as vacuoles, from which *Listeria* escapes to access the cytoplasm and begin the cycle again ❺.



REROUTING MEMBRANE TRAFFIC

Some intracellular bacteria, such as *Legionella pneumophila*, inhabit membrane-bound compartments inside host cells ❶. Once there, the microbes typically interact with host membranes and secrete so-called effector proteins that help the microbes wield control over them ❷. *Legionella* in particular interacts with the Golgi apparatus and the endoplasmic reticulum, pilfering some of the organelles' proteins and rerouting their vesicular traffic. Later, the newly formed membranes become studded with ribosomes ❸ that may help the bacterium make certain host proteins—or could simply be a byproduct of the membrane's ER-like identity. *Legionella* replicates inside this compartment before bursting out of the cell ❹.



phocholination to one of the cell's Rab proteins, *Legionella* was able to cause a "massive and quick collapse of the [host] trafficking pathway," Mukherjee says.[7] This was in addition to its recruitment of that same Rab protein through a separate mechanism to the surface of its own intracellular compartment, converting its hideaway into something resembling the endoplasmic reticulum. Labs including Roy's have since showed how this membrane conversion is part of a process that preps *Legionella*'s compartment for bacterial replication. Similar membrane-copying or -hijacking processes have been described in other microbes, too. The sexually transmitted pathogen *Chlamydia trachomatis*, for example, conspires to reorganize Golgi membranes around its intracellular compartment, while rerouting the organelle's vesicles to itself as a source of lipids.

Other intracellular bugs have found different ways to mess with a cell's membranes. The single-cell parasite *Toxoplasma gondii*, which the Centers for Disease Control and Prevention estimates currently infects more than 40 million people in the US alone, survives in an intracellular vacuole, from which it deploys proteins to subvert host cell function. Researchers observed decades ago that the *T. gondii* vacuoles, like several types of bacterial compartments, often become surrounded by mitochondria—a phenomenon that's now thought to be related to a cellular anti-pathogen response, says Lena Pernas, a parasitologist-turned-cell-biologist at the Max Planck Institute for the Biology of Ageing in Cologne, Germany.

Pernas's team recently found that *T. gondii* is able to subvert and exploit this mitochondrial mobbing by secreting a protein that causes the organelles to cast off the outer of their two membranes.[8] "We're not sure exactly how that happens . . . and this is the subject of ongoing work in the lab," Pernas says. But her team's experiments do suggest that the shedding behavior is a natural mitochondrial response to outer membrane stress that can occur even in uninfected cells, and that *T. gondii* has hijacked this process—perhaps by mimicking a host protein that normally triggers the reaction. Whether other pathogens exploit this or related pathways remains to be seen, although some researchers have noted that at least one of the mitochondrial proteins mentioned in Pernas's study also seems to be targeted by viruses such as SARS-CoV-2.

It wouldn't be surprising to discover new types of membrane manipulation, Mukherjee says, adding that the behavior offers a versatile way to exert influence over everything from cell division to the intracellular positioning of organelles and even pathogens themselves. "That's why various bacteria target various membranes inside the cell."

> "If you slow down the leading ribosome, the ribosome after it goes and hits it . . . just like a pile-up in a freeway."
>
> —Shaeri Mukherjee, UC San Francisco

Altering hosts' destiny

Intracellular bacteria and other parasites carry their own protein-making machinery, so it might sometimes make sense for them to shut down or pause most of the host's protein production. "The host will most likely make proteins that are deleterious for the bacteria," explains Mukherjee, who recently showed that *Legionella* is able to block protein synthesis by targeting a host peptide involved in protein folding.[9] On the other hand, an intracellular bacterium doesn't want to kill off its host before it's ready to move on, or to miss out on the opportunity to get the host to perform energy-intensive tasks on its behalf.

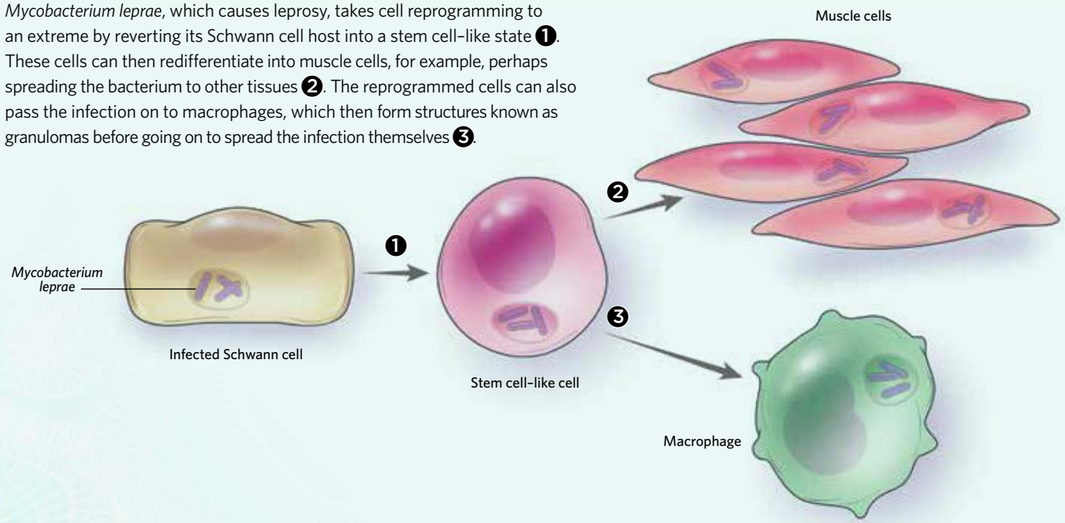
Faced with these trade-offs, some bacteria launch a well-choreographed effort to manipulate what a cell can and can't make at different times during infection. For example, Mukherjee and colleagues recently identified a tRNA-mimicking toxin secreted by *Legionella* that stalls the movement of host ribosomes along RNA, causing collisions. "If you slow down the leading ribosome, the ribosome after it goes and hits it . . . just like a pile-up in a freeway." This sets off a cascade of events in the cell, the researchers found, including large-scale alterations to gene expression that allow just a few key transcripts to bypass the traffic jam and get translated into proteins. The response leads to controlled cell death, which is good news for *Legionella*: the host breaks open, releasing the bacteria to go on to infect other cells.[10] It's yet another case of a microbe teaching biologists about how cells work, says Mukherjee, who described the research in a preprint on bioRxiv—the team only discovered the genetic response to ribosome collisions thanks to *Legionella*'s ability to target that pathway.

Some intracellular bacteria appear to take a different tack: instead of prompting cells to self-destruct, they can cause changes in their host's cell type. The University of Edinburgh's Anura Rambukkana has studied reprogramming by *Mycobacterium leprae*, which causes leprosy. These bacteria infect Schwann cells, glial cells that surround neurons and help develop and repair peripheral nerves. Infection typically triggers widespread neurological injury and, eventually, loss of sensitivity to pain or touch in affected limbs. But in 2013, Rambukkana and colleagues reported results from a series of in vitro and mouse experiments that suggested the bacterium was first hijacking their hosts' gene expression, apparently reprogramming Schwann cells back into a stem-cell-like state.[11] (See illustration below.)

This tweaking of gene expression seems to aid *M. leprae*'s spread in at least two ways. First, altered cells can go on to differentiate into other cell types, including muscle cells, says Rambukkana, potentially seeding the bacteria in these other tissues. Second, reprogrammed cells attract macrophages, which themselves can pick up the infection and spread it to other tissues. The team is currently working to understand more about the mechanisms underlying this cellular rewiring, as well as exploring potential therapeutic applications of the phenomenon. For example, studying the factors the bacteria use to reprogram cell state might offer new techniques for regenerative medicine, Rambukkana says. The team is currently testing some of these principles in nine-banded armadillos (*Dasypus novemcinctus*)—not an ideal model for human biology, but one of *M. leprae*'s few natural hosts other than humans.

REPROGRAMMING THE HOST

Mycobacterium leprae, which causes leprosy, takes cell reprogramming to an extreme by reverting its Schwann cell host into a stem cell–like state ❶. These cells can then redifferentiate into muscle cells, for example, perhaps spreading the bacterium to other tissues ❷. The reprogrammed cells can also pass the infection on to macrophages, which then form structures known as granulomas before going on to spread the infection themselves ❸.



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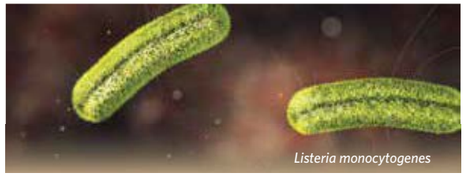
Cossart, now a visiting scientist at EMBL Heidelberg in Germany, calls the work on Schwann cell reprogramming a "very interesting" line of research and notes that these kinds of findings highlight just how varied intracellular pathogens are in their attempts to subvert cell function. "There are different types of result with different types of pathogens," she notes, adding that in addition to studying species differences, researchers should dig into variation among different strains of bacteria and under different conditions if they want to understand the biological consequences of infection. It's only relatively recently, for example, that scientists have started to consider intracellular bacteria's interactions—direct or indirect—with the microbiome of the organisms they infect, a research area that Cossart says deserves more attention.

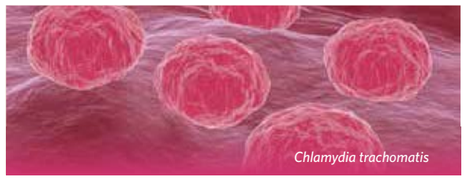
In many cases, though, the wider impact of research using intracellular microbes may be hard to estimate in advance, says Mukherjee, noting that tools such as CRISPR grew out of basic research—in that case, on a system bacteria use to defend themselves from infection by viruses. "We are basic scientists, we want to study fundamental processes," she says. That work can "have an impact down the road." ■

> Some bacteria launch a well-choreographed effort to manipulate what a cell can and can't make at different times during infection.

BUG BOX

Intracellular bacteria vary considerably in how dependent they are on their hosts. So-called **facultative** intracellular bacteria such as *Listeria monocytogenes* and *Legionella pneumophila* do not need to be inside a host cell to reproduce. By contrast, **obligate** intracellular bacteria such as *Chlamydia trachomatis* and *Mycobacterium leprae* do, and this trait makes them a challenge to culture and study in the lab. Obligate intracellular pathogens will often have reduced genomes compared to the facultative variety, a genetic tendency that is reflective of their more limited lifestyle.


Listeria monocytogenes


Chlamydia trachomatis

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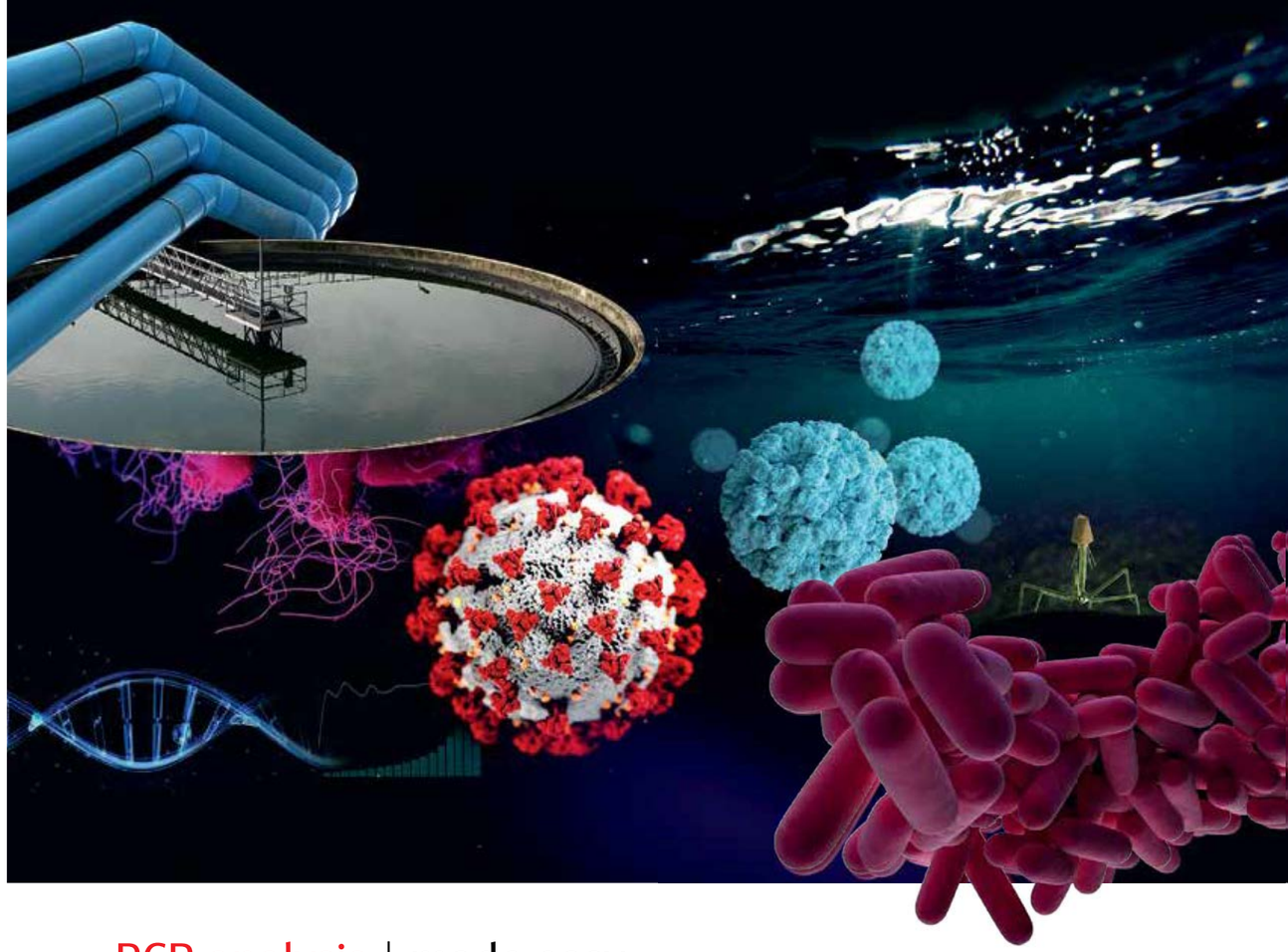

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Protecting Pregnancy

Carefully calibrated immunological changes in mothers are critical for a healthy pregnancy. Understanding how the body balances immune tolerance and protection from infection will allow us to improve pregnancy outcomes.

BY TOBIAS R. KOLLMANN, ARNAUD MARCHANT, AND SING SING WAY

We first met Kate and her husband Adam in 2016 when she was 26 weeks pregnant and in labor. Within hours, she gave birth to twins, James and Fraser. The newborns weighed in at just around 1 kilogram each. "Seeing them—so tiny and fragile, but alive—flooded my body with sheer relief," Kate wrote of the experience in an email. "I touched their little hands before they were wheeled away into the Neonatal Intensive Care Unit (NICU)."

Kate pumped breast milk that was fed to the newborns to help them grow. But before long, James took a turn for the worse, developing a devastating intestinal condition known as necrotizing enterocolitis. Surgeons needed to cut a tiny hole in his belly to allow gas to be released from his bowel, but bacteria from his damaged colon had spread through his body. After just three and a half weeks, we had to tell Kate and Adam that James wouldn't survive, and that it was time to say goodbye.

Unfortunately, James's story is not unique. Millions of little lives are lost around the time of birth every year. Some babies don't make it as long as James did, dying even before delivery. Every 16 seconds a baby is stillborn somewhere in the world; this amounts to more than 2 million stillborn babies globally every year. Of babies that are born alive, shockingly high numbers of them are born too early. A baby is born prematurely every two seconds, resulting in 15 million preterm babies every year. Sadly, 1 million of these preterm babies die every year due to direct complications from preterm birth, and another 800,000 of them die from infections associated with preterm birth. Preterm birth remains the leading cause of death for babies and young children the world over.

Preterm birth is also the leading cause of childhood disability, with 1.3 million preemies every year suffering major disabilities such as breathing difficulties, blindness, and cerebral palsy. Moreover, susceptibility in the perinatal window is not limited to babies. Nearly 300,000 mothers die every year due to complications of pregnancy and childbirth. Together, adverse pregnancy outcomes and the associated deaths and

disabilities constitute the deadliest and longest pandemic of human history.

Despite the high and steady death toll, vulnerability during pregnancy and early in life has for too long been accepted as unavoidable. Although adverse pregnancy outcomes consistently rate as one of the top three causes of death across the entire human lifespan, research to address it receives less than 1 percent of total funding. This disconnect has been thrown into stark relief by the ongoing COVID-19 pandemic. Not only does SARS-CoV-2 represent yet another pathogen that increases risk for stillbirth, preterm birth, and maternal death, uncertainty surrounding the risks to pregnant people and their fetuses who are exposed to the virus highlights the broader reality that the immunology of pregnancy remains largely enigmatic and understudied.

Indeed, much remains unknown about the factors associated with adverse pregnancy outcomes. Nevertheless, the little that is known strongly supports the idea that modulation of the mother's immune system—for example, through diet or maternal vaccination—can improve pregnancy outcomes. Furthermore, given the scalability of these interventions, finally addressing this massive crisis is within reach.

For the duration of pregnancy, immune tolerance of the baby, who is genetically foreign to the mother's body, is critical. So is immune resilience—avoiding undue inflammation, for example, in the face of benign commensal microbes. But inflammation is required for the separation of the maternal and fetal layers of the placenta that occurs leading up to birth. The timing and induction of this inflammation is tightly controlled by physiological signals from both the fetus and the mother around 37 to 42 gestational weeks. Anything that activates this inflammatory cascade too early—what's known as aberrant immune activation—can result in the premature separation of the maternal and fetal placental layers, ultimately causing preterm birth. Interventions that avert unnecessary inflammation thus should reduce the risk of pregnancy complications. By applying the tools of modern science to understand the immunological dynamics of gestation, we may be able to save millions of lives and put an end to this substantial cause of suffering.

Immunobiology of pregnancy

Pregnancy is an immunological marvel, representing the only natural physiological state where genetically foreign cells and tissues lie in close physical contact with the host immune system without rejection. What prevents maternal immune cells from attacking fetal tissues remains unclear. Aberrant activation of maternal immune components associated with pregnancy complications such as prematurity likely represents defects in pregnancy-induced immune tolerance and resilience.

Interestingly, prior pregnancies appear to protect against such complications in future pregnancies. Mothers of sons immunologically remember their babies thanks to long-lived T cells with specificity for Y-chromosome-encoded antigens.[1] Recent characterizations of these fetal-specific T cells in mice have revealed that pregnancy stimulates maternal T cells to adopt functionally unique properties. Researchers have shown, for example, that maternal $CD8^+$ killer T cells develop an exhaustion-prone phenotype, meaning that they selectively silence killer-cell properties upon re-encountering fetal antigens in subsequent pregnancies.[2,3] At the same time, one of us (S.S.W.) and colleagues have shown that pregnancy stimulates the differentiation of $CD4^+$ T cells, which are dedicated to suppressing, instead of activating, other immune cells.[4] Persistence of these immune-suppressive T cells after pregnancy may explain why the incidence of pregnancy complications is sharply reduced in second compared with first pregnancies[5,6]—and why these protective benefits appear to be paternity-specific.[7] Such immune tolerance may be further enforced by fetal cells that continue to circulate in the mother's bloodstream.[8]

A mother cannot afford to totally suppress her immune system, however, as pathogens are an ever-present threat. In addition to warding off infection as well as possible during pregnancy, a mother's body will send immune sentinels across the placenta to provide protection to the baby after it's born. For example, we and others have found that transfer of maternal antibodies to the fetus occurs in utero, ramping up significantly at 30–34 weeks gestation.[9] Transfer of immunological experience continues postnatally through breast milk, which provides protective benefits to babies beyond the neonatal window.

Adverse pregnancy outcomes and the associated fetal, neonatal, and maternal deaths constitute the longest, deadliest pandemic of human history.

Although such vertical transmission of antibodies has long been recognized, details of such immune sharing continue to be unveiled. Earlier this year, one of us (S.S.W.) and colleagues found that pregnancy actively modifies the molecular structure of antibodies, expanding their protective scope beyond extracellular pathogens to include immunity against microbes that live inside cells.[10] This resolves a long-standing conundrum for how antibodies work against pathogens such as HIV, tuberculosis, or Zika virus that live inside cells and thus were once thought to be hidden from antibodies. It also implies that maternal antibodies are not simply immunological effectors, but also serve to activate and regulate an infant's developing immune system, supporting the idea that vaccinating expecting mothers or reproductive-age women (and other individuals capable of pregnancy) prior to conception helps young babies in developing their own defense against microbes.

Of course, not all microbes that we encounter are pathogenic; many are harmless or even beneficial. In the context of pregnancy, controlling inflammation induced by microbes in the birth canal is likely important, as vaginal dysbiosis has increasingly been linked with prematurity and other pregnancy complications. For example, spontaneous preterm birth is consistently linked with depletion of *Lactobacillus crispatus* species and high diversity of other vaginal microbiota.[11] From the babies' perspective, recognizing the difference between microbial friend and foe is critical at birth, as they undergo an abrupt transition to the external world and its plethora of commensal microbes.[12] In this context, microbe-induced inflammation is likely to be more damaging than helpful. Here again, immune molecules, including antibodies, transferred from the mother to the newborn are likely key for the regulation of baby's response to unharmful microbes.

The intricacy of immune regulation in pregnancy remains largely a black box, with many fundamental questions left unanswered. Answering such questions will be essential to understanding and addressing the aberrant maternal immune activation that can lead to adverse pregnancy outcomes.

Immunological interventions to improve pregnancy outcomes

Even with the current, relatively rudimentary understanding of the immunological processes at play during pregnancy, there are immune-modulatory approaches clinicians are already employing to protect pregnancies and newborns. The most direct approach relates to maternal vaccination against common diseases. As infections are generally associated with adverse pregnancy outcomes, protecting mothers against infection can help reduce the risk of preterm births and stillbirths. Such protective effects have been noted with maternal influenza and pertussis immunization already, where reduction of risk for stillbirth or preterm birth was as high as 50 percent in mothers who received either influenza or pertussis vaccinations, or both. When it comes to COVID-19, data point to a higher risk of preterm birth and stillbirth following maternal infection, indicating that maternal vaccination not only protects the mother from severe disease but can also prevent adverse pregnancy outcomes.

Maternal vaccination against various pathogens has also long been recognized for its ability to protect newborn babies, thanks to the antibodies transferred in utero and in breast milk.[9] For example, maternal tetanus vaccination, together with improved birth and umbilical cord hygiene practices, has reduced neonatal mortality from tetanus by nearly 90 percent over the last decade. Similarly, maternal pertussis vaccination prevents severe whooping cough early in the baby's life.

Importantly, vaccines modulate the immune system in ways far beyond pathogen-specific immune responses as well. This has been well documented in non-pregnant vaccine recipients but is likely also occurring in women vaccinated during pregnancy.[13] This suggests that maternal vaccination could be deployed to intentionally modulate the immune system of pregnant women to reduce aberrant immune activation and thereby protect against adverse pregnancy outcomes. For example, mothers experienced improved pregnancy outcomes following maternal influenza vaccination even outside of flu season, indicating pathogen-agnostic rather than only pathogen-specific benefits following vaccination during pregnancy. Additionally, a recent study demonstrated that BCG immunization of women prior to pregnancy also reduced the incidence of adverse pregnancy outcomes, suggesting vaccine-induced modulation of immune trajectories impacting pregnancy may extend to before the gravid period.[14]

Given that the upstream causes of adverse pregnancy outcomes are not sufficiently well understood, it comes as no surprise that insights into vaccine-induced pathogen-agnostic immune-modulatory effects of maternal vaccination improving pregnancy outcomes are limited. Mechanisms could relate to overall immune regulation, such as increased resilience to various immune perturbations. They also could relate to increased innate immunity providing broad pathogen-agnostic protection from a variety of potential microbial culprits, akin to the concept of trained immunity.

Even less is known about how more-indirect immune-modulatory approaches such as dietary interventions reduce adverse pregnancy outcomes. For example, omega-3 fatty acids supplements, which have been shown to protect against preterm labor in populations where omega-3 deficiency is common, may act via systemic immune modulators of fatty acid origin (eicosanoids). Another supplement, the amino acid L-arginine, similarly appears to protect against preterm birth, as well as stillbirth, especially in malaria-infected women, possibly due to a reduction of inflammatory processes in the placental vascular bed.

Irrespective of the missing insight, data showing improved pregnancy outcomes for vaccinated mothers or those taking omega-3 or L-arginine supplements are proof that the global burden of pregnancy complications might be reduced through targeted interventions. Moreover, such interventions offer ideal opportunities to decipher underlying protective mechanisms. Our hope is that, in the future, maternal immunization and nutritional strategies can be improved and better integrated to optimally protect both mother and child.

Call to action

The scale of human suffering during the early life developmental window from conception to birth and beyond constitutes an ongoing public health emergency of frightening proportions. Tackling this problem also constitutes a massive opportunity. Specifically, implementing interventions of promise to pro-

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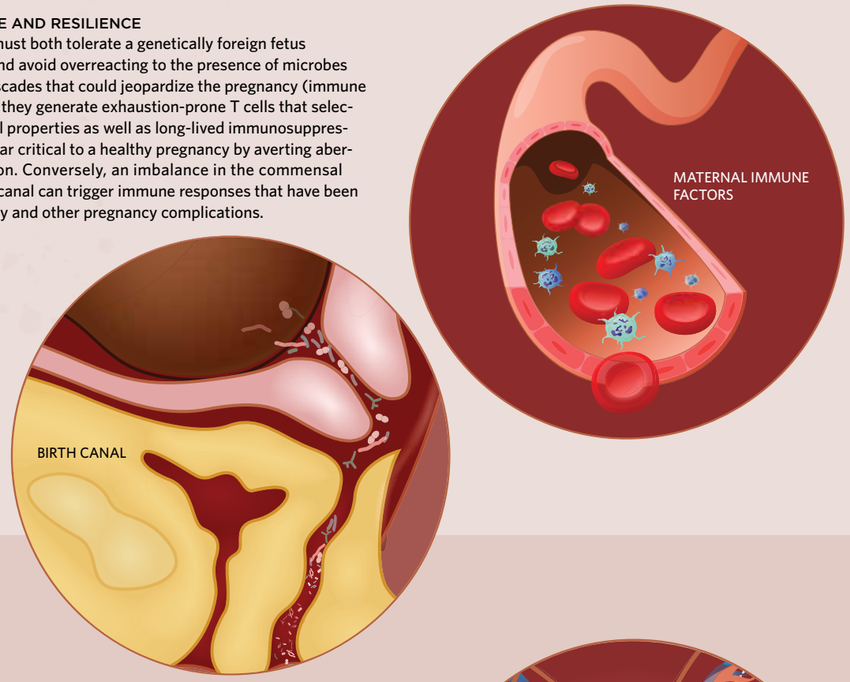
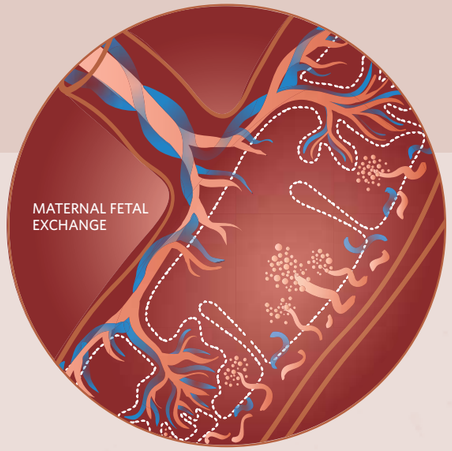
IMMUNOLOGY DURING PREGNANCY

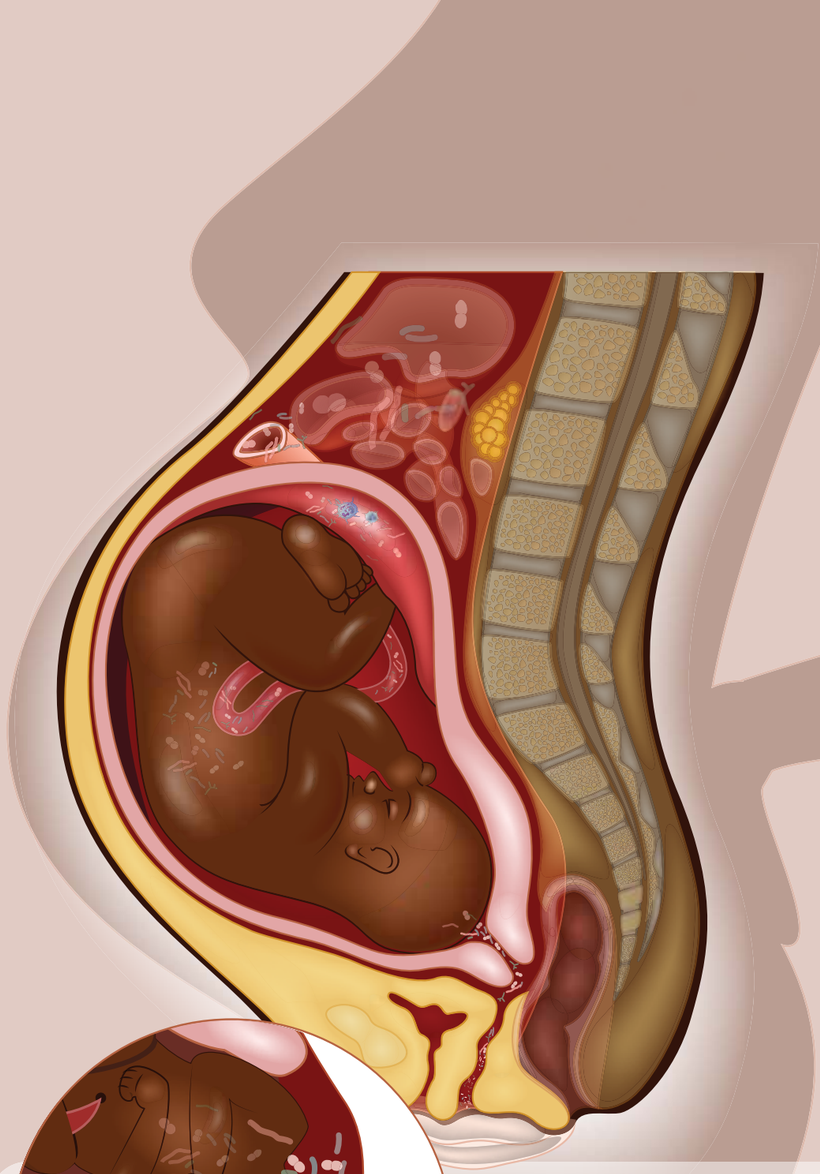
During pregnancy, the immune system adapts to support the baby's development and coordinate birth. When immunity goes awry, so can the pregnancy, with adverse outcomes such as preterm birth and stillbirth often resulting from aberrant immune activation. Diet or maternal vaccination are examples of how to modulate the immune system to improve pregnancy outcomes.

IMMUNE TOLERANCE AND RESILIENCE
Pregnant individuals must both tolerate a genetically foreign fetus (immune tolerance) and avoid overreacting to the presence of microbes with inflammatory cascades that could jeopardize the pregnancy (immune resilience). To do this, they generate exhaustion-prone T cells that selectively silence killer-cell properties as well as long-lived immunosuppressive T cells. Both appear critical to a healthy pregnancy by averting aberrant immune activation. Conversely, an imbalance in the commensal microbes of the birth canal can trigger immune responses that have been linked with prematurity and other pregnancy complications.



VERTICAL TRANSFER OF MATERNAL IMMUNITY
Antibodies and other immune factors can pass across the placenta from mother to child, as well as through breast milk after birth. This means that a mother's acquired immunity to pathogens, including through vaccination, can protect the baby after birth. In addition to providing postnatal protection against specific pathogens, maternal immune molecules transferred to the baby can regulate the fetal and newborn immune system. Such factors can support the baby's in utero immune tolerance to the genetically foreign mother as well as its immune resilience before and after birth, avoiding excessive immune activation by commensal microbes.





INFLAMMATORY CASCADE CONTROLS BIRTH TIMING
While aberrant immune activation can be disastrous, inflammation plays an important role in the process of birth. Typically initiated starting around 37 to 42 gestational weeks, these inflammatory signals can be prematurely activated and trigger the separation of the maternal and fetal placental layers, leading to preterm birth or stillbirth. Certain dietary interventions such as supplements of omega-3 fatty acids or the amino acid L-arginine have been shown to protect against preterm labor in some populations, and may act by reducing inflammatory processes.

THE SCIENTIST STAFF; COMPOSITE FROM © ISTOCK.COM, GUZALIIA FILIMONOVA, VIKIVECTOR; RENDIXALEXTIAN; SFISCHKA

vide rapid reprieve, along with deciphering how these interventions work, would establish the framework to safeguard all women and babies.

Groups around the world, such as the Born Strong Initiative, where one of us (T.R.K.) is chief executive officer, are actively pursuing this mechanistic understanding. The Born Strong Initiative's studies on adverse pregnancy outcomes across the globe focus on interventions that are scalable and feasible for deployment in disadvantaged populations. The initiative's basic approach of contrasting intervention arms with standard-of-care controls aims to reveal the missing mechanistic insight regarding how available interventions can most effectively prevent adverse pregnancy outcomes.

Deploying vaccines not only to prevent infection with specific pathogens, but for their immune-modulatory potential, could save millions of lives.

For these interventions to reduce the global incidence of adverse pregnancy outcomes, access and scalability are key. As stated in the World Health Organization's pioneering Every Newborn Action Plan, "High-quality universal maternal and newborn care is not a privilege but the right of every child and every pregnant woman everywhere." However, lack of access to quality healthcare is estimated to cause two-thirds of neonatal deaths and half of maternal deaths worldwide, with adverse pregnancy outcomes disproportionately affecting disadvantaged and marginalized populations.

Even among privileged factions of society, however, adverse pregnancy outcomes shockingly attract the lowest level of investment along the continuum of care. Compared with COVID-19, which received such a windfall of money and energy that effective vaccines were developed and made available within a year's time, the resources available to scientists interested in understanding and improving pregnancy outcomes seems paltry, despite a death toll that has long outpaced that of the new coronavirus. In addition, there is a disconnect between maternal and newborn healthcare, each involving its own specialists with their own priorities. Yet, models of integrated mother-and-child care are now emerging, and the benefits are becoming apparent. For example, the management of maternal HIV infection and the prevention of newborn infection have led healthcare providers across the globe to join efforts in a multidisciplinary approach. This approach revealed the importance of effective control of maternal HIV infection for the health of the child, beyond the prevention of mother-to-child transmission of the virus. An integrated approach is clearly needed to optimize immune-modulatory interventions aimed at reducing stillbirth and preterm birth.

Although Kate and Adam's son James was tragically lost, his twin brother Fraser eventually graduated from his 123-day stay in the NICU and is now "a typical six-year-old boy in every way," Kate wrote in her note to us. Given his perseverance, his parents gave Fraser the nickname "The Beast." In the future, we are hopeful that fewer babies born too early will have to fight as hard as Fraser did, the onus shifting more to the medical providers overseeing their mothers' pregnancies, with better immunological control converting millions of missed opportunities into more healthy lives lived to their fullest potential. ■

Tobias R. Kollmann is a pediatric infectious disease clinician at Perth Children's Hospital, director of Systems Immunology for the Human Vaccines Project, and chief executive officer of the Born Strong Initiative, a partnership between the Human Vaccines Project and Telethon Kids Institute aimed at reducing adverse pregnancy outcomes. Arnaud Marchant is codirector of the European Plotkin Institute for Vaccinology *and director of the Institute for Medical Immunology of the Université libre de Bruxelles, Belgium.* Sing Sing Way *is a pediatric infectious disease clinician at Cincinnati Children's Hospital, as well as director of the Center for Inflammation and Tolerance and the March of Dimes Ohio Collaborative on Preterm Birth.*

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2022 The Scientist TOP 10 INNOVATIONS

This year's crop of winning products features many with a clinical focus and others that represent significant advances in sequencing, single-cell analysis, and more.

BY *THE SCIENTIST* STAFF

As the acute phase of the COVID-19 pandemic recedes further into the global rearview mirror, life science research—and in particular, the tools that fuel it—continues to forge ahead. The past couple of iterations of *The Scientist*'s annual Top 10 Innovations featured many products that directly addressed the (hopefully) once-in-a-generation disease outbreak, but also highlighted technological advances that pressed forward even in the face of that massive global disruption.

This year's winners reaffirm that the research enterprise has not only persevered but gained momentum as the world emerges from the worst that SARS-CoV-2 threw at us. These include technology that can sequence a human genome for $100, highly sensitive imaging platforms for studying individual cells and subcellular compartments, and an assay system that facilitates protein discovery. There are also several tools with a clinical focus, such as personalized sequencing panels to detect residual cancer cells left after tumor removal, a software tracking system for overseeing gene and cell therapies from bench to bedside, and a DNA processing tool that improves technicians' ability to analyze fetal DNA in a mother's blood.

We are happy to announce the new products that our panel of independent judges has chosen as this year's Top 10 Innovations.

NeXT Personal™
Personalis

Small quantities of cancer cells can linger in the body after tumor removal, a phenomenon known as minimal or molecular residual disease (MRD) that, if left undetected, can lead to recurrence. Personalis's NeXT Personal™ assay, unveiled in December 2021, uses a patient's own tumors to detect, quantify, and monitor circulating tumor DNA in order to spot MRD and track responses to therapy.

Using 1 mm^3 of tumor tissue sampled from a patient, Personalis's lab performs whole genome sequencing to identify up to 1,800 single-nucleotide variants. These patient-specific mutations are used to design a panel of primers targeting those regions for sequencing in future blood biopsies. The assay also includes primers for other known cancer-related genes, says Dan Norton, associate director for product management at Personalis. "We can see if there are other variants emerging that have a precision therapy associated that may be more effective for that patient."

Medical oncologist Jonathan Loree of BC Cancer and the University of British Columbia began partnering with Personalis in August to use NeXT Personal in a study of patients who'd had tumors removed from their pancreases or colons, testing the technology's ability to diagnose disease recurrence "earlier when there is a window of opportunity for patients to still be cured," he says in an email to *The Scientist*. Loree says that if the assay could replace conventional diagnostic CT scans and blood tests, "[t]hat has the potential to save money [and] improve outcomes."

NeXT Personal is currently used in research only, with plans to expand to clinical trial settings next year, Norton says. Personalis declined to provide a cost for NeXT Personal, explaining that the price varies depending on user needs.

KAMDAR: *"NeXT Personal offers [the potential to] address a number of tumors that are not fully analyzed by other technologies to help identify and manage a patient's disease."*

PERSONALIS

WINNER

THE JUDGES

KIM KAMDAR
Managing partner at Medical Excellence Capital, a healthcare-focused venture fund creating and investing in biopharma and diagnostic companies. She began her career as a scientist and pursued drug-discovery research at Novartis/Syngenta for nine years.

MAHENDRA RAO
CEO at Implant Therapeutics. Rao has published more than 200 papers on stem cell research and is the cofounder of the neural stem cell company Q Therapeutics, based in Salt Lake City, Utah. He has served on advisory panels for the US Food and Drug Administration (FDA), as well as for the governments of the US, Singapore, and India on policies pertaining to human embryonic stem cells. He continues to work with the FDA and other

WEI-JUN QIAN
Bioanalytical chemist at Pacific Northwest National Laboratory. His research centers primarily on the development and applications of mass spectrometry–based approaches to better quantify the dynamic changes in protein abundances and protein post-translational modifications in biological and clinical applications.

KRYSTYN VAN VLIET
Michael (1949) and Sonja Koerner Professor in the departments of materials science and engineering and biological engineering at MIT. She also leads the Singapore-MIT Alliance for Research and Technology's Critical Analytics for Manufacturing Personalized-Medicine (CAMP) research team.

2 CosMx™ Spatial Molecular Imager
NanoString Technologies

The CosMx™ Spatial Molecular Imager (SMI) visualizes and quantifies RNA and protein levels at the single-cell and even subcellular levels. The platform, developed by NanoString Technologies, Inc., allows users to follow a standard protocol to prepare and hybridize specific probes and antibodies to their samples, which can be frozen tissue slices or formalin-fixed, paraffin-embedded slices. In the automated instrument, reporter sets hybridize and are imaged, then the fluorescent dyes are cleaved with UV light and washed off before the next reporter set hybridizes with the sample, allowing researchers to image multiple targets in one sample.

The CosMx SMI, priced at US$295,000, contains a high-resolution microscope and "allows researchers to visualize and quantify 1,000 RNA and 100 protein targets at a subcellular resolution across more than 1 million cells," Vikram Devgan, senior director of Spatial Genomics Business at NanoString, says in an email to *The Scientist*. He adds that users can also purchase the AtoMx™ Spatial Informatics Platform, a subscription-based software produced by NanoString, to visualize and analyze the data generated by the CosMx.

"The CosMx is the only instrument that has provided us with the opportunity to simultaneously visualize thousands of genes, at subcellular resolution, and across all cells in a tissue," says Miranda Orr, an Alzheimer's disease researcher at Wake Forest University School of Medicine in North Carolina who, after using another NanoString product, bought the CosMx SMI. "We are able to develop maps of the brain at an unprecedented resolution."

QIAN: *"This will transform the in situ spatial biology and molecular pathology fields."*

NANOSTRING TECHNOLOGIES

3 UG100™
Ultima Genomics

Ultima Genomics announced this May that it had developed technology to sequence an entire human genome for US$100. Thus far, only early-access customers have had the chance to use the company's new platform, called UG100™, but Ultima Genomics expects to release the product to the broader market in the first half of 2023. Compared to other sequencers, UG100 has several advantages, including higher speed, better efficiency, and less waste, says Josh Lauer, the company's chief commercial officer.

Lauer attributes many benefits of the UG100 to a unique feature: a circular, open flow cell. Reagents are applied directly to a spinning silicon wafer that distributes them more efficiently than reagents pumped through a traditional flow cell, Lauer

explains. In addition, the revolving design increases the speed of data collection and imaging, enabling Ultima's sequencer to complete one run in about 20 hours, which he says is about twice as fast as existing technologies. "Much like a CD player, this enables ultra-high-speed scanning of genetic material."

"I'm excited about the throughput of the platform, as well as the cost," says Reuben Saunders, a genetics graduate student at the Whitehead Institute who collaborated with Ultima Genomics to use its UG100 in recent research. "It's heralding an exciting era where very large-scale experiments . . . will become accessible methods that can really drive advancements in our understanding of genetics and cell biology."

Lauer says the $100 per genome cost includes wafers and chemical reagents, but Ultima Genomics declined to release the price of the refrigerator-sized hardware that performs the sequencing.

RAO: *"This is the first under $100 genome, and they have achieved it with an innovative use of technology."*

4 Proteograph™ Product Suite
Seer

Proteomic studies have traditionally faced two key challenges, says Rebecca Rutherford, Director of Product Management at biotech company Seer: The need to tag proteins has restricted research to known proteins, and the large, diverse nature of the proteome has made investigating low-abundance proteins difficult. Seer's Proteograph™ Product Suite, launched in January 2022, addresses both challenges, she says, using a nanoparticle-based assay that allows unbiased sampling of all peptides in a biofluid sample. "The innovation in the Proteograph Product Suite is really around our proprietary engineered nanoparticles that have unique surface functionalization that attract proteins across the entire dynamic range." This allows researchers to track small molecular changes associated with disease and reveals distinct protein variants produced by post-transcriptional modifications, Rutherford says, enabling the identification of novel and biologically relevant proteoforms.

"From a [discovery] proteomics perspective, the complex liquid biopsies like blood, serum, and plasma that we work with have just been inaccessible," says Mark Flory of the Cancer Early Detection Advanced Research Center (CEDAR) at Oregon Health & Science University Knight Cancer Institute who collaborated with Seer to test the new platform before becoming the first client to purchase it. Proteograph enables "deep sampling in those very complex liquid biopsy types." His research team has been applying the platform to a large cohort study of prostate cancer to identify new biomarkers. The product is also being applied to research on lung cancer and Alzheimer's disease, according to Rutherford.

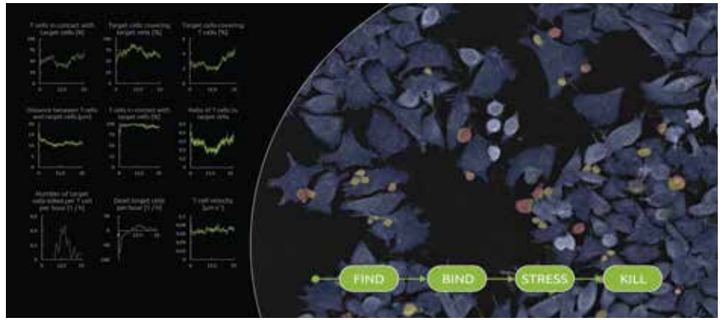
Seer declined to provide a price for the Proteograph™ Product Suite.

KAMDAR: *"Provides access to the proteome in an unbiased way and does for proteomics what next-generation sequencing has provided in genomics."*

SEER © 2022; ULTIMA GENOMICS

LIVE T Cell Assay
Nanolive

5



Over the past decade, Nanolive has developed imaging platforms based on technologies that reconstruct three-dimensional holograms of label-free samples. In September 2021, they launched their first application-specific digital assay, LIVE T Cell Assay, which examines how T cells locate, bind, stress, and kill their targets, such as infected, foreign, or cancerous cells. The assay measures phenotypic and morphological parameters of both the T cells and the target cells, but "what's really novel about the product is the metrics that we can extract from the interaction between the two," says Emma Gibbin-Lameira, scientific communications manager at Nanolive. Such information can be very powerful in testing the efficacy of a drug, she adds. For instance, you can assess whether a specific antibody brings T cells closer to the desired target and whether it increases the cells' killing rate.

Valery Moine, a unit head in the Pharmacology group at Switzerland-based Light Chain Bioscience who collaborates with Nanolive, says he started using LIVE T Cell Assay a year ago to create "marketing videos to highlight and promote the mode of action of bispecific antibodies" developed by his company. More recently, he says he has been using the platform to further characterize these antibodies. The metrics it provides, he adds, are valuable for ranking the best candidates.

Nanolive declined to share the price of the assay, but Lisa Pollaro, the chief marketing officer at the company, writes in an email that "it comes with an annual license with a price in the same range of chemical assay kits available in the market."

RAO: *"A non-end point assay for the rapidly developing field is a huge advance in enabling therapy."*

MARS® Bar
Applied Cells, Inc.

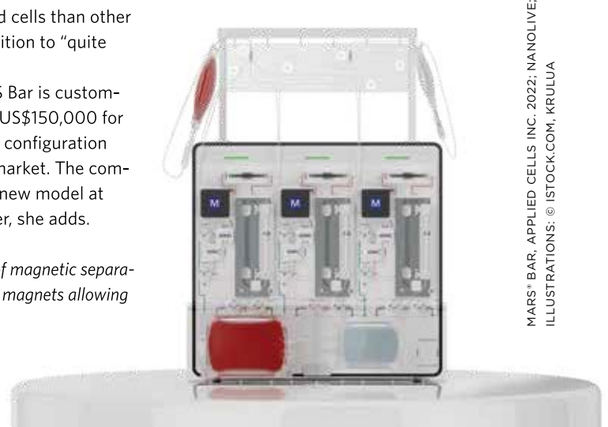
One of the most efficient methods to select cells is immunomagnetic separation, where the isolation is based on the presence of magnetic beads attached to specific cell surface antigens. There are several products on the market that apply this method, but a new system presented by Applied Cells, Inc., called MARS® Bar, has various advantages, says Liping Yu, vice president of applications at the company. For instance, MARS Bar works as a closed system with sterile fluidic kits, which makes it "much easier to manage," says Yu, because its use is no longer restricted to a clean room or biosafety cabinet. Additionally, the device contains three modules, allowing it to process three samples in parallel.

Sergei Rudchenko, an assistant professor at Columbia University who has an ongoing scientific collaboration with Applied Cells, has been using a version of the product, MARS Bar Flex—quite similar but with open fluidics—since February for a protocol that aims to remove naive T cells from peripheral blood mononuclear cells during cell transfusion. According to Rudchenko, who says he is interested in developing the protocol into a clinical application to prevent graft-versus-host disease, MARS Bar Flex achieves about eight times higher recovery of blood cells after depletion of unwanted cells than other published protocols, in addition to "quite competitive purity."

According to Yu, MARS Bar is customizable but will cost around US$150,000 for the standard three-module configuration once it is available on the market. The company started demoing this new model at customer sites in November, she adds.

RAO: *"Very innovative use of magnetic separation technology without fixed magnets allowing a high throughput."*

6



MARS® BAR, APPLIED CELLS INC. 2022; NANOLIVE; ILLUSTRATIONS: © ISTOCK.COM, KRULUA

Single Cellome™ System SS2000
Yokogawa Electric Corporation

The Single Cellome™ System SS2000 by Yokogawa Electric Corporation is an automated subcellular sampling system, which contains both a dual spinning-disk confocal microscope to visualize tissue and a sampling setup to collect whole cells or intracellular components from a single cell.

The system, launched in February 2022 in Japan, the US, and China, can be fully automated, giving users the ability to define which cells should be sampled based on their cytoplasm area, nucleus size, or other morphological features. Cells that fit a particular profile are aspirated and deposited in a 96-well plate for further analysis. The system can also directly sample intracellular components such as organelles or parts of the cytoplasm and combine these insights with whole-cell sampling. The SS2000's "high-resolution 3D image allows researchers to control the location of [cell and subcellular] sampling in a highly precise way and retains the spatial context," says Takanobu Kiuchi, head of global marketing at Yokogawa.

"We can sample multiple times from the same cell, you collect a small intracellular sample for metabolomics, and then collect the rest of the cell for single-cell transcriptomics," says Carla Newman, associate director of Cellular Imaging and Dynamics at GSK, who received an SS2000 from Yokogawa as part of a research collaboration agreement for beta testing, Especially for rare phenotypes, she notes, the ability to target sampling to specific features is highly useful, as well as being able to sample small numbers of patient cells. "It allows for the granularity of the single-cell level to pick up rare events." Newman adds that the SS2000 is faster and easier than traditional micromanipulators, improving the sampling speed by at least 10 times.

Depending on the technical configuration and required support, the instrument list price ranges from €650,000 (US$651,800) to €725,000 (US$727,088).

QIAN: *"This is a highly innovative system coupling high resolution cellular imaging with subcellular sampling technology."*

7

YOKOGAWA ELECTRIC CORPORATION; YOURGENE HEALTH

LightBench® Detect
Yourgene Health

8

The LightBench Detect® is a DNA processing tool that is useful for noninvasive prenatal testing (NIPT), which involves fetal DNA collected from the mother's blood. To help find the tiny strands of fetal DNA among the clusters of maternal DNA, the LightBench Detect separates the fragments by length, explains Yourgene Health product manager Becky Underwood.

The product employs Yourgene's imaging system, Ranger Technology, to image the gel and make real-time adjustments to the voltage to optimize strand separation, Underwood says, adding that the LightBench Detect is the only instrument on the market that can use plastic EDTA blood collection tubes, which are cheaper, less susceptible to breakages, and more efficient than the industry standard glass ones. These features yield 50 to 75 percent more fetal DNA per sample, according to Yourgene's product page.

The instrument, which costs US$50,000, launched in early June and is still in its testing stages with company scientists and a few outside researchers, Yourgene tells *The Scientist*. Bhavika Patel, director of the Yourgene genomics services lab that is using the product, says its usage of EDTA tubes has significantly cut down the lab's costs and failure rates, potentially putting less strain on patients and getting quicker results. "It's quite a nice, neat instrument," Patel says.

In addition to its use for NIPT, Underwood says the LightBench Detect has a wider range of applications, including scanning blood samples for tumor DNA or infectious pathogens. "We want to positively influence clinical pathways and improve patient outcomes," Underwood tells *The Scientist*.

VAN VLIET: *"This can be a game changer for widely used diagnostics globally, especially given price point and ease of sample preparation."*

9
Molecular Cartography™
Resolve Biosciences

Resolve Biosciences's Molecular Cartography™ workflow is a single-molecule fluorescence in situ hybridization technology that offers a three-dimensional view of gene expression within cells without damaging the tissue section or cell culture sample. The company launched the platform as a mail-in service last year, and that service won a spot in the 2021 Top 10 Innovations list. Resolve Biosciences began installing hardware and software for the fully automated Molecular Cartography workflow in customer laboratories in January 2022, for a cost of US$400,000.

"We're able to bring the assay into the actual disease state and map interactions at the single molecule level within tissues," says Jason T. Gammack, cofounder and CEO of the firm. The platform produces high-resolution images of subcellular gene expression, which, in addition to providing unique insights into the transcriptional landscape of the cell, are "quite breathtaking," Gammack says. "You now see the beautiful symmetry of biology."

"It is a ready-to-go system that needs very little optimization," says Jan-Philipp Mallm, head of the Single-Cell Open Lab at the German Cancer Research Center (Deutsches Krebsforschungszentrum, DKFZ). A major advantage of Molecular Cartography is its fully customizable panel of probes, says Mallm, who used the mail-in service before purchasing the full workflow for his laboratory this year. "I think the versatility is a big asset of the system." Mallm and his colleagues are applying the technology to study the cancer microenvironment, where "a tumor can be regarded as a whole complex tissue and thus needs single-cell spatial resolution in order to understand its function and capabilities."

KAMDAR: *"This view of subcellular gene expression activity can facilitate new insights into the interactions and complexity of critical biological mechanisms."*

10
OCELLOS 3.0
TrakCel

Cell and gene therapy have started to revolutionize medicine, but they've also presented new challenges in tracking the materials involved, including patients' own cells. To address these challenges, TrakCel introduced a cloud-based software called OCELLOS in February 2021 and released a new iteration, OCELLOS 3.0, this July. "It's a really simple computer interface that's easy to access throughout the whole supply chain," says Matthew Lakelin, a cofounder of TrakCel and the company's vice president of scientific affairs and product development. OCELLOS 3.0 not only bolsters the safety and quality of such treatments, but it manages the chain of identity and chain of custody data that regulatory agencies require, Lakelin notes.

Edward Armstrong, senior director of quality assurance at Mustang Bio, has been partnering with TrakCel since 2018, using their technology to track Mustang's autologous CAR T cell products and cell and gene therapies during clinical trials. "Chain of custody and chain of identity are critical to our process and are looked at very heavily by the Food and Drug Administration," says Armstrong, who is currently switching to OCELLOS 3.0. When Mustang Bio was looking to enroll its first patient, Armstrong considered tracking the products on paper, but quickly realized that "to do on paper what TrakCel does electronically would drive mortal men insane."

TrakCel's OCELLOS 3.0 starts at $300,000, but the price may increase based on the amount of involvement and customization desired.

VAN VLIET: *"This product can help focus, simplify, and reduce errors in the complex supply chain and task logistics of cell/gene therapy (CGT) development and production."*

RESOLVE BIOSCIENCES; TRAKCEL: ILLUSTRATIONS: © ISTOCK.COM, KRULUA

Ankur Jain: Neurodegeneration Explorer

Assistant Professor, Department of Biology, MIT

BY HANNAH THOMASY

Ankur Jain didn't always know he wanted to be a biologist. During his undergraduate studies at the Indian Institute of Technology Kharagpur, he originally focused on engineering, but soon realized that he wasn't actually that interested in understanding how human-made things work. Instead, he wanted to study the mechanisms that make things tick in the natural world, he says. Jain ended up graduating with a degree in biotechnology and biochemical engineering in 2007, and moved to the United States to pursue his doctorate in biophysics and computational biology at the University of Illinois Urbana-Champaign.

During his PhD, Jain focused on developing better techniques to study what proteins do in cells. "The machines inside the cell—the proteins—they often act in conjunction. Maybe one protein alone does not do its task, but [it does when] it's bound to something else," he says. "The question is, what is it working with? What are its partners in crime?"

One method for determining protein partners is a pull-down assay—anchoring one type of protein to beads in a column and identifying which other proteins bind to, or are "pulled down" by, it. But this technique runs into difficulty when proteins have multiple partners.

To fix this problem, Jain led the effort to develop a single-molecule pull-down (SiM-Pull) assay in which protein A is attached to a slide, while proteins B and C are tagged with fluorescent peptides (*Nature*, 473:484–88, 2011). By overlaying fluorescent microscopy images of single-molecule resolution, this technique can determine whether protein A can bind to B and C at the same time, or only separately. Jain later used this technique to study the components of mTOR complexes, which play important roles in normal cell growth but also in many diseases, including cancer (*PNAS*, 111:17833–38, 2014).

After receiving his doctorate in 2013, Jain took a postdoctoral position with biochemist Ronald Vale at the University of California, San Francisco. During this time, he pivoted to studying the internal organization of cells. Many intracellular structures are enclosed in membranes, but others aren't, Jain says. "Just like oil and water separate from each other, there are compartments inside the cell which don't have a physical boundary surrounding them, but still maintain their identity."

Jain wasn't initially studying this phenomenon, called liquid-liquid phase separation, in any particular disease. But in 2014, the Ice Bucket Challenge, which highlighted amyotrophic lateral sclerosis (ALS), took the internet by storm. Jain learned that some forms of familial ALS are associated with abnormal nucleotide repeats in a specific gene. He noticed that these disease-associated patterns in patient's DNA—and the RNA it is transcribed into—looked similar to the synthetic DNA and RNA he was making to study RNA aggregation and phase separation.

In a 2017 *Nature* paper, Jain demonstrated that RNA molecules with repeat expansions, including those associated with diseases such as ALS and Huntington's, bond with each other in specific ways, forming gels and undergoing phase separation to create structures called RNA foci (546:243–47, 2017). While RNA foci had been observed in the cells of patients with repeat expansion disorders, this study revealed the mechanisms by which repeat expansions in DNA lead to RNA foci and suggested that these abnormal RNAs, and not just abnormal proteins, may be involved in driving disease.

Vale tells *The Scientist* that he was "very impressed with the originality of [Jain's] work as a graduate student . . . and that spark of original thinking and independence came through in his postdoc as well." On top of that, Vale adds, Jain is "just a really wonderful person to interact with and do science with."

In 2018, Jain joined the Whitehead Institute at MIT, where he continues to study the role of RNA foci in neurodegenerative disease. This year, he was named a Pew Biomedical Scholar, and he says he plans to use the accompanying funding to investigate the role of polyamines in RNA aggregation.

Among his other projects, Jain recently teamed up with Jing-Ke Weng, an MIT biologist who studies plant metabolism, to screen plant-produced small molecules for their abilities to inhibit the aggregation of RNAs and proteins relevant to human neurodegenerative disorders. This collaboration is scientifically promising, but also enjoyable for the scientists involved, Weng says. Both he and Jain are "quite interested in the fundamental mechanisms of how things work, how molecules behave in the cells," he adds. "We just happen to be trained in very different disciplines. So when we meet and discuss, we have a lot of fun." ■

GRETCHEN ERTL, WHITEHEAD INSTITUTE

Chantell Evans: Mitochondria Tracker

Assistant Professor, Department of Cell Biology, Duke University

BY HOLLY BARKER

Growing up in a small town in Illinois, Chantell Evans recalls spending much of her time gazing up at the skies, searching for constellations, or down at the ground, picking out new rocks for her collection. At school, she gravitated toward science, enrolling in a bachelor's program in chemistry at Southern Illinois University in 2005. It was here, earning extra money by organizing lab equipment, that she got her first real glimpse of research. In a world beyond the pH tests and titrations of her early chemistry classes, Evans watched scientists in the labs using sophisticated technology to chip away at life's unknowns.

Keen to become part of this world, Evans began a PhD in molecular and cellular pharmacology at the University of Wisconsin–Madison in 2009. She joined the lab of neuroscientist Edwin Chapman, where she investigated synaptic vesicle exocytosis, a signaling process in neurons that is mediated by influxes of calcium ions. To demonstrate that the speed of synaptic transmission depends on a protein called synaptotagmin 1 (syt 1), Evans engineered a version that interacts with the neuronal membrane for longer, enabling her to untangle syt 1's impact on vesicle exocytosis.

Evans took the syt 1 backbone and grafted on loops from other naturally occurring, slower-acting forms of synaptotagmin, assembling an enormous panel of chimeras. It was a challenging protocol, Chapman tells *The Scientist*, and yet "she was a natural." Compared to the normal protein, Evans's chimeras prolonged neurotransmitter release, demonstrating that syt 1 determines the time course of synaptic transmission (*J Neurosci*, 35:11769–79, 2015).

According to Chapman, Evans became a "lab leader" in other complex techniques too, including isothermal titration calorimetry, fluorescence labeling, and spectroscopy. When it came time for a postdoc, Evans says she was eager to tackle skills missing from her repertoire. She was drawn to University of Pennsylvania cell biologist Erika Holzbaur, whose group was using live-cell imaging to spy on cellular organelles in real time. Evans arrived in 2016, focusing on mitophagy—the targeted degradation of damaged mitochondria—and its role in neurodegenerative disease. The dynamic nature of the organelles made the research interesting from a technical perspective, says Evans, and investigating their role in neurodegeneration gave the project purpose.

Previous cell culture work by Holzbaur's group had revealed how optineurin, a protein associated with amyotrophic lateral sclerosis (ALS), is recruited to damaged mitochondria as part of the PINK1/Parkin signaling pathway. Mutations in both *PINK1* and *PARK2*, genes that encode components of this pathway, are also known to cause Parkinson's, supporting the idea that mitochondrial dysregulation may be a common feature of neurodegenerative diseases. The PINK1/Parkin pathway had been investigated in a nonneuronal cell line, but Evans knew it needed to be explored in neurons.

She set about establishing a protocol to induce mitochondrial stress in cultured hippocampal neurons, improving on previous methods that were inconsistent or produced unrealistic levels of damage. "She was very successful in identifying simple conditions that work consistently and give mild damage," says Holzbaur. "It was a huge step forward."

Live-cell imaging confirmed that optineurin is recruited to mitochondria in response to stress, and revealed how damaged mitochondria stick around for an unexpectedly long time, lingering within autophagosomes for more than 24 hours (*eLife*, 9:e50260, 2020). These findings offered a clue to mitophagy's links with neurodegeneration: Natural aging or mutations in mitophagy proteins may delay an already sluggish cleanup process, leading to a buildup of damaged mitochondria that leads to neuronal death.

It was during her postdoc that Evans was awarded a highly competitive Hanna H. Gray fellowship, including $1.4 million in funding. As part of her application, Evans was asked to write a long-term proposal, which forced her to take a future-focused perspective early in her research career. "It really allowed me to take a big step back from my project and say, 'If you could run a lab, what would you want to do?' It helped me to start thinking about those things way earlier than maybe a lot of other postdocs would have."

In 2021, she launched her own lab at Duke University, where she continues to probe mitophagy in the brain. Evans has assembled a team of scientists from diverse backgrounds, not only in terms of ethnic or religious identity, but with different thinking styles too, she says. Her group today includes a cell biologist, a neuroscientist, and an ecologist, each "coming with their own unique experiences" to answer the same question from different perspectives. ■

JEFF FUSCO AND HOWARD HUGHES MEDICAL INSTITUTE (HHMI)

Expert JeWell-ry Designers

Imaging organoids has proven slow and cumbersome for scientists. But a new technique may speed things up, producing 3D images of hundreds of organoids per hour.

BY NATALIA MESA

Organoids, three-dimensional miniature organs grown from stem cells, are powerful tools for studying development and disease, for testing new drugs, and potentially even for transplantation. But so far, growing and imaging them at scale has proven difficult. Now, a team at the University of Bordeaux, in collaboration with scientists at the National University of Singapore, has designed an automated technique that takes mere seconds to image an organoid in 3D (*Nat Meth*, 19:881–92, 2022).

Organoids grown under the same conditions can still develop differently, so "you need to have a lot of organoids in the same condition to be able to understand what's going on," explains National University of Singapore cell biologist Anne Beghin, who helped develop the technique. But capturing a 3D image of the entire structure is tricky, she adds, because light is toxic to cells, and most 3D imaging methods are light-intensive, not to mention slow. Previous methods could image just "ten or twenty" organoids at a time, Beghin says.

In 2015, Bordeaux physicist Jean-Baptiste Sibarita and his colleagues developed a method to capture super-resolution 3D images of live single cells. The technique, Single-Objective Selective-Plane Illumination Microscopy (soSPIM), differs from traditional cell-imaging methods because it only illuminates a single plane of the sample at a time and uses only one objective, minimizing cells' light exposure (*Nat Meth*, 12:641–44, 2015).

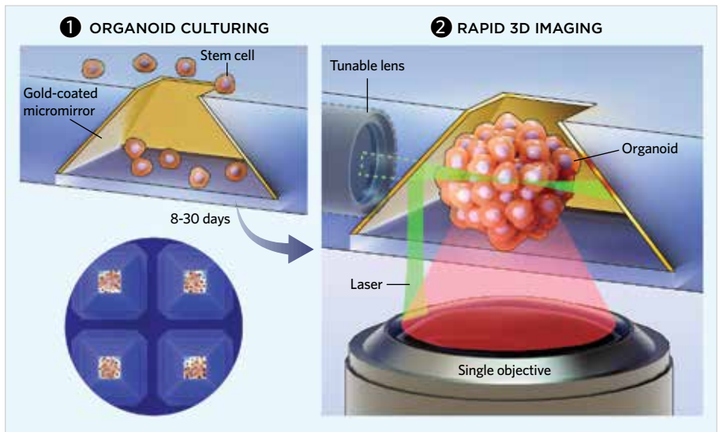
To apply soSPIM to organoids, Sibarita, Beghin, and their colleagues designed JeWell chips: high-density arrays that contain cavities composed of four mirrors arranged in a pyramid. The shape of each JeWell cavity keeps the organoid grown within from spilling out and facilitates soSPIM imaging. A laser positioned below the JeWell chip bounces off a mirror to illuminate thin slices of either fixed or live organoids tagged with fluorescent markers. A camera then captures the reflected light and assembles a 3D image, layer by layer.

Beghin successfully grew neural, liver, and cancer cell–derived organoids, among others, inside the JeWells and applied the imaging approach. The researchers also adapted machine learning–based tools to pick out cells undergoing mitosis or apoptosis and taking on properties of organoids.

Using this approach, the researchers imaged a single organoid in seven seconds—and roughly 300 organoids in an hour—employing a single color of fluorescence. Using three colors to tag three separate proteins, they could image about 96 organoids per hour. "There is a huge demand in terms of getting organoids close to the pipeline to drug discovery," says Beghin, adding that this approach should help meet it by helping pharmaceutical companies integrate organoids into high-throughput drug screening protocols.

Cardiff University cell biologist Trevor Dale, who researches organoids but was not involved in the study, says he worries the JeWell's unique shape may prevent important structure-giving molecules from reaching the organoids, rendering the technique unsuitable for growing certain types of organoids. However, he adds that the imaging definitely "appears to increase the rate at which you can acquire 3D data," whereas similar techniques he's tried have "taken ages." ■

SPARKLING JEWELLS: (1) Researchers seed JeWells—pyramid-shaped wells in a high-density array, each made of four highly reflective gold-plated mirrors—with stem cells. After a period of growth, the resulting fixed or live organoids are stained with fluorescent dyes (2) and imaged via Single-Objective Selective-Plane Illumination Microscopy (soSPIM), an adapted form of light-sheet fluorescence microscopy that allows for subcellular resolution imaging. This technique uses a simple inverted microscope to take 2D cross-section images of the sample layer by layer, which are assembled into a 3D picture of the organoid.



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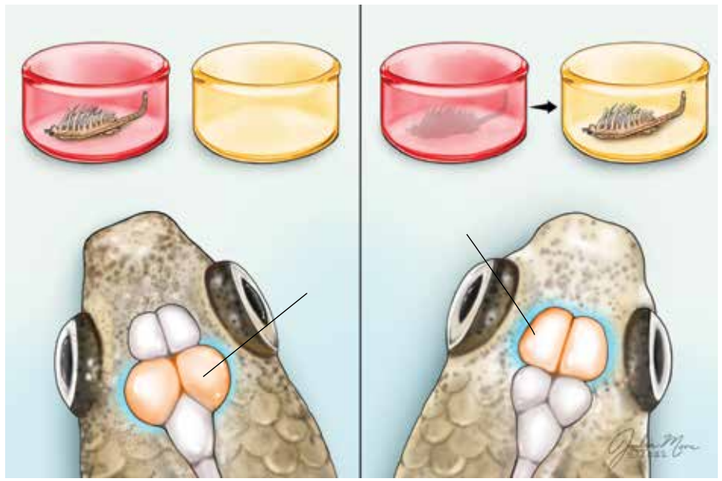
The Literature

NEUROSCIENCE

Gifted Guppies

THE PAPER
Z. Triki et al., "Brain morphology correlates of learning and cognitive flexibility in a fish species (*Poecilia reticulata*)," *Proc R Soc B*, 289:20220844, 2022.



© JULIA MOORE

FISH BRAINED: 1 The optic lobes are thought to be involved in visual processing. In this study, researchers found that guppies with larger optic lobes more quickly learned a visual discrimination task—identifying which color well contained food. 2 The fish telencephalon is thought to be involved in spatial learning, memory, and inhibitory control. Here, the researchers found that a larger telencephalon might enhance the fish's cognitive flexibility, allowing them to more quickly associate food with a new color after the researchers switched it.

Despite their vacant stares, fish are surprisingly brainy. They can quickly learn tasks, pilot vehicles, and may even be able to count. Some fish are able to pick up on complex tasks more quickly than others, and researchers previously attributed these individuals' smarts to bulkier brains. But research published July 13 in *Proceedings of the Royal Society B* finds that some individual differences in cognitive ability among fish of the same species may stem from relative size differences between specific brain regions, not just brain size overall (289:20220844, 2022).

Zegni Triki, a biologist at Stockholm University, had an inkling that brain region size might influence task performance. For example, she knew that among wild gobies, another family of small fish, species that dwell in craggy rocks have bigger telencephalons, while sand-dwelling species have larger optic lobes. The telencephalon is a brain region associated with cognitive skills including memory and decision making, while optic lobes are brain areas that process visual information.

So Triki selected guppies (*Poecilia reticulata*) with large or small telencephalons and optic lobes. She bred the two groups separately over three generations, then measured how well each group performed at two tasks. First, guppies learned to discriminate which of two differently colored wells contained food. The second task added a twist. Once the fish had learned the first task, the colors were reversed. "That means, once your animals learn an association successfully, you make them unlearn it," Triki says. The researchers measured the time it took the guppies to adjust—cognitive flexibility that Triki says represents "quite a difficult task for animals."

She then compared the brains of the two groups and found that, while the average brain size remained the same between the two, fish with larger optic lobes excelled at the initial color discrimination task. Triki says this makes sense since the area "is mainly used for visual information processing."

Meanwhile, fish with larger telencephalons fared better at the second task. That came as more of a surprise, Triki says, adding that it's the first evidence that the telencephalon is involved in cognitive flexibility.

B. Wren Patton, a graduate student in marine biology at Pennsylvania State University who was not involved in the research, says that the study "was really clean . . . they did a really good job being very precise with their descriptions."

Patton says that she appreciated that the study focused on brain regions, rather than the brain as a whole. She also says that the artificial selection in the study was a "really interesting aspect of the story" and she'd like to see the researchers breed the fish for a few more generations before testing. The experimental design "make[s] sense in the field of animal behavior," she adds. "If you want to make comparisons between what these parts of the brain are really driving . . . [in terms of] the behavioral and cognitive capabilities of the individual, you really need exactly this kind of design."

—Natalia Mesa

DEATH BY LIGHTNING: While lightning is a major source of mortality for most large tropical trees, species vary in their susceptibility to lighting damage.

EDITOR'S CHOICE IN ENVIRONMENT

Tree Killers

THE PAPER

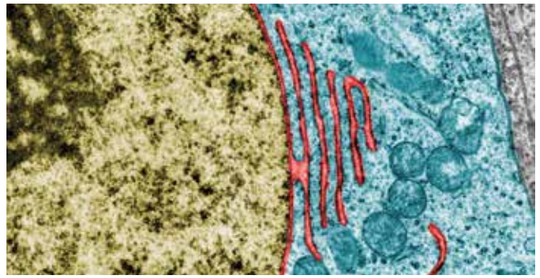
J.H. Richards et al., "Tropical tree species differ in damage and mortality from lightning," *Nat Plants*, 8:1007-13, 2022.

Although historically overlooked, lightning may play a surprisingly large role in shaping tropical forests, accounting for as much as 40 percent of large tree mortality. But like other drivers of mortality, it likely doesn't affect all trees equally. "Species differ in their susceptibility to drought, their tolerance of fire, and all of these other hazards that they're exposed to," says Jeannine Richards, a plant ecologist at the University of Wisconsin–Madison. "Certainly, there should be differences in how they respond to the electrical current of a lightning strike."

To explore this hypothesis, Richards and her colleagues used a combination of cameras, electrical field change meters, and field surveys to pinpoint lightning strikes and assess the damage to trees at those locations. Of the 30 species the group identified, palm trees were highly likely to die if struck by lightning, while four species of broadleaf trees—a diverse group of seed-bearing, flowering trees—had comparatively little mortality following a strike. Among all the species included in the study, trees with higher wood density seemed to have greater lightning tolerance. Richards says that the mechanism underlying this finding isn't clear and could be the basis for further study.

Nate McDowell, a forest ecologist at the Pacific Northwest National Laboratory in Washington who was not involved in the research, says that this study has important implications for the future of tropical forests under climate change, which is expected to make lightning more common. "If lightning does increase in frequency in some regions . . . as expected," McDowell says, "we would anticipate that this will cause changes in community demography—the winners and the losers." He adds that changes in tree communities will likely affect the carbon cycle, although exactly how this will play out remains to be seen.

—Hannah Thomasy



LIPID PROCESSING: Researchers detected the TMEM63C protein in the endoplasmic reticulum (red), the cellular hub for processing lipids, of human cells.

EDITOR'S CHOICE IN CELL BIOLOGY

Misshapen Organelles

THE PAPER

L.C. Tábara et al., "TMEM63C mutations cause mitochondrial morphology defects and underlie hereditary spastic paraplegia," *Brain*, awac123, 2022.

Geneticists Emma Baple and Andrew Crosby previously discovered mutations in more than 15 genes that cause hereditary spastic paraplegia (HSP)—a group of rare inherited disorders characterized by leg muscle weakness and stiffness. Recently, the University of Exeter duo identified new variants in yet another gene called *TMEM63C* in seven patients from three affected families.

Nobody knew what the TMEM63C protein did inside cells, but the researchers had a hunch. Their previous work suggested that cellular pathways involved in processing fat molecules may be a common factor underlying HSP and related motor neuron diseases. "Lo and behold," says Baple, the TMEM63C protein was located "exactly where we'd expect to find it if it was involved in those lipid metabolism processes": in the endoplasmic reticulum (ER), the cellular hub for protein packaging, as well as at sockets where the ER joins with mitochondria to exchange lipids. Both organelles became misshapen in cells engineered to lack TMEM63C, which also suggests that the protein helps shape organelle morphology.

The next step, Baple adds, is to confirm if, and how, the protein is involved in lipid processing, and if cells from people with HSP are deficient in the protein. In the meantime, identifying these gene variants "brings immediate diagnostic benefits" to affected families, Crosby tells *The Scientist*.

Kishore Kumar, a neurogeneticist at the Garvan Institute of Medical Research in Australia, says the discovery is "one more piece in the [HSP] puzzle" that could help diagnose previously unexplained cases. More than 80 mutated genes have been identified in HSPs, making this one of the most genetically heterogenous inherited diseases. Kumar says larger cohorts of patients should be screened to determine how frequent *TMEM63C* mutations are. They're likely to be fairly rare, he says, "but we just don't know yet."

—Clare Watson

FATAL FUNGUS: A dead Peron's tree frog (*Litoria peronii*) infected with a chytrid fungus.

EDITOR'S CHOICE IN ZOOLOGY

Deadly Plasticity

THE PAPER

M. Torres-Sánchez et al., "Panzootic chytrid fungus exploits diverse amphibian host environments through plastic infection strategies," *Mol Ecol*, 31:4558–70, 2022.

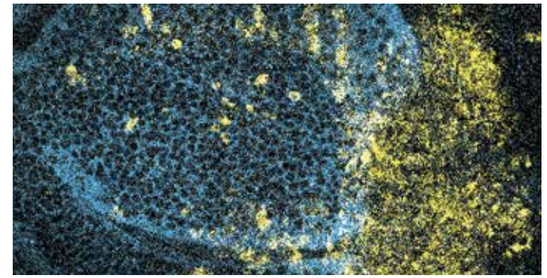
Since the 1970s, the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) has spread globally amongst amphibian populations, wiping out entire species and decimating others. Yet while the pathogen, which infects an amphibian's porous skin and disrupts gas and water exchange, is deadly and ubiquitous, some species are more susceptible than others. Past studies have focused on animals' immune responses to *Bd* infection, but not on how the fungus might be adapting to different hosts. "It was not clear if the fungus itself was doing the same thing in the different species it is infecting," says María Torres-Sánchez, a postdoc at the University of Florida.

To find out, Torres-Sánchez took datasets from those early experiments and turned them on their heads, looking instead at what genes the fungus was expressing on the skins of different amphibian species with varying susceptibility to *Bd*. She and her colleagues compared the transcriptomes of *Bd* growing on 14 species of frogs, newts, and salamanders, and of *Bd* grown on plates without a host.

While the fungus maintained a consistent set of housekeeping genes, the team found that *Bd* tailored the expression of other genes to each host, allowing it to pursue multiple infection strategies. For example, in more-vulnerable species, genes essential for attaching to and invading leukocytes, cells that defend a host from pathogens, were upregulated. In more-resistant species, genes promoting quicker reproduction, perhaps to evade or overwhelm a host's defenses, were elevated.

The results are "really exciting," according to Amy Ellison, a molecular parasitologist at Bangor University in Wales who was not involved with the study. The list of differently expressed genes could provide "interesting targets" for further studies looking at the mechanism of *Bd* infection, Ellison adds, or in "identifying populations of amphibians that might be more at risk" for severe disease.

—Tess Joosse



STRESS DEATH: Cells engineered to downregulate Gr64 expression experience cell death via apoptosis (shown in yellow), while control cells where Gr64s were not manipulated (shown in blue) persist.

EDITOR'S CHOICE IN CELL BIOLOGY

Taste of Survival

THE PAPER

M.E. Baumgartner et al., "The Gr64 cluster of gustatory receptors promotes survival and proteostasis of epithelial cells in *Drosophila*," *PLOS Biol*, 20:e3001710, 2022.

While combing through a list of genes that are differently expressed in *Drosophila* cells with and without certain ribosomal mutations, researchers in developmental biologist Eugenia Piddini's lab at the University of Bristol stumbled upon a surprise. A cluster of genes encoding six receptors known as gustatory receptors 64 (Gr64s) was upregulated in the epithelial cells of mutant larvae, which experience cellular stress as a result of the buildup of misfolded or otherwise dysfunctional proteins. It was an "intriguing and serendipitous" find, Piddini says, as Gr64s sense sugar molecules in adult flies but had no other known functions.

Piddini and her colleagues decided to investigate further, knocking out Gr64 function in larval epithelial cells containing the stress-inducing mutations. Losing the taste receptors resulted in "a spectacular amount of death" among the stressed cells, Piddini says, hinting that the receptor cluster might somehow be involved in cellular homeostasis. Indeed, the team found that the loss of Gr64s prevented the cells from digesting aggregated proteins via autophagy compared to cells with functioning receptors.

In their typical role as taste receptors in sensory cells, Gr64s also oversee calcium flow, which is involved in protein regulation. The researchers imaged calcium influxes into the mutant cells and found less activity in Gr64-free cells compared to controls, making calcium signaling a likely candidate for how the receptors might maintain proteostasis, Piddini says.

The study is "intriguing," says Craig Montell, a neurobiologist at the University of California, Santa Barbara, who studies gustatory receptors and was not involved with the research. He adds, however, that the authors didn't fully connect calcium activity to protein regulation. This is also top of mind for Piddini, who says the calcium activity "is a very important unknown for the proper understanding of how this receptor functions."

—Tess Joosse

Revising the Genome

A biotech startup called Tessera Therapeutics has made a splash with its claims to have developed "Gene Writing" technology. Is the excitement justified?

BY DAN ROBITZSKI

For the past few years, a biotech company called Tessera Therapeutics has been working away with a singular, lofty goal in mind: revolutionizing the technology used to edit DNA and RNA for scientific or therapeutic purposes.

The company is developing a suite of technologies it calls Gene Writing, all intended to expand the range of possible insertions, deletions, or edits that can be made to genetic material, while reducing the number of off-target alterations produced by more-traditional methods such as CRISPR-Cas9. The company claims that its techniques to "rewrite" or "write into" the genome without cleaving the DNA molecule, will help usher in a new era of highly effective, specific, and mechanistically simple gene editing. However, Tessera has shared precious little data with the world, and the approach that it's championing is riddled with technical hurdles, raising questions among some researchers about whether the company can deliver what it's promised.

Since CRISPR-Cas9 gene editing was first described in 2012, scientists have grown increasingly competent at editing, inserting, or deleting specific stretches of an organism's DNA. In the early years, the most tried-and-true approach was to harness the abilities of natural or engineered nucleases—such as various Cas enzymes, transcription activator-like effector nucleases (TALENs), and zinc fingers—to break both strands of DNA's signature double helix. Such breaks can be difficult for the cell to repair, however, leaving the genome vulnerable to errors in addition to the alterations researchers want to make. Newer and gentler approaches have since emerged in which the brute force of a nuclease's double-strand breaks is replaced by single-strand breaks (also known as nicks) or sometimes no breaks at all, greatly increasing the efficiency and precision of gene edits.

Some successes of this new generation of genome-editing technologies include prime editing and base editing, which involve nicking DNA with a Cas nuclease and relying partly on the cell's own DNA repair machinery to make precise changes. But researchers are limited in the types of changes they can make using these technologies—base editing, for example, can currently only substitute purine bases for other purines or pyrimidines for pyrimidines. Tessera instead plans to use mobile genetic elements (MGEs)—stretches of genetic material that are thought to make up half the human genome and are capable of moving around that genome without making double-strand breaks. In theory, explains company cofounder and board chair Geoffrey von Maltzahn, this could allow researchers to swap any individual base pair for any other.

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For all of its investor interest, Tessera remains something of an enigma to researchers within both academia and industry.

It's far from the only company to try to make this technology a reality. Massachusetts-based SalioGen Therapeutics, which closed a $115 million Series B financing round in January, is testing an MGE-based gene therapy platform for clinical applications that it calls Gene Coding. And Integra, based in Barcelona, Spain, has conducted preclinical tests of a similar MGE system, also dubbed Gene Writing, first described last December in *Nature Communications*. In March, Integra completed a roughly $6.3 million seed round of government and private funding. "We are continuously improving the technology," says Marc Güell, cofounder and chief scientific officer of Integra Therapeutics. "Last year we had the year of base editing, prime editing. . . I think this year will be the year of gene writing."

Yet Tessera seems to have stolen the spotlight. Spun out of the biotech venture capital company Flagship Pioneering in 2018, the company issued a press release about its trademarked Gene Writing technology—a trio of techniques that von Maltzahn says all use MGEs—in 2020. "At the outset of Tessera . . . we didn't have clear perspectives as to how fruitful this endeavor was going to be," von Maltzahn tells *The Scientist*, adding that the team shared a belief "that putting a lot of chips in the exact same place that Mother Nature put a bunch of chips was probably a worthwhile endeavor to do."

Tessera placed its chips and a windfall of investor cash followed. Seven months after raising $2.7 million in a 2020 seed round, Tessera collected another $230 million in a January 2021 Series B, with Flagship increasing its financial stake alongside outside contributions. Earlier this year, Tessera completed its Series C, raising more than $300 million of additional funding, much of which came from the same cadre of investors. At the time, von Maltzahn told *FierceBiotech* that the company would continue "aggressively investing" in its platform and hoped to have operational technology by October 2023.

As is the case with many privately held companies, it is true that we have not yet shared much of our data publicly.

—Geoffrey von Maltzahn, Tessera Therapeutics

That funding has helped the company recruit skilled scientists who are well-regarded in the gene editing community. Dana Carroll, a University of Utah molecular biologist who helped pioneer early nuclease-based editors and previously licensed that tech to Sangamo Therapeutics, tells *The Scientist* that he recognizes a few of the scientists on Tessera's team and that he respects their earlier work. "They got some experienced entrepreneurs in the biotech space, they've got some good scientists on board, and they got a lot of money," Carroll says. "So, if they have some good ideas, the prospects may be bright."

However, for all of its investor interest, Tessera remains something of an enigma to researchers within both academia and industry who spoke to *The Scientist*. The company has yet to publish peer-reviewed research or a white paper on its technology. Instead, it has primarily communicated progress through the occasional conference presentation, an approach that has caught the attention of prominent members of the community but left many questions unanswered—and some scientists skeptical.

"They've been on all of our radars through the grapevine," University of California, San Diego, gene editing researcher Alexis Komor—a coauthor on the 2016 *Nature* paper first describing base editing and a former consultant at Beam Therapeutics, which commercialized that technology—says of academics' perspective on Tessera. "We're always like, 'What are they doing, exactly?' But no one actually knows. We're all very interested."

Three Gene Writers

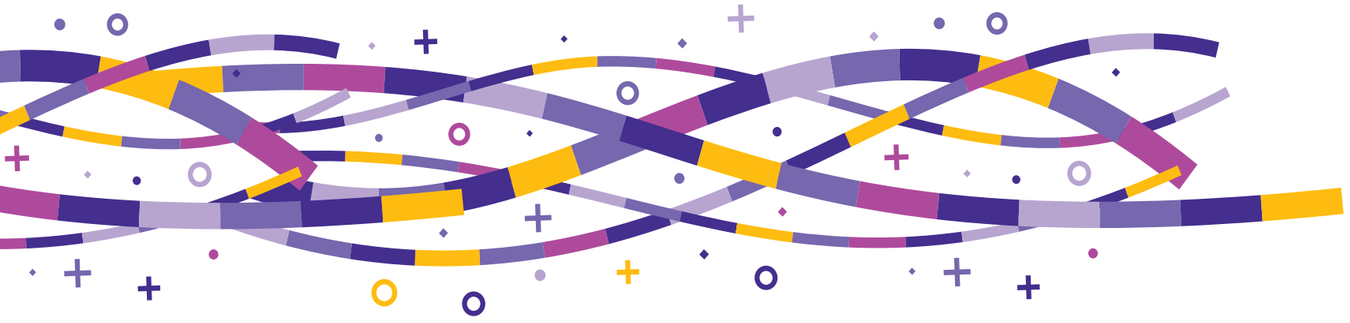
Von Maltzahn tells *The Scientist* that Tessera is simultaneously developing and testing three categories of Gene Writing systems—DNA Gene Writers, RNA Gene Writers, and RNA Gene Rewriters—that are delivered to the target site inside a lipid nanoparticle. The company hopes the trio will ultimately form a comprehensive platform allowing for a wide variety of alterations ranging from single-base-pair substitutions to the insertion of entire genes.

For small alterations, there are the RNA Gene Rewriters, derived from MGEs called retrotransposons that essentially copy and paste their own sequences into new locations of a genome by means of an RNA intermediate and reverse transcription. Specifically, Tessera's Rewriters use a process called target-primed reverse transcription, von Maltzahn explains, which involves writing the payload DNA sequence into a desired location by nicking one DNA strand at the target site, delivering an RNA template transcribed from the retrotransposon sequence, and assisting the cell's transcription machinery in the reverse transcription of a complementary DNA strand. It's these Rewriters that von Maltzahn says can outperform base editing by performing any kind of base pair substitution. They can also make substitutions, insertions, or deletions of up to roughly 100 base pairs in length, he says.

The other two systems are both designed to perform whole-gene insertions, vastly expanding the lengths of DNA that can be added by current cutting-edge techniques such as prime editing, which struggles to insert sequences longer than a few dozen base pairs. DNA Gene Writers are derived from recombinases, enzymes involved in the mobilization of MGEs that genetic engineers have used for decades and that are capable of rearranging or combining DNA sequences. With its own technology, Tessera aims to provide a DNA template that the recombinase would use to overwrite the DNA sequence at the target site via reverse transcription after inducing a single-strand break.

Then there are the RNA Gene Writers, which like the RNA Gene Rewriters are derived from retrotransposons, but which are capable of much longer insertions thanks to differences in the specific RNA template–encoding enzymes they contain. They include an RNA template and the RNA for that retrotransposon-encoding enzyme. "The RNA Gene Writer protein goes to the genome [and] nicks one strand, which leads to local unraveling, which leads to the hybridization of RNA bases to that location," von Maltzahn says. "One letter at a time, it will write that RNA into DNA."

Von Maltzahn compares Tessera's technology to existing CRISPR transposase systems that combine programmable MGEs with nucleases, as well as to *piggyBac*. The latter is a



transposase-based gene delivery system that's proven useful for in vitro and in vivo applications, but historically has run the risk of inserting DNA at unintended locations. Von Maltzahn also points to similarities between Tessera's systems and naturally occurring transposons, some of which can insert their payload nearly anywhere in the genome. Like *piggyBac*, such transposons show a lack of specificity in insertion sites, and so have proven difficult to harness for gene editing therapies.

Part of the team's work, von Maltzahn says, involves "brute force" testing of thousands of candidate MGEs to find sequences that precisely bind and deliver their payload to specific DNA sequences of interest—and then finding a way to reprogram them to deliver a desired or therapeutic payload sequence rather than whatever sequence they'd evolved to shuttle in nature. "Our endeavor at Tessera has been to continuously expand on top of the natural resource [that is] MGEs. We've tested over 20,000 MGEs inside of human cells at Tessera to date," he says.

Initially, von Maltzahn tells *The Scientist* over email, Tessera was working with MGEs for RNA and DNA Gene Writers that more or less randomly insert their payload. However, the company has since managed to replace "the native DNA binding domain with a synthetic DNA binding domain," which he says has allowed them to more precisely target desired integration sites. With the company's RNA Gene Writers, "[a]s far as we know, we are the first to have shown that whole genes can be written into the genome merely by introducing two molecules of RNA into human cells," he adds, clarifying that human cell testing is rare for the industry.

"I think large gene insertion can be interesting for a variety of diseases," especially loss-of-function diseases such as spinal muscular atrophy, Duchenne muscular dystrophy, or cystic fibrosis, says Alexandra Urman, an analyst who covers therapeutics and the gene editing industry at ARK Investment Management. "And it's a challenge that's plagued gene editing in general." If Tessera can figure out MGE gene editing that is specific, efficient, and programmable—and back up its claims with solid evidence—"that would be very interesting," she adds.

Accumulating Critiques

According to von Maltzahn, the company has kept its head down to work on developing and automating its technology—specifically running tests in human cells and animal models—but plans to publish in the future. Tessera shared very little data or findings on the performance of its trio of technologies with *The Scientist*, with von Maltzahn saying that the company has "prioritized filing intellectual property over scientific publications thus far." The company did share a PowerPoint presentation that its scientists gave at the Precision Genome Engineering Keystone Symposia in April, describing how the company screened for MGEs that could target and overwrite mammalian genes and tested them in human cells.

Tessera is simultaneously developing and testing three categories of Gene Writing systems—DNA Gene Writers, RNA Gene Writers, and RNA Gene Rewriters.

Tessera also shared its presentation from the Federation of American Societies for Experimental Biology (FASEB) Genome Engineering Conference in June, in which the company provided some preliminary, preclinical information on its Gene Rewriters—a rare disclosure of specific data from a company that's otherwise remained tight-lipped. The presentation suggested that Tessera could perform individual thymine-to-adenine conversions in target alleles with roughly 65 percent efficiency—20 times the rate at which it performed unintended or errant edits.

Still, the dearth of data and publications has raised more than a few eyebrows, as well as questions about whether Tessera's technology will live up to the hype generated by the company's financing. Urman, for example, expressed some hesitation about Tessera's technology due to the lack of publications supporting its work. She tells *The Scientist* that she feels similarly to the way she did when writing a 2020 newsletter for ARK. Back then, she wrote: "Now Tessera Therapeutics . . . is claiming to write DNA without breaking it, something impossible to date without off target effects. Tessera uses mobile genetic elements (MGEs) to move or copy DNA into new locations, another technology that is not new." She continued: "We believe other technologies will have to be supported by strong patents and data before they can compete [with any gene editing technology]."

Carroll and other researchers emphasize in particular the complexity of the challenge Tessera has taken on. Programming a therapeutic payload, combined with finding or programming MGEs to target a DNA sequence that's specific enough to prevent MGEs from making the same edit throughout the human genome, has historically proven to be an all-but-insurmountable hurdle, experts tell *The Scientist*. Some MGEs "will just integrate [their payload] randomly throughout the genome everywhere," says Komor. And even some of the more specific retrotransposons out there can still integrate into thousands of different genomic sites—far too imprecise for a therapeutically useful delivery system. Retrotransposons essentially "find a particular sequence and insert the RNA or DNA" on the spot, Komor explains. "And so, if that sequence happens to be the exact location of the genome where you want to do your editing, then that's great. Chances are it's not."

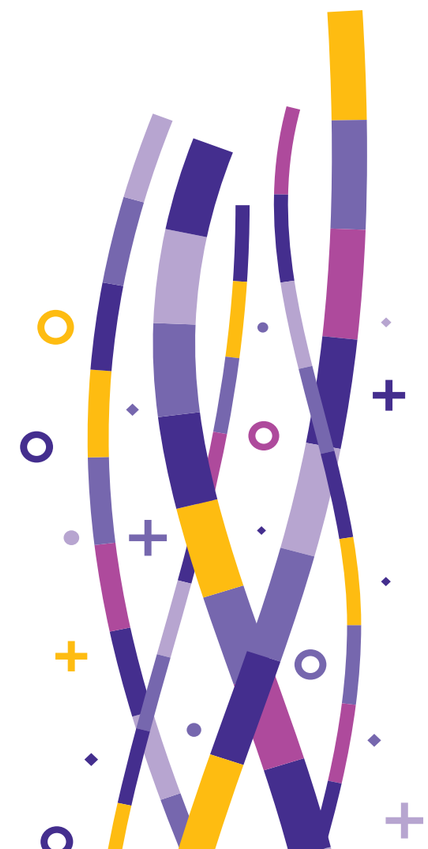
Carroll notes that previous experiments with transposons by various researchers have found that "the enzymatic activity and the recognition activity are tightly linked within the proteins. So if you try to add some specificity by engineering, you'll often decrease the efficiency. And if you try to increase the efficiency of it, you'll decrease the specificity." He also adds that those researchers typically avoided the headache by linking the transposon to a precision-boosting nuclease such as Cas9. "It has turned out to be extremely challenging and no one as far as I know has made it work," Carroll says of the myriad attempts to use MGEs as gene editors, adding that he is hesitant to comment on Tessera's research because of how little is known and because he hasn't seen the company "give any references to the literature" that their work builds on.

Even when the audience at Tessera's presentation at the Keystone Symposia pressed for more information on how—and how well—Tessera's platform worked, they were left without many details about how the company was going to get around the issues associated with MGE editing, says Komor, who attended. "The talk at the conference was really exciting," she says. "It was the first time we were going to see data." However, she adds, very little information was presented, and "they had no actual information about the actual editor aside from that it uses RNA as a template."

The presentation slides, shared with *The Scientist* by Tessera, do indicate that the company ran into problems with imprecise integration and had experimented with using Cas proteins to improve target site specificity. This use of Cas proteins is also a feature of existing gene editing techniques such as prime editing and, in an email to *The Scientist*, von Maltzahn refers to papers showing high site specificity in prime editing as a proof of concept for Tessera's Gene Rewriters, since both use target-primed reverse transcription. In a follow-up email, von Maltzahn clarifies that Tessera's technology differs from prime editing because, even though prime editing and Gene Rewriters both rely on a similar mechanism, "there are differences in the molecular machinery itself in both the template RNA and enzymatic protein" between the two technologies. Von Maltzahn additionally claims that Gene Rewriters can insert considerably longer stretches of DNA.

As for criticisms regarding a lack of information about Tessera's technology, von Maltzahn responds: "As is the case with many privately held companies, it is true that we have not yet shared much of our data publicly, and this may limit the ability of some researchers who are not affiliated with Tessera to share informed opinions on our work."

Nevertheless, even the more skeptical scientists who spoke to *The Scientist* say they remain hopeful that the challenges of MGE-based genome editing are on the way to being solved. After all, "nobody's been able to get [CRISPR transposases] to work really well in mammalian cells," Komor says, so it would be an exciting feat if Tessera has overcome the challenges associated with this or related approaches. Urman adds that if scientists believe in the promise of a technology, a proliferation of studies that use or validate it will follow in the next few years, as it has for CRISPR, base editing, and prime editing. Perhaps, she suggests, that phase for Gene Writing is still to come. ■



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Growth of the Green Lab

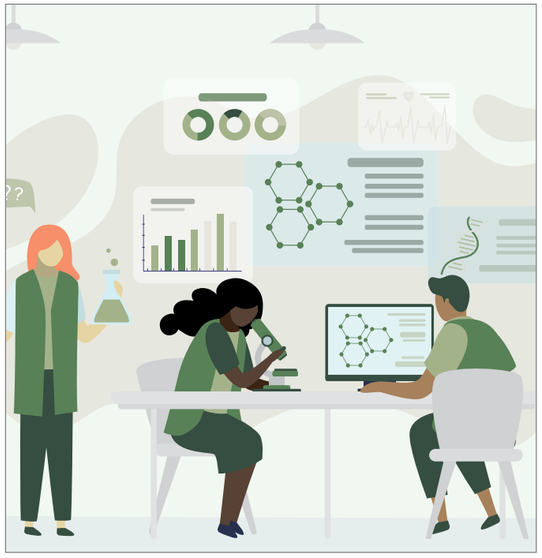
Across the world, scientists, students, and administrative staff are working to mitigate research's impact on the environment.

BY NATALIA MESA

During the pandemic, postdoc Julie Sesen started to feel overwhelmed by the amount of plastic used by the scientific community. Sesen studies pediatric tumors and cerebrospinal disease at Boston Children's Hospital (BCH), where in 2020 many researchers were testing the community for COVID-19. Every day, researchers there and at scientific centers across the country inevitably threw away hundreds of single-use masks and plastics. Everyone she spoke to agreed that the volume of plastic waste was an issue, she says. "And we needed to do something about it."

Even before the pandemic, researchers worldwide produced several million tons of plastic waste per year, according to some estimates. But when Sesen looked into how she could recycle the waste she was generating in her lab, she learned that plastic waste was only part of a larger problem. Scientists also use massive amounts of energy, exacerbating pollution and contributing to climate change. Various estimates indicate that a research lab can consume more than three times as much energy as an office of the same size. Common equipment such as fume hoods and ultra-low temperature freezers can consume as much as an average household. So when Sesen discovered My Green Lab, a nonprofit that seeks to help scientists improve sustainability in research labs, she soon joined their Ambassador program, which provides free online courses on sustainable science. She and another postdoc went on to found BCH Greenlabs, an initiative to support other research groups at the institution in reducing their carbon footprint and plastic waste.

The huge environmental impact of laboratory research has led many other institutions to try to make research eco-friendlier, too. There are now hundreds of such programs around the world, developed either in-house or in partnership with organizations such as My Green Lab. Some initiatives, like the one at BCH, are mainly run by volunteers, typically students and postdocs, while other institutions have a sustainability office overseen by one or more paid specialists. Taking advantage of the increased uptake of sustainable lab practices, advocates are now taking the opportunity to push for larger, systemic change. While the COVID-19 pandemic has hindered some of these efforts, it has also motivated people to do more, researchers tell *The Scientist*. In a handful of countries, sustainable practices may even soon be tied to grant funding, notes Anna Lewis, a sustainable science manager at the University of Bristol in the UK, making a green approach an integral part of life sciences research.



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"The momentum is incredibly good right now. . . . We're seeing an explosion of green labs," says My Green Lab CEO James Connelly. "But we do need those systemic levers [for science] to be part of the climate solution and not part of the climate challenge."

A green wave

Although most researchers are open to adopting greener laboratory practices, the "scientific industry as a whole has been a bit slow to address climate change," says Connelly. Indeed, when My Green Lab first started in 2013, it had partnerships with only 10 schools. Now, it works with more than 1,000 labs in 36 countries, including several biotechnology companies. The organization offers a voluntary certification process in which researchers assess a lab's current energy usage, equipment usage, and chemical and waste disposal system. They send these assessments to My Green Lab, which makes suggestions for improvement. "These are low-cost or zero-cost things that any researcher can do to help improve the sustainability of scientific research that also don't undermine or interfere with . . . research," says Connelly. Simple suggestions include actions like closing the fume hood sash, which can reduce energy consumption by up to 30 percent, and setting ultra-low temperature freezers to –70 ˚C instead of the standard –80 ˚C, which can reduce the appliances' yearly energy consumption by 30 percent.

Another popular certification scheme is the Laboratory Efficiency Assessment Framework (LEAF), which was developed at University College London and is now used at several schools in the UK. Schools that adopt it typically have a centralized office to help implement LEAF, but it's up to individual labs to opt in. Like many other sustainability initiatives, LEAF mainly focuses on life science laboratories because they "have a lot of common, energy-intensive equipment," says Lewis, who helps oversee LEAF at Bristol. Most of the guidance encourages behavioral changes, such as remote participation in conferences, reusing solvents, and cataloging chemicals and samples to avoid over-purchasing supplies. Although participation is voluntary, 100 percent of Bristol's 1,000 laboratories have adopted LEAF, Lewis says.

Increasingly, green life science efforts are being seen as part of larger, institute- or municipality-wide commitments to improving sustainability.

Some institutions have launched their own sustainability programs. The University of Colorado Boulder started its initiative, CU Boulder Green Labs, back in 2009. Program manager Kathryn Ramirez-Aguilar says that the effort initially focused on "energy savings, water savings, waste diversion, [and] scientists' engagement," which meant getting individual labs to change their practices. More-recent initiatives include university-wide equipment sharing programs, which Ramirez-Aguilar says has not only saved energy and reduced unnecessary purchases but improved equal access to resources. She says she hopes that in the future, CU's lab startup packages will include access to this shared equipment.

Increasingly, green life science efforts are being seen as part of larger, institute- or municipality-wide commitments to improving sustainability. The University of California (UC) system, for example, has partnered with My Green Lab as part of its pledge to achieve carbon neutrality by 2025, and UC has made it a goal to certify three laboratories on every campus under its umbrella by the end of this year. Similarly, the University of Bristol adopted LEAF to reach a 2030 carbon neutrality target, set after the city of Bristol passed a resolution in 2015 to hit a similar target.

Because sustainability programs are largely voluntary, it's difficult to know exactly how many labs at certain institutions have adopted green practices. Similarly, because academic institutions are large and labs may share building space, it can be hard to track how much energy purely behavioral initiatives save.

Still, the organizers of many green lab projects say they've saved energy and diverted waste from landfills. LEAF's pilot program, which took place from 2018 to 2020 at 23 universities, reportedly saved 648 tons of carbon, the equivalent of taking 140 passenger vehicles off the road for those two years. Recently, the University of British Columbia's Michael Smith Laboratories (MSL), a group of more than 300 researchers, participated in UBC's Chill Up Challenge, its version of the Freezer Challenge competition organized by My Green Lab and the International Institute for Sustainable Laboratories (I2SL). The University saved 45,000 kilowatt hours of electricity in a year, equivalent to the annual usage of four single-family homes. And since 2009, CU Boulder's Green Labs program has saved 9.1 gigawatt hours of energy (equivalent to $1 million), says Ramirez-Aguilar, as well as conserved 61 million gallons of water and diverted 376,000 pounds of waste.

Obstacles to sustainability

Overwhelmingly, sustainability coordinators and volunteers say that scientists are enthusiastic about making their research more environmentally friendly, even though they sometimes lack the tools and know-how to do it. But some also say that getting busy scientists to take action is a challenge. "Even if scientists are aware of the environmental impact of research . . . there's this sense that 'There's nothing I can do about it' or 'This science is too important that the environmental impacts are worth it,'" says Connelly. In addition, not all schools have the resources to put these programs into practice. "The challenge is funding. I don't really have funding for large-scale changes," says Carrie Metzgar, a sustainability and planning analyst at UC Irvine.

Costs can accumulate in various ways. For example, some changes require support from technical staff and environmental health and safety experts, all of whom are ideally also trained in sustainability. At the University of Bristol, Lewis says that technicians, who are knowledgeable about how to adapt their lab's protocols and practices, provided the necessary support for Bristol to reach 100 percent LEAF certification. But she also admits that not all schools have this technical support.

Many waste mitigation strategies, not to mention equipment procurement and replacement, are more than scientists can do on their own, and some are costly up front. Recycling can be an especially difficult organizational task, as many research products must be recycled outside the municipal waste stream. For example, there are only three companies in the US that recycle single-use nitrile gloves, a laboratory fixture.

Similarly, while many biotech companies have begun taking back plastic waste, "the problem with that is often it's restricted just to the items that they have sold you. That means the lab needs to have multiple lab plastic bins in the lab for different suppliers," says Andrew Arnott, a climate strategy, biodiversity and sustainability manager at the University of Edinburgh. He adds that Edinburgh is attempting to incorporate recycling of these items into the municipal waste stream.

In some cases, thinking long-term may help universities provide labs with the needed up-front costs. The University of Bristol, for example, has a fund to replace laboratory equipment with energy-efficient models, giving researchers the amount of money that they would save over the course of seven years. And they're not alone; many other universities have similar programs that help laboratories buy green equipment.

Systemic change

Despite the growing enthusiasm for green labs, some proponents still say that they are working within a system that deprioritizes sustainability. Some behaviors like plastic use remain entrenched in scientific practice and continue to increase. Making sustainable laboratory practices standard in research laboratories will require incentives for researchers and companies to divert waste and save energy. "The climate crisis requires us to spend money on things which won't necessarily give us a quick payback," says Arnott.

One way to get the whole scientific enterprise more involved in sustainability may be to tie grant funding to green practices, says Ramirez-Aguilar. "I find that scientists want access to sustainable products and supplies," she says. Prioritizing sustainability when allocating research funding "can drive the [systemic] changes needed." In the UK, some grant funding agencies are already welcoming sustainability statements in grant applications. UK Research and Innovation (UKRI), the UK's national science funding agency, has expressed interest in adopting LEAF as a standard for laboratory sustainability and incorporating it into grant decisions, says Lewis. "Green lab certification, LEAF for example . . . is very likely to be linked to grant funding opportunities in the next year or so," she says. UKRI did not respond to a request for comment.

The US may soon follow. In October, the Department of Health and Human Services released its Climate Action Plan, which listed enacting sustainable grant policies as one of its priorities. So far, this hasn't translated into any changes in the grant application process for any federal funding agencies, although National Institutes of Health (NIH) spokesperson Elise Rabin tells *The Scientist* in an email that the agency is "aware of the Department of Health and Human Services' (HHS) climate goals as outlined in the HHS 2021 Climate Action Plan. . . . While NIH awaits further direction from HHS, it has been reviewing NIH policies to see how we can achieve the objectives identified in the plan." The NIH also says it supports green science initiatives and runs its own green labs program. HHS did not respond to a request for comment.

To push for faster change, My Green Lab and the I2SL have sponsored an effort called Million Advocates for Sustainable Science, a letter-writing campaign to funding agencies requesting that they do their part to promote sustainability in research. Martin Howes, the assistant carbon manager at Cambridge University, says he hopes that researchers won't view and talk about sustainability and research as separate issues for much longer. "We've long had a strong safety culture. Sustainability needs to be the next one of those needs to integrate with safety culture and best practice." ■

Natalia Mesa is a freelance science journalist based in Seattle.

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SIMPLE TIPS FOR A MORE SUSTAINABLE LAB

The Scientist assembled advice from sustainability experts on how to reduce your research's environmental impact

Chill out: Regularly service your ultra-low temperature freezer and increase the temperature, if possible. Many samples can be held at –70 ˚C as opposed to the current standard default temperature of –80 ˚C.

Shut off: Power down equipment when not in use and set up timers on lights so that they're not running all night.

Shut up: A single fume hood can consume as much energy as 3.5 homes. Shutting the sash on your fume hood can save up to $9,100 each year.

Recycle: Reach out to your facilities manager to learn more about how you might partner with companies that recycle common single-use items such as gloves, masks, and pipette tips and boxes.

Take stock: Regularly checking what your lab already has in stock can prevent unnecessary purchases.

Share it: Set up sharing programs with laboratories nearby to avoid purchasing unnecessary equipment and improve equal access to equipment.

The Conservation Power of Animal Creativity

When species disappear, more than their genomes are lost. Their potential to benefit ecosystems through innovation vanishes as well.

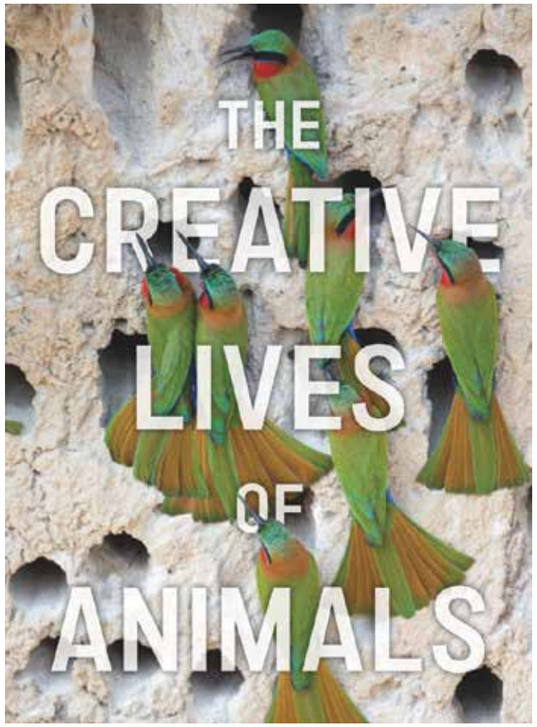
BY CAROL GIGLIOTTI

To some people—road engineers, for instance—beavers and their dams may seem like the ultimate foe of human progress. But to the scientists who study them, beavers exemplify animal creativity. In a recent study on methods for rewilding freshwater wetlands, researchers found that the reintroduction of beavers as ecosystem engineers often creates unique habitats that benefit biodiversity at numerous spatial scales. Importantly, beavers actively creating and maintaining their ponds also produces aquatic habitats superior to those that are human-made. In other words, by exercising their unequaled creativity, beavers benefit not only themselves, but myriad other species, large and small, that share their ecosystems in ways humans simply cannot accomplish.

The beaver, at once a potential solution to biodiversity loss and a troublesome force acting against the goals of human development, illuminates our conflicted relationship with the approximately 2.1 million other animal species who share our planet. If we understand that nonhuman animals—and not only beavers—also have inherently valuable skills, unique to individuals and to species, might we widen our tunnel vision to see them as collaborators and guides in conserving their habitats and biodiversity?

Although I have a computer science and philosophy background, I am essentially an artist. My long career of teaching not only traditional visual art students but those studying computer animation programming, graphic narratives, design, interactive media, and video has offered me access to primary research on how the creative process works. It was obvious to me that creativity existed in many domains, not only in the arts, and across individuals of all cultures. I became interested in animal creativity when working on my edited book, *Leonardo's Choice: Genetic Technologies and Animals*. What do we lose, I wondered, when we genetically modify animals to suit our needs?

In 2004, I learned of a 2003 book, *Animal Innovation*, edited by biologists Simon Reader and Kevin Laland, and an article by the biologist Allison Kaufman and the psychologist James Kaufman titled "Applying a Creativity Framework to Animal Cognition," both of which encouraged me to continue this line of investigation. As I did so, research on animal creativity was blossoming, much of this interest coming from scientists who were spending years in the world of a particular species. Interviews with some of these researchers and the published research of many more support the ideas in my latest book, *The Creative Lives of Animals*.



NYU Press, November 2022

In the book, I define creativity as a dynamic process in which novel and meaningful behaviors are generated by individuals with the possibility of affecting others at cultural, species, and evolutionary levels. That is, individual animals are creative in unique ways that influence their culture, and that accumulated creativity may have an evolutionary effect on biodiversity. Both domestic animals and those who live as part of larger communities, such as ants or bees, express creativity.

Being open to the possibility that creativity exists across species requires open minds, a willingness to see behaviors in a new way, and a comfort with complexity.

I emphasize the intricate workings of creativity for several reasons. We must be able to appreciate the sometimes complex and iterative processes by which an animal solves problems or achieves goals. This is complicated by the way the creative process often takes a zigzagging course, driven by ongoing exploration. Sometimes that pursuit produces an innovation, such as a song, a tool, or a dam. Sometimes, appropriation of another's work is a creative act. Indeed, beavers recognize human-made dams and modify them to meet their needs. This ability to recognize an opportunity is key to the creative process, and beavers demonstrate their flexibility and ingenuity in doing so.

At other times, nothing new seems to come out of the creative undertaking, but the behaviors involved may be new for the individual and useful in other facets of their existence, now or in the future. Sometimes that exercise may lead to more creativity. The process may not be visible to an outsider, existing only as a thought experiment.

Growing interest within the humanities and sciences in how the creative impulse works across many domains, not only in the arts, has fostered a reluctance to limit creative license to only a few special human individuals. The idea that creativity may be a common thread that runs throughout human activity has become accepted throughout the academy just as ideas about animal creativity are gaining traction in the biological sciences. Appreciating beavers for their contributions to biodiversity is not a hard sell among many biologists. But being open to the possibility that creativity exists across species requires open minds, a willingness to see behaviors in a new way, and a comfort with complexity. These qualities, the same ones often associated with creative behaviors, will assist humans in understanding that the creative agency of animals is a foundation of biodiversity. The world loses their genomes when species disappear, but what also disappears are creative pathways to saving ecosystems and habitats for all on this planet. ■

Carol Gigliotti is professor emerita of Dynamic Media at the Emily Carr University of Art + Design in Vancouver, British Columbia, Canada. Read an excerpt of The Creative Lives of Animals *at the-scientist.com.*

Cats as Sociological Bellwethers

Whether a feline is considered a pet or a pest depends not on what the animal does, but on what scientists and nonscientists alike believe about cats' place in the world.

BY BETHANY BROOKSHIRE

The internet seems built for humans to consume as much cat content as quickly as possible. Cats leap cup barriers and wear adorable outfits. Social media sites abound with heartwarming stories of people hearing the plaintive wails of a bedraggled kitten in the brush or in a storm drain, and transforming the sad, skinny waif into a beautiful, playful pet.

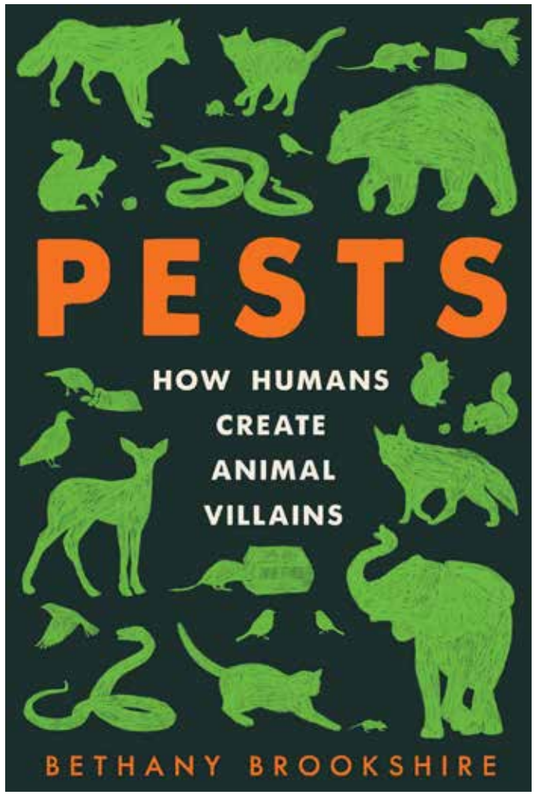
Cats—unsupervised and often unowned, either stray or fully feral—may also kill as many as 4 billion birds and 22 billion mammals every year in the lower 48 states. Many ecologists and conservationists consider cats invasive species. Harmful. Pests. In natural reserves and on islands, invasive cat populations are managed with the use of traps, shotguns, and even poisoned pellets carefully implanted in their potential prey. Anything to kill the killers.

Everyone, from the most fervent cat meme lover to an ardent wildlife conservationist, can agree—the world would be better with fewer stray and feral cats. How we get there, though, reveals a major cultural divide. In my soon-to-be-published book, *Pests: How Humans Create Animal Villains*, I investigate why some animals annoy us so very much, while others never seem to no matter what harms they cause. It's not a list of pests and their habits. Instead, it's a story of people—including the scientists who study the animals we love to hate.

One of the best examples of differences in how people view animals exists in the form of the house cat (my house, for one, is a temple to two very spoiled specimens). In a 2020 study, Kirsten Leong, a social scientist at the National Oceanic and Atmospheric Administration in Honolulu, and Ashley Gramza, a conservation social scientist then with the Arkansas Game and Fish Commission, investigated cultural models people had constructed around outdoor cats. They found a divide between wildlife managers and animal welfare groups. These jobs, like any jobs, have cultural expectations that come with them—beliefs and perspectives that lend people credibility in their profession. Scientists, for example, have cultural beliefs about matters such as author order and which scientific journals are the most valuable. The beliefs are often unspoken and are learned over time as one marinates in their particular field.

Conservation scientists and wildlife managers see their jobs as protecting natural spaces and the wild species that inhabit them. Their scientific understanding, and what they have learned to value, informs how they see the world and the animals that live in it. Any animal that threatens biodiversity is considered a harmful invasive species—and that includes cats. In this conceptual framework, an outdoor cat, particularly a stray or feral one, is an invasive pest. Then there are animal welfare groups. The cultural values that pervade the animal welfare community position stray and feral cats as homeless pets. These wayward felines represent an opportunity to be an animal's savior, and a potential source of love.

Both groups can agree that stray and feral cats are problematic. Animal welfare groups might promote a trap-neuter-return approach along with adoption or feeding and caring for outdoor colonies. Conservationists and wildlife managers, on



Ecco, December 2022

Many ecologists and conservationists consider cats invasive species. Harmful. Pests.

the other hand, might view colonies of neutered cats with horror, knowing that these cats are supplementing their cat food diet with hapless songbirds. For them, cat removal is the solution, whether via adoption, housing ferals in large enclosures, or euthanasia.

Neither of these views is wrong. But when the population of a country, city, or town fights over the designation of pet and pest, it can make it difficult to ensure there are fewer cats dining on the wildlife. If humans focus on this shared goal of fewer outdoor cats, it seems easy to achieve. In reality, though, cultural ideas and beliefs about cats—what we owe them and what they deserve—can complicate the practical solutions.

Scientists sometimes believe that pushback from the public results from a lack of information. They throw facts into what they perceive as a hole of ignorance, hoping that when they fill it, they will get the public to agree with their views. All too often they forget that disagreement isn't always about lack of knowledge. It's about experience, belief, and culture.

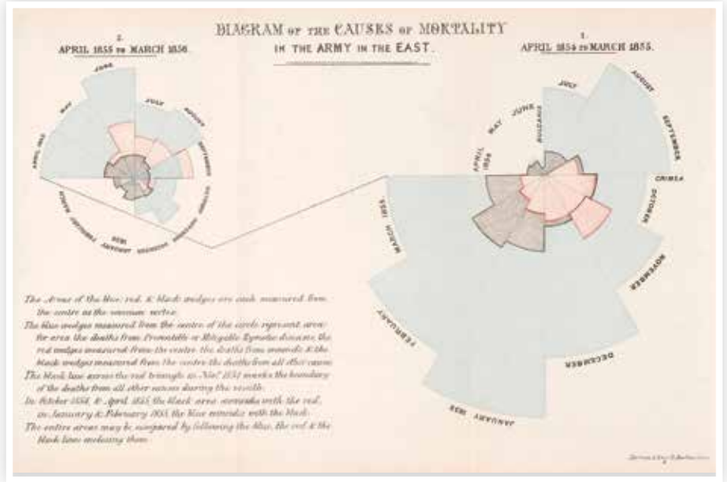
In the case of cats, conservation scientists often report the numbers of animals that cats affect, whether it's dozens of species pushed to extinction or billions of individuals killed. They hold up these vast body counts as evidence for feline misdeeds. But these facts don't always win arguments. They might horrify, but they might also make outdoor cat lovers defensive. *Their* cat, after all, would never do such a thing. On the other side, the scientist might get defensive too, asking how many birds Fluffy has brought home lately.

Coming to agreement on this and other complex conservation issues has to involve more than just fighting a war of facts. It's about understanding attitudes and culture. Not of the cats, but of the people, both those who love them and those who want to save wildlife from them. To deal with cats, we must first understand ourselves. ■

Bethany Brookshire is a science journalist and author of Pests: How Humans Create Animal Villains. *Read an excerpt of* Pests *at the-scientist.com.*

Diagrammatic Wars, 1858

BY ANDY CARSTENS



WELLCOME COLLECTION

DEATH WHEELS: In these rose-like diagrams, the numbers of deaths are proportional to the size of each wedge: pink wedges (inner) are deaths due to combat, blue wedges (outer) are due to disease, and grey wedges (middle) are deaths resulting from all other causes. The right and left diagram, representing before and after the launch of a widescale sanitation effort in March of 1855, respectively, are to scale, showing a drastic decline in mortality due to illness.

When nurse Florence Nightingale arrived at the British Army's hospital in Constantinople in 1854 to help treat soldiers wounded in the Crimean War, she immediately encountered squalor: nonfunctional sewer systems, vermin, and only a single bedpan for every 40 men. "The hospital was a chamber of horrors," says R.J. Andrews, a freelance data scientist. "This is why everybody [was] dying of communicable diseases," such as cholera, dysentery, and typhus.

When the war ended in 1856, Nightingale returned to London, worried that society's memory of these deaths would quickly fade, Andrews tells *The Scientist*. Before that happened, she intended to ensure that future conflicts would result in fewer needless deaths unrelated to combat.

To convince people that sanitation and mortality were inextricably linked, Nightingale waged a war of her own—one of information, fought primarily against the British government's War Office. The bureaucrats in charge there resisted change because, in their eyes, the infantry were "chattel," according to Andrews, whereas Nightingale's religious beliefs led her to view each individual as deserving of protection. Directing a team of lithographers, statisticians, and scientists, she often worked 20-hour days to create graphics that illustrated how improved sanitation would save lives, says Andrews, who edited a new book on the nurse's innovative data visualizations. "She was relentless."

Her most influential diagrams, published in 1858 and 1859, were part of a three-act story. Act one illustrated the problem: The death rate among British soldiers during the Crimean War was extremely high (roughly 23 percent). In act two, Nightingale and her colleagues showed that most of these soldiers died from disease, not combat. And finally, act three revealed the solution: A massive sanitation effort launched in the spring of 1855 to clean the Constantinople hospital caused death rates to plummet.

The second graphic in particular, called the "Diagram of the Causes of Mortality in the Army in the East" (pictured), caused a splash because its two circular figures were so unique, Andrews says. "We don't have a great name for what these are." In fact, one of the major criticisms of the chart was that its color-coded, differently sized wedges were too unconventional and, as a result, inaccessible. However, Andrews argues that nobody remembers the more typical bar or line graphs published by the War Office, whereas people are still talking about this one today.

While her work garnered media publicity, Nightingale herself remained anonymous as the campaign's orchestrator. Part of the reason, Andrews says, was that nameless pamphleteering was common, but perhaps more importantly, it was likely because she was a woman. "She's already really rocking the boat in terms of how much power a woman can hold in society," Andrews says, "So she has to be very careful about how she exerts that power."

In the end, her visualizations prompted real change. Reduced crowding and better sanitation during subsequent wars shrank disease-related death rates of British soldiers to below that of civilians. And in 1875, functioning sewer systems and access to clean water became part of Britain's Public Health Act, spreading Nightingale's reforms beyond military hospitals and into people's homes. ■

Multiplex Testing: A Solution to Manage Surge in Respiratory Illnesses as Concerns over "Multi-demic" Rise

Q: What should we expect during this year's flu season?

A: Over the past three years, we have been paying particular attention to personal hygiene such as frequent hand washing and the use of face masks. Due to reduced exposure and lowered immunity to respiratory viruses including influenza and colds during the pandemic, we have become more vulnerable to such illnesses.

The UK Health Security Agency says that emergency department attendances for acute respiratory infections and influenza-like illness have increased. According to the Centers for Disease Control and Prevention, early increases in seasonal influenza activity have been reported in most of the United States. South Korea has also seen a surge in influenza cases outside of their expected season. In addition, South Korea and the United States are both witnessing the early and fast spread of respiratory syncytial virus which usually causes mild, cold-like symptoms but can sometimes cause serious complications especially in very young infants and older adults.

Health officials are especially concerned about the early return of influenza in Australia this year. The intensity of the flu season in Europe and North America can often be predicted by the flu season in the southern hemisphere. This year, Australia experienced its worst flu season in five years. At its height in June, more than 30,000 cases were reported each week. Australian Influenza Surveillance Report shows that the most prominent flu strain during their winter was influenza A (H3N2), which is more likely to lead to severe morbidity and increased mortality than influenza B or seasonal A (H1N1) strains.[1]

It is expected that the circulation of respiratory viruses could return to pre-pandemic levels spreading widely at the same time, fueling concerns of a potential winter "multi-demic", which is why multiplex testing is more important than ever this year.

Eunsin Bae, M.D
Specializes in laboratory medicine and leads the Institute of Clinical Research at Seegene Inc. Her research focuses on microbiology, molecular biology, and hematology. Dr. Bae is currently working toward implementing a global clinical study and establishing an international network of clinical investigations.

Q: What is multiplex testing?

A: Multiplex testing in molecular tests refers to PCR tests that simultaneously detect multiple pathogens in a single reaction with one sample. While most singleplex PCR tests for respiratory infection only detect a single pathogen such as COVID-19 or Influenza A and B, multiplex PCR tests for respiratory infection can detect and differentiate COVID-19, influenza and respiratory syncytial virus using a single tube. However, some will extend beyond that to include adenovirus, parainfluenza virus, human rhinovirus and metapneumovirus.

About Seegene's Allplex™ SARS-CoV-2/FluA/FluB/RSV Assay

The Allplex™ SARS-CoV-2/FluA/FluB/RSV Assay detects six targets associated with the four respiratory viruses. For the detection of COVID-19, three distinct genes (S, RdRp, N) are included as targets to reliably identify positive cases, even as new variants emerge. The test also includes two internal controls for proper sampling and testing validation. The product can deliver results within two hours when extraction-free, making it both time and cost-efficient. The assay is CE-IVDD marked but is available for research use only in the U.S.

Seegene

Q: What are the benefits of using multiplex testing?

A: ***Clinicians and laboratories***

Multiplex PCR testing enables clinicians to make informed decisions for patient management, including the need for isolation and appropriate treatment. The ability to identify infectious pathogens using a single assay rather than multiple tests will allow laboratories to create an efficient workflow and conserve important testing materials that are in short supply.

Multiplex testing can also identify coinfections. Research in 2020 found that people diagnosed with flu and COVID-19 at the same time had an increased risk of death compared to those who only tested positive for COVID-19.[2] Based on a meta-analysis result, coinfection of COVID-19 and flu was associated with a higher risk of ICU admission compared with infections caused by COVID-19 only.[3]

Patients and health authorities

Getting multiple results from a single test means less discomfort and lower costs for patients. Respiratory illnesses may present similar symptoms such as cough, sneezing, stuffy or runny nose, sore throat, and fever. Despite the similarities in symptoms and signs, there are preferred treatment options for each respiratory virus. Influenza B virus, for example, is highly resistant to amantadine and rimantadine, antiviral drugs that are used to treat Influenza A virus.

Multiplex testing also helps early detection of outbreaks by screening multiple viruses in a single test, enabling rapid public health response to limit spread, particularly in hospitals and long-term care facilities.

Q: Can we apply molecular diagnostics to other disease diagnosis areas?

A: Conventional laboratory tests for the detection of infectious pathogens are based on microbiological culture, which requires long incubation times, special facilities, and laboratory personnel highly trained in clinical microbiology. Furthermore, many bacteria and viruses require specific conditions for growth and are, therefore, difficult to culture. In this respect, rapid and highly accurate molecular diagnostic tests will be excellent alternatives for clinical microbiology laboratories.

Other than respiratory illnesses, there is a demand for multiplex molecular diagnostic tests for infectious diseases such as gastrointestinal tract infections, sexually transmitted infections, human papillomavirus infections, meningitis, and urinary tract infections. High multiplex PCR tests covering these disease areascould offer speed and sensitivity that was impossible to achieve with standard microbiology and play a significant role in improving clinical care for patients.

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Trans Medicine, 1919

BY NATALIA MESA

Recorded trans history can be traced back at least 4,500 years, to ancient Sumerian texts documenting priests known as *gala* who may have been transgender. But according to Jules Gill-Peterson, a historian at Johns Hopkins University, the word transgender as we use it today, to refer to people whose gender identities do not align with the sex they were assigned at birth, "is a contemporary, Western concept" popularized in part by German physician Magnus Hirschfeld around the turn of the 20th century.

In early-1900s Germany, homosexuality was illegal. After witnessing several of his gay patients commit suicide, Hirschfeld, a gay man, left his practice to advocate for gender and sexual minorities. In 1919, he founded the Institute for Sexual Research in Berlin to establish sexual science as its own discipline and to provide sexual health services to the community.

At the institute, Hirschfeld conducted extensive research on gender and sexuality, amassing a library of more than 20,000 books and manuscripts. "The research . . . was basically hanging out in bars and getting to know the queer and trans community," Gill-Peterson says. In recording these stories, Hirschfeld developed prescient hypotheses, including the existence of gender and sexuality spectra.

Hirschfeld employed medical professionals to administer hormone replacement therapy and perform early iterations of gender-affirming surgeries, and many of his patients lived and worked at the institute. He was able to provide some with legal "transvestite passes," which protected them against police violence and incarceration. Gill-Peterson says this is an early example of medicine decriminalizing trans identity "by saying it's more appropriate for them to see a doctor or a psychiatrist instead of ending up in prison." But, she adds, it "was a tricky bargain" that also created new power imbalances.

MAGNUS HIRSCHFELD SOCIETY

INSIDE AND OUT: In the 1920s, the Institute for Sexual Research in Berlin was a haven for queer people, many of whom came to the institute seeking to express their identities without fear of being imprisoned. This undated photo depicts a costume party at the institute; its founder, Magnus Hirschfeld (second from right, in glasses), can be seen holding hands with his partner, Karl Giese (center).

Even as the facility provided a valuable public service, many of Hirschfeld's personal views and those of his colleagues "reflected their middle-class values and presumptions," she says, including the idea that science was the best way to understand gender. And although he was Jewish and thought of himself as antiracist, Gill-Peterson notes that Hirschfeld still held views that women were less intelligent than men and that Black people were inferior to whites.

Despite these complexities, Hirschfeld had by the 1930s become a world-renowned sexologist. But backlash followed when, after months of threats, Nazis raided his institute in 1933. Hirschfeld was in France at the time and remained in exile until his death two years later. At least one doctor who performed operations at the institute went on to voluntarily serve as a chief medical adviser at the Dachau concentration camp.

Now, as anti-trans legislation sweeps across the US, Gill-Peterson says that while she doesn't think we're repeating the past, perhaps we haven't fully divested ourselves of it. Trans people continue to struggle with police harassment and poor treatment within the carceral system, as well as for access to gender-affirming procedures and other medical care. In addition, there's rampant disinformation "saying that medical research on trans people is brand new," she says, which isn't the case. To refute this idea, Hirschfeld's story "is important to come back to." ■

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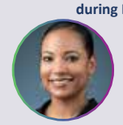


Crystal Rogers
UC Davis

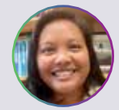
Verena Ruprecht
Centre for Genomic Regulation (CRG), Barcelona

Developmental Plasticity and Regeneration

Alissa R. Armstrong
University of South Carolina

Irene Miguel-Aliaga
Imperial College, London

Environmental Change and Cell Biology



Rosie Alegado
University of Hawai'i Mānoa

Beronda L. Montgomery
Grinnell College

Extracellular Vesicles and Their Roles in Development and Disease

Ayuko Hoshino
Tokyo Institute of Technology

Neta Regev Rudzki
Weizmann Institute of Science, Rehovot

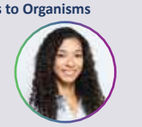
How Structure Enables Function in Differentiated Cell Types

Chantell Evans
Duke University

Raghu Padinjat
National Centre for Biological Sciences, India

Metabolism from Cells to Organisms

Monther Abu-Remaileh
Stanford University



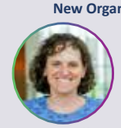
Sonya Neal
UC San Diego

Cellular Changes to Host and Pathogen during Infection

Pascale Guiton
California State University - East Bay

Xiaofeng Wang
Virginia Tech

New Organisms; New Directions



Carrie Adler
Cornell University

Bob Goldstein
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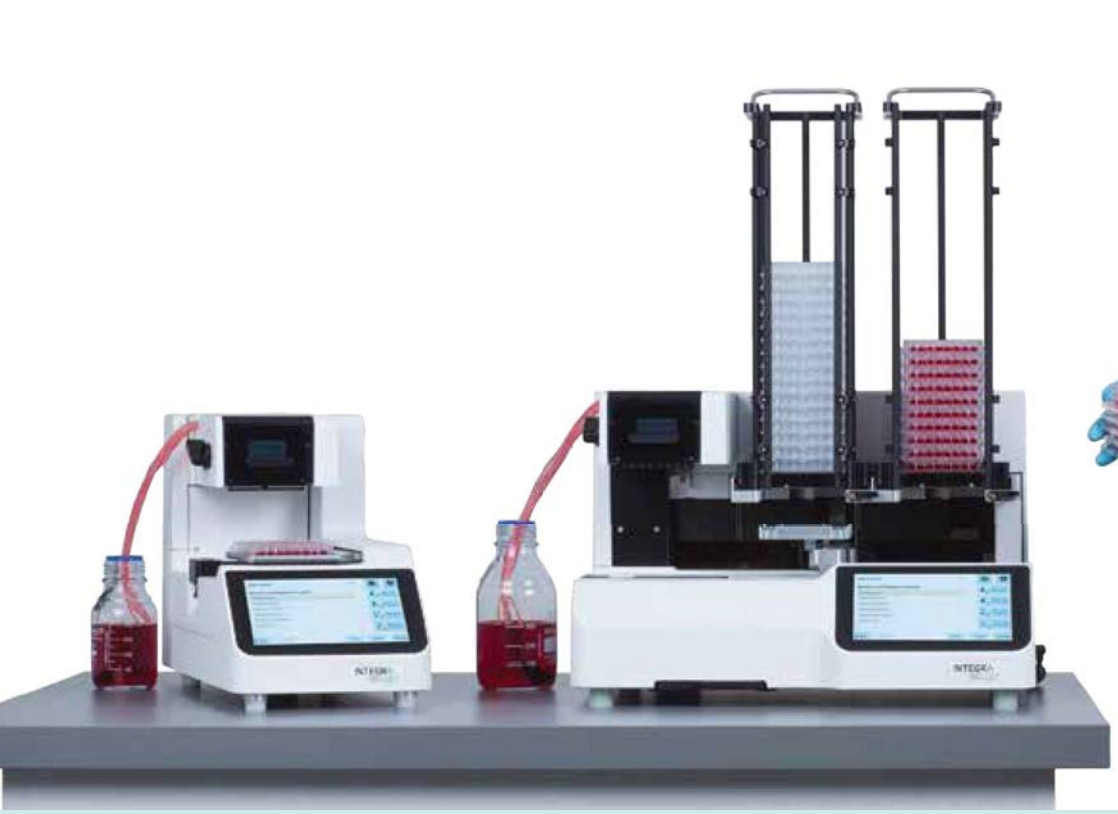

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