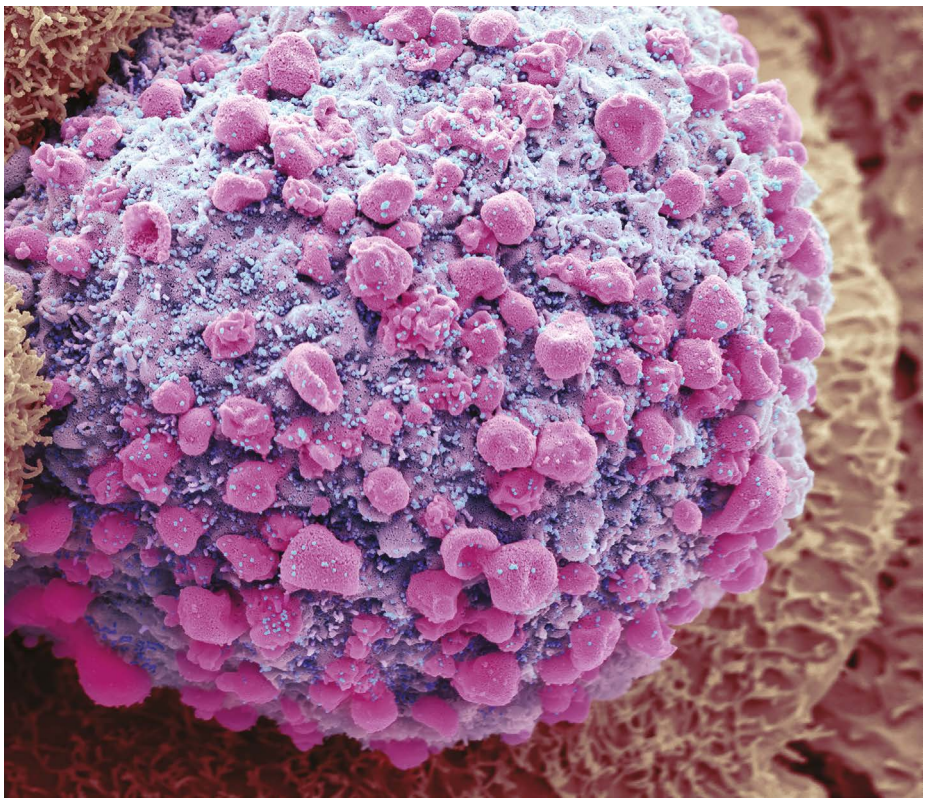


STEVE GSCHMEISSNER/SPL

Particles of SARS-CoV-2 (blue; artificially coloured) bud from a dying intestinal cell.

CORONAVIRUS 'GHOSTS' CAN LINGER FOR MONTHS IN THE GUT

Scientists are studying whether long COVID could be linked to viral fragments that persist in various tissues.

By Heidi Ledford

In the chaos of the first months of the coronavirus pandemic, oncologist and geneticist Ami Bhatt was intrigued by widespread reports of vomiting and diarrhoea in people infected with SARS-CoV-2. "At that time, this was thought to be a respiratory virus," she says. Bhatt and her colleagues, curious about a possible link between the virus and the gastrointestinal symptoms, began to collect stool samples from people with COVID-19.

Thousands of kilometres from Bhatt's lab at Stanford Medicine in California, gastroenterology internist Timon Adolph was also puzzled by accounts of gut symptoms. He and his colleagues at the Medical University of Innsbruck in Austria started to assemble gastrointestinal-tissue biopsies.

Two years into the pandemic, the scientists' foresight has paid off: both teams published results[1,2] last month suggesting that pieces of SARS-CoV-2 can linger in the gut for months after an initial infection. The findings add to a growing pool of evidence supporting the hypothesis that persistent bits of virus — coronavirus "ghosts", Bhatt has called them — could contribute to the mysterious condition called long COVID.

> **"There is anecdotal evidence, but there are a lot of unknowns."**

Even so, Bhatt both urges scientists to keep an open mind and cautions that researchers have not yet nailed down a link between persistent viral fragments and long COVID. "Additional studies still need to be done — and they're not easy," she says.

Long COVID is often defined as symptoms that linger beyond 12 weeks after an acute infection. More than 200 symptoms have been associated with the disorder, which ranges in severity from mild to debilitating. Theories about its origins include harmful immune responses, tiny blood clots and lingering viral reservoirs in the body.

An early hint that the coronavirus might persist in the body came in work[3] published in 2021 by gastroenterologist Saurabh Mehandru at the Icahn School of Medicine at Mount Sinai in New York City and his colleagues. By then, it was clear that SARS-CoV-2 can infect the gut.

Mehandru and his team found viral nucleic acids and proteins in gastrointestinal tissue collected from people who'd been diagnosed with COVID-19 an average of four months earlier. The researchers also studied participants' memory B cells, which are pivotal players in the immune system. The team found that antibodies produced by these B cells were continuing to evolve, suggesting that, at six months after the initial infection, the cells were still responding to molecules made by SARS-CoV-2.

Inspired by this work, Bhatt and her colleagues found that a few people continued to shed viral RNA into their stool seven months after an initial mild or moderate SARS-CoV-2 infection, well after their respiratory symptoms had ended[1].

Virus goes for the gut

Adolph says the 2021 paper inspired his team to look at its samples for signs of coronavirus. The group found that 32 of 46 study participants who had had mild COVID-19 showed evidence of viral molecules in their gut seven months after acute infection. About two-thirds of those 32 people had long-COVID symptoms.

But all of the participants in this study had an autoimmune disorder of the bowel, and Adolph cautions that his data do not establish that there is active virus in these people, or that the viral material is causing long COVID.

In the meantime, more studies have suggested lingering viral reservoirs beyond the gut. Another team of researchers has studied tissue collected during autopsies of 44 people who had been diagnosed with COVID-19 and found evidence of viral RNA in many sites, including the heart, eyes and brain[4]. Viral RNA and proteins were detected up to 230 days after infection. The study has not yet been peer reviewed.

Nearly all of the people in that sample had had severe COVID-19, but a separate study of two people who had had mild COVID-19 followed by long COVID symptoms found viral RNA in the appendix and the breast[5]. Pathologist Joe Yeong at the Institute of Molecular and Cell Biology at the Agency for Science, Technology, and Research in Singapore, who is a co-author of the report, which has not been peer reviewed, speculates that the virus might hide out in immune cells called macrophages.

All of these studies support the possibility that long-term viral reservoirs contribute to

long COVID, but researchers will need to do more work to conclusively show a link, says Mehandru. They will need to document that the coronavirus is evolving in people whose immune systems are not compromised, and they will need to link such evolution to long COVID symptoms. "Right now there is anecdotal evidence, but there are a lot of unknowns," Mehandru says.

Bhatt is hopeful that samples will become available to test the viral-reservoir hypothesis. The US National Institute of Health, for example, is running a large study that aims to tackle the causes of long COVID and will collect intestinal tissue from some participants.

But Sheng says he does not need to wait for a billion-dollar study to get more samples: an organization of people with long COVID has contacted him and offered to send samples from members who have had biopsies for various reasons, such as a cancer diagnosis, after their infections. "It's really random, the tissue can come from everywhere," he says. "But they don't want to wait."

1. Natarajan, A. *et al. Med* https://doi.org/10.1016/j.medj.2022.04.001 (2022).
2. Zollner, A. *et al. Gasteroenterology* https://doi.org/10.1053/j.gastro.2022.04.037 (2022).
3. Gaebler, C. *et al. Nature* **591**, 639–644 (2021).
4. Chertow, D. *et al*. Preprint at Research Square https://doi.org/10.21203/rs.3.rs-1139035/v1 (2021).
5. Goh, D. *et al*. Preprint at Research Square https://doi.org/10.21203/rs.3.rs-1379777/v1 (2022).

DID LIFE BEGIN WITH RNA–PROTEIN HYBRIDS?

Researchers propose an amino-acid twist to the 'RNA world' theory of life's origins.

By Davide Castelvecchi

Chemists say they have solved a crucial problem in a theory of life's beginnings, by demonstrating that RNA molecules can link short chains of amino acids together.

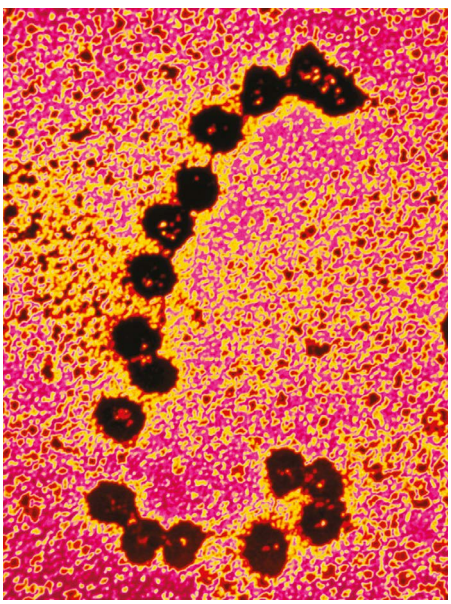
The findings, published on 11 May in *Nature*, support a variation on the 'RNA world' hypothesis (F. Müller *et al. Nature* **605**, 279–284; 2022). This proposes that, before the evolution of DNA and the proteins it encodes, the first organisms were based on strands of RNA, a molecule that can both store genetic information — as sequences of molecules called nucleosides, containing the bases A, C, G and U — and act as catalysts for chemical reactions.

The discovery "opens up vast and fundamentally new avenues of pursuit for early chemical evolution", says Bill Martin, who studies molecular evolution at Heinrich Heine University Düsseldorf in Germany.

In an RNA world, the standard theory says, life could have existed as complex proto-RNA strands that were able to both copy themselves and compete with other strands. Later, these 'RNA enzymes' could have evolved the ability to build proteins and ultimately to transfer their genetic information into more-stable DNA. Exactly how this could happen was an open question, partly because catalysts made of RNA alone are much less efficient than the protein-based enzymes found in all living cells today.

"Although [RNA] catalysts were discovered, their catalytic power is lousy," says Thomas Carell, an organic chemist at Ludwig Maximilian University of Munich in Germany.

While investigating this conundrum, Carell and his collaborators were inspired by the part that RNA plays in how all modern organisms build proteins: a strand of RNA encoding a gene (typically copied from a sequence of DNA bases) passes through a large molecular machine called a ribosome, which builds the corresponding protein one amino acid at a time.



OMIKRON/SPL

The researchers were inspired by ribosomes — shown here translating a strand of RNA.

Unlike most enzymes, the ribosome itself is made of not only proteins, but also segments of RNA — and these have an important role in synthesizing proteins. Moreover, the ribosome contains modified versions of the standard RNA nucleosides that contain A, C, G and U. These exotic nucleosides have long been seen as possible vestiges of a primordial broth.

Experimental ribosome

Carell's team built a synthetic RNA molecule that included two such modified nucleosides by joining two pieces of RNA commonly found in living cells. At the first of the exotic sites, the synthetic molecule could bind to an amino acid, which then moved sideways to bind to the second exotic nucleoside adjacent to it. The team then separated their original RNA strands and brought in a fresh one, carrying its own amino acid. This was in the correct position to form a strong covalent bond with the amino acid previously attached to the second strand. The process continued step by step, growing a short chain of amino acids — a mini-protein called a peptide — that developed attached to the RNA. The formation of bonds between amino acids requires energy, which the researchers provided by priming the amino acids with various reactants in the solution.

"This is a very exciting finding," says Martin, "not only because it maps out a new route to RNA-based peptide formation, but because it also uncovers new evolutionary significance to the naturally occurring modified bases of RNA." The results point to an important part played by RNA in the origins of life, but without requiring RNA alone to self-replicate, Martin adds.

Loren Williams, a biophysical chemist at the Georgia Institute of Technology in Atlanta, agrees. "If the origins of RNA and the origins of protein are linked, and their emergence is not independent, then the math shifts radically in favour of an RNA–protein world and away from an RNA world," he says.

To show that this is a plausible origin of life, scientists must complete several further steps. The peptides that form on the team's RNA are composed of a random sequence of amino acids, rather than one determined by information stored in the RNA. Carell says that larger RNA structures could have sections that fold into shapes that 'recognize' specific amino acids at specific sites, producing a well-determined structure. And some of these complex RNA–peptide hybrids could have catalytic properties and be subject to evolutionary pressure to become more efficient. "If the molecule can replicate, you have something like a mini organism," says Carell.