Nanobacteria - Are They Alive?

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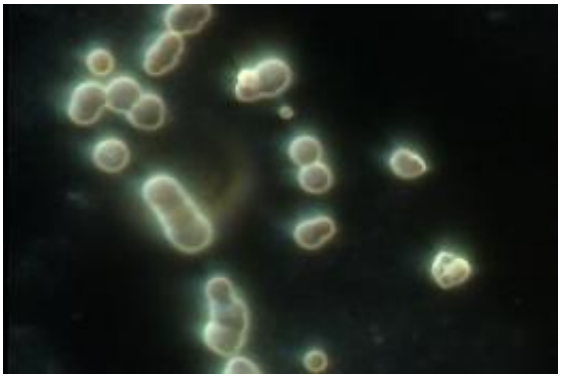

Calcium carbonate crystals (nanobacteria-like particles) have a cellular appearance, but the new study shows that nanobacteria are not alive. Image credit: Martel and Young. ©2008 PNAS.

Tiny particles called nanobacteria have intrigued researchers in many ways since their discovery 20 years ago, but perhaps the most controversial question they pose is whether or not they are alive.

Nanobacteria – which sometimes go by the name “nanobes” or “calcifying nanoparticles” – don’t seem to fit scientists’ criteria for life. Researchers at a workshop hosted by the National Academy of Sciences for this specific reason concluded that the minimal cellular size of life on Earth must exceed 200 nm in diameter in order to contain the cellular machinery based on DNA replication. But nanobacteria can be as small as 80 nm – so, unless they contain some novel replicating mechanism, it seems unlikely that they constitute a form of life.

That's just one piece of evidence against living nanobacteria named in a recent study by Jan Martel of Chang Gung University in Taiwan and John Ding-E Young from The Rockefeller University in New York, which was published in *PNAS*. Martel and Young have studied healthy human blood serum that contains what they call "nanobacteria-like particles" (NLP), composed of the compound calcium carbonate ($CaCO_3$), or limestone. The researchers performed a series of experiments showing that the tiny particles contain no traces of DNA or RNA, and suggest that their formation can be explained by non-biological means.

"We believe that this study provides substantive proof that nanobacteria are not living entities," Young told *PhysOrg.com*. "Some previous studies have hinted that this is the case, but have not provided a chemical composition or formulation that could explain the nanobacteria phenomenon in its entirety."

One thing about nanobacteria that's clear is that they're very widespread, occurring in practically all human material tested. Under an electron microscope, nanobacteria (and the NLPs) look like typical bacteria, and even resemble cells undergoing division. They're also hardy: when the researchers bombarded the NLPs with 30 kGy (kiloGray) of gamma radiation, it didn't prevent them from growing in cultures, in accordance with previous studies.

Another bacteria-like property of NLPs is that they have the ability to nucleate hydroxyapatite (HAP), a calcium phosphate crystal that largely composes the bones and teeth of humans and animals. Previous research has suggested that this might be how the nanobacteria self-replicate. When Martel and Young investigated this issue in their study, however, they found that HAP only forms around NLPs under certain conditions. For example, when mixed with some crystal-growth-inhibiting proteins, NLPs stop nucleating HAP, indicating that HAP is not really necessary

for NLP formation.

Instead, their experiments lead Martel and Young to suggest a chemical rather than biological model for NLP formation. Based on this hypothesis, they could control the speed and shape of NLP formation in vitro by simply varying the substrates needed for the precipitation of calcium carbonate.

These findings could also shed light on nanobacteria that have shown up in a variety of other areas, from sandstones of the Triassic and Jurassic eras to meteorite fragments from Mars. The chemical process that the researchers describe here for nanobacteria formation could be the same for these nanobacteria, as well.

“Nanobacteria have been heralded as the smallest cellular forms on Earth and as candidates to explain how cellular life began on Earth and other extraterrestial bodies, like meteorites and Mars,” Young said. “Our results clearly disprove that nanobacteria are living organisms. We have shown that all the previous vast body of literature in nanobacteria can actually be explained by a chemical and abiotic mechanism involving the simple deposition of limestone or calcium carbonate.”

Nano-pathogens?

Previous research has suggested that nanobacteria could be the cause of a wide variety of diseases, from kidney stones to atherosclerosis – a prospect which now must be tested with the new nanoparticles. Because they multiply faster in low-gravity environments, NASA is particularly concerned in light of astronauts’ increased risk for developing kidney stones. According to Martel and Young, these nanoparticles may be part of a much wider family of organic mineral complexes that seem to assemble and propagate as if they are alive – in fact, much like prions, the self-assembled proteins that cause mad cow disease.

“We believe that we have uncovered a whole family of organic mineral complexes that give the seeming appearance of replication and self-assembly as if they are live entities,” Young said. “They appear to be ubiquitous entities found in living and non-living substrates.”

Some researchers have even been developing antibodies to try to combat the “pathogenic” nanobacteria. A company called Nanobac Oy, owned by Nanobac Life Sciences and founded by the discoverers of nanobacteria, has antibodies that are commercially available and sells diagnostic kits for detecting the nanobacteria. The antibodies come from mice cells that have been immunized with nanobacteria obtained from cows.

To try to understand the nature of the reaction between the antibodies and nanobacteria, Martel and Young tested the antibodies on NLPs, which gave positive reaction, as expected. Surprisingly, however, the same antibodies also reacted with albumin, the most common protein in the blood serum. Since proteins like albumin can not possibly have been produced by any living bacteria, they’re probably attached to the calcium carbonate particles, and reacting with the antibodies, the researchers explain.

“Since nanobacteria have now been disproved as living entities, it is unlikely that they can produce diseases as bacteria would,” Young added. “Their common distribution in living and non-living environments – from blood to soil to meteorites – must be taken into account when speculating a role for them in disease. This is not to say that such nanoparticles are incapable of causing disease – with which they may very well be involved – but any such claims must be rigorously established through verifiable documentation, which is lacking at the present moment.”

More information: Martel, Jan, and Ding-E Young, John. “Purported

nanobacteria in human blood as calcium carbonate nanoparticles." *Proceedings of the National Academy of Sciences*. April 8, 2008. vol. 105, no. 14, 5549-5554.

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