Revealing the earth's secrets

August 22 2006

In biology textbooks the world over you can read about how important bacteria in the soil are for plants and animals. Now the textbooks might have to be rewritten.

The textbooks tell us namely that there are two types of nitrifying bacteria which account for all the oxidisation of ammonia (NH_3) into nitrite (NO_2) and subsequently into nitrate (NO_3). It turns out that these bacteria are probably much less important in the nitrogen cycle than previously assumed. Instead, it appears that most of this job may be done by a completely different type of micro-organism: archaea.

Professor Christa Schleper of the Department of Biology at the University of Bergen, Norway, is a specialist in archaea, and she is behind this discovery, which may prove expensive for the textbook publishers. A lot of research remains to be done, however - archaea is namely a type of organism about which little has so far been known.

“Many of them are difficult to cultivate in the laboratory, since the soil is a complicated system that is impossible to copy in the laboratory. Only archaea that live in hot springs or other extreme environments thrive under such artificial conditions,” she explains. That is why scientists also believed for a long time that archaea only lived in extreme environments. And these simple organisms were assumed to play a much less important role than bacteria in the earth and the ocean, for example. Quite simply because they had not been observed there to any great extent.

Archaea enough to go around

But that was before gene technology made it possible to sequence DNA from just about anything. Because if you take a sample of soil from your garden and extract all the DNA you find in it, you will find that there are more than just bacteria in it. There are archaea also. Huge amounts of them.

“In one gram of soil there are an average of at least 10,000 species of micro-organisms,” explains Professor Schleper. And the more scientists study soil samples, the more archaea they find.

Professor Schleper has specialised in cloning DNA from soil samples and extracting long strings of the genetic material. She is thus able to find out how many different organisms are the source of all the genetic material in the samples - and by studying the genes in the long DNA fragments, she can also work out which attributes they have.

Finding the identity of a micro-organism is done by looking for a so-called phylogenetic marker gene, the rRNA gene or the gene for ribosomal RNA. This gene is namely found in all living organisms. By analysing which other genes are present in various long DNA fragments that include the rRNA gene, Professor Schleper can then find out whether they stem from the same species or from different species.

“That is basically how scientists discovered that archaea are also present in soil samples and not just in hot springs or in deep oceans,” she explains.

Needed rock solid evidence

Professor Schleper and her collaborators caused a stir when they were able earlier this year to reveal in Nature that some of the archaea genetic material that was present in soil samples also contained genes for the oxidisation of ammonia. Just like nitrifying bacteria did. She immediately suspected that archaea could thereby be important pieces in the nitrogen cycle. And her research team started to gather evidence for this theory:

•They first looked in soil samples for the enzyme that was the product of the oxidisation process – and found large quantities of it.
•In addition, they also looked for certain lipids that are well-known products of archaea, and not of bacteria. They also found these in large quantities.

The soil samples were taken from several locations, from the Greek island of Santorini, via Germany to Bergen. It seemed to be the case that bacteria were ousted by archaea. And the deeper they dug, the fewer bacteria they found - while the amount of archaea remained constant.

She presented the results to the journal Nature, which liked what it read. But it refused to publish it - at least right away.

“They said: ‘If you are going to knock the ground from under a century-old dogma, the documentation has to be rock solid’,” explains Professor Schleper. The team therefore reconvened with the samples and performed a new type of analysis.

A revolution under way

This time she looked for RNA copies of the exact gene that is expressed when the archaea organisms oxidise ammonia. When she also found it in large quantities, the editors of Nature - and the scientists who assessed the article - were finally satisfied, and the article was accepted for publication.

“It is still too early, however, to talk about rewriting the textbooks,” Professor Schleper underlines. Because even though she has shown that archaea are present in large quantities in the soil, and even though they indisputably oxidise ammonia, it is still conceivable that bacteria make a more effective contribution to the process. The next step for Professor Schleper is therefore to find out what is most important to the nitrogen cycle - archaea or bacteria.

“Either by trying to halt the process in the one group and seeing what effect that has on overall oxidisation, or trying to characterise the biochemistry that underlies each of them,” explains Professor Schleper. If archaea then prove to be the most important contributor, this could have major ripple effects.

“Perhaps first and foremost for agriculture, where the nitrogen cycle is absolutely vital. Nitrification is also important in the purification of sewage,” she explains. Understanding nitrification in the soil is also important in understanding the process of eutrification, or overfertilisation, of lakes and coastal waters.

The revolution that is under way in research into the micro-organisms in the soil is a result of new genetic methods. But Professor Schleper points out that scientists at the University of Bergen foresaw the enormous diversity long before the new techniques were available:

“Already during the 1980s, Professor Vigdis Torsvik of the Department of Biology here calculated that the diversity of species in the soil would be as large. At the time, not everyone fully believed her, but modern technology has shown that she was completely right,” says Professor Schleper.

Reference: *Nature* 442, 806-809 (17 August 2006);
doi:10.1038/nature04983

Source: University of Bergen