to the insulator–metal transition. This would be a major advantage over the FeSi system, because the effects of the type and density of carriers could be studied in detail. In FeSi only aluminium, which substitutes for Si and introduces additional holes, can have this role.

Broader applications of the principles described by Manyala *et al.*[1] to wide classes of well-characterized semiconductors offer the opportunity of new approaches to the understanding of the breakdown of conventional Landau–Fermi liquid theory and with it a possible route towards the control of strong electronic correlations in solids for useful purposes. ■

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BIOGEOCHEMISTRY

Who lives in the sea floor?

Ann Pearson

The sediments that blanket the sea floor contain tremendous numbers of microorganisms. This deep marine biosphere, which is probed by deep-sea drilling, is a new frontier for microbiologists and geochemists.

On page 991 of this issue, Lipp *et al.*[1] add to the debate over the nature of life in the sea floor. They report that most cells in deep-sea sediment are members of the domain Archaea and not of the other domain of the prokaryotes, the Bacteria. Although both are differentiated from the eukaryotes — which include ourselves — in lacking a nucleus, Archaea and Bacteria are fundamentally different from each other in their biochemistry, metabolism and evolutionary history. This in turn profoundly influences their role in Earth's biogeochemical cycles.

A century ago, the suggestion that the ocean's deep sediments had inhabitants — organisms far removed from the oxygenated, light-filled surface of Earth — would have been preposterous. Life *on* the sea floor was well known; such fauna were sampled during the voyage of the HMS *Challenger* (1872–76), in an expedition that is widely recognized as marking the birth of modern oceanography (Fig. 1). The first hint of a rich microbial biosphere *in* the sea floor came only much later[2], and it was not until the 1990s that microbial cells in deep sediments were enumerated systematically[3,4]. In parallel, geochemists showed that inorganic solutes in sediment pore-waters usually react in the order predicted by thermodynamic principles, but at rates exceeding expectations for abiotic processes[5–7]. Thus biologists and chemists agree: there must be a 'deep biosphere'. But what populates it? And how do the organisms concerned adapt their metabolic strategies to sustain life at the limits of energetic viability[8,9]?

The deep biosphere is large, and, at least in aggregate, it is 'alive'[10,11]. The data almost defy imagination — on the order of a million cells in every cubic centimetre of sediment buried half a kilometre below the sea floor, or more than 50% of all microbial cells on Earth[12]. The overall order of magnitude is the same as the biomass of all surface plant life. In investigating what kind of cells they are, Lipp *et al.*[1] conclude that, deeper than 1 metre below the sea floor, it is Archaea, not Bacteria, that contribute the bulk of sedimentary microbial biomass. If true, Archaea would be the most abundant cell type in the marine system, rivalling Bacteria in total numbers globally.

Lipp *et al.*, however, are far from the first to approach the question of the phylogeny of sedimentary prokaryotes. Previous efforts generated conflicting results[10,11,13], and a hung jury has been declared[14] over the question of the predominance of Bacteria or Archaea. But why the protracted debate? Part of the problem lies in the distinction between 'living' cells, total cells (including inert or dead cells), and/or cells that are in between, persisting in an undefined degree of stasis. This leads to ambiguity about what should or should not be counted. Adding to the confusion, nearly every report so far also has used a different combination of analytical techniques, and consideration of those techniques is essential to understanding the issues.

The fluorescent stains acridine orange and DAPI detect all cells — alive or dead — that contain any remnant of DNA. But they cannot reveal phylogeny or activity. So, to discriminate between Bacteria and Archaea, molecular methods have been used in various combinations. These include approaches that detect ribosomal RNA (rRNA) or intact polar lipids (IPLs) of cell membranes (both are proxies for live cells), or that quantify rRNA gene copies from extracted DNA. Of these techniques, *in situ* fluorescent tagging of ribosomes in methods known as FISH or CARD-FISH produce the most disparate (indeed, opposite) results[10,11,13]. Although potentially an ideal

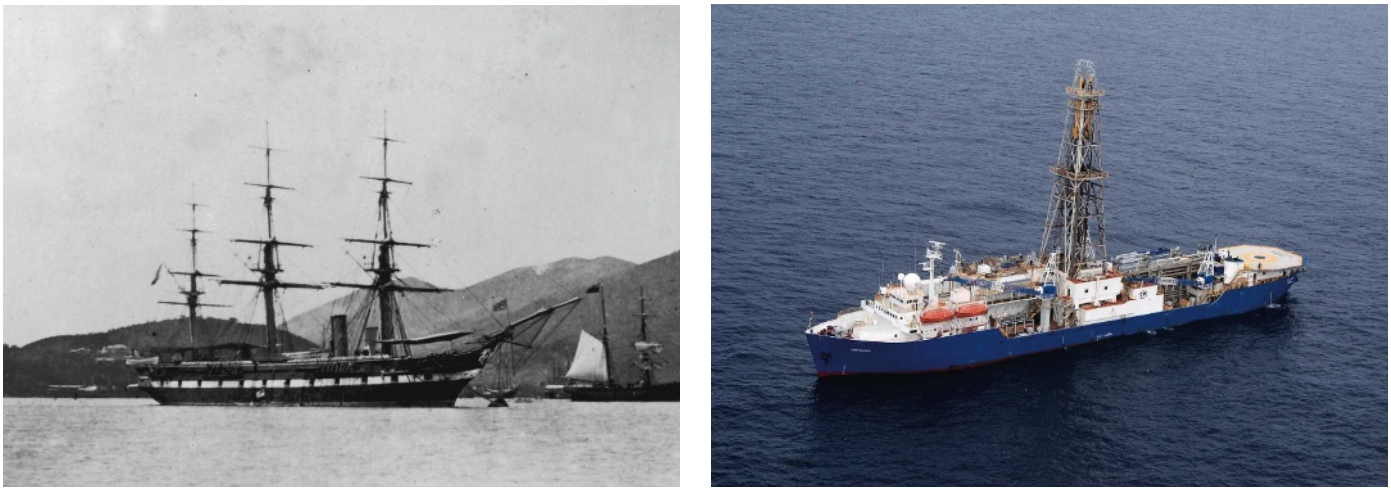

Figure 1 | Sea change. HMS *Challenger* (left), whose voyage in the 1870s opened eyes to the fauna living on the floor of the deep sea, and the drill ship *JOIDES Resolution*, mainstay of the Ocean Drilling Program and subsequent Integrated Ocean Drilling Program, which has provided the cores from which much of the information about life in deep-sea sediments has been gleaned.

ARCHAEOLOGY

An oasis in time

Today's Sahara is arid and inhospitable, but this was not always the case. In the early Holocene (between about 10,000 and 4,500 years ago), monsoon rains created a lush savannah rich in animal and plant life. Complex human societies settled there beside ancient lakes, as demonstrated by a recently reported archaeological site documenting nearly 5,000 years of human occupation (P. C. Sereno *et al. PLoS ONE* **3,** e2995; 2008).

The site, named Gobero and situated in central Niger, contains about 200 burial sites, which, along with several rubbish dumps, provide a record of two distinct periods of human settlement. It was originally occupied 9,500 years ago by a tall, well-muscled people who fished the lake for Nile perch and large catfish with the use of bone harpoons and hooks. These people abandoned the site a little over 8,000 years ago when an extended arid period dried up the lake.

Gobero was recolonized 1,000 years later by a slighter and shorter people who ate clams and small catfish from the now much shallower lake, as well as antelope and other vertebrates from the surrounding savannah. This population had sophisticated burial practices involving jewellery and grave goods, and what appear to be ritual poses. One grave, dated to be about 5,300 years old, contained a woman and two children buried together with clasped hands (pictured). Pollen found in this grave suggests that they were buried on a bed of wool flowers (*Celosia*).

Occupation of the Gobero site came to an end around 4,500 years ago, when changing climate returned this region to the arid desert conditions that persist to this day.

Christopher Surridge

MIKE HETTWER/PROJECT EXPLORATION

method of identifying metabolically active cells, the data suggest that consistent application of FISH remains a challenge. To achieve a similar goal of studying only 'living' cells, direct extraction of rRNA followed by reverse transcription to DNA has been successful[13], but attempts to make this approach quantitative face major hurdles.

Theoretically, there are fewer challenges when examining DNA or lipids. Quantitative amplification of extracted DNA by using the polymerase chain reaction (qPCR) allows rRNA genes to be counted, rather than seeking ribosomes directly. Even after accounting for the variable copies per cell of these genes in Bacteria and Archaea, early results from qPCR invariably declared the winner to be Bacteria[11,15]. How, then, is it possible that Archaea could have been underestimated?

The key word is 'extracted' — Lipp *et al.*[1] resolve the qPCR dilemma, showing that a more aggressive approach to obtaining total DNA is essential. It is revealing to view their Supplementary Fig. 3: depending on the method used, as many as 80% or as 'few' as 15% of cells escape lysis, the latter under optimized conditions. The implication is that DNA from these escapees would be overlooked during qPCR, and most of them would be Archaea with their more durable cell envelopes.

Improved extraction brings estimates from qPCR in line with earlier claims of archaeal abundance as derived from IPLs[13]. Polar lipids are presumed to reflect living biomass, because their labile (often phosphate-containing) head groups are quickly lost after cell death. Lipp *et al.* also offer expanded IPL data covering seven different locations. Nearly 90% of IPLs below a depth of 1 metre are specific to Archaea, and the total abundance is proportional to the total organic carbon content of the sediment in which they are found. This suggests that archaeal production fundamentally scales to the available organic resources, whatever the type of metabolism involved.

In considering Archaea and Bacteria, it has been proposed that Archaea are united by a universal ecological ability to cope with energetic stress[16]. It is therefore reasonable that in a sub-seafloor world, where it has been estimated that cell turnover times could be centuries or longer[8,9], organisms with honed strategies to conserve energy would dominate.

Nevertheless, Lipp and colleagues' results will be controversial. Much of their argument rests on the interpretation that all IPLs represent living cells — that is, that the degradation time of IPLs after cell death is infinitely fast relative to other processes in the system. Although IPLs degrade rapidly in experiments, little is known about their persistence in complex communities with extraordinarily low rates of enzymatic activity. Specifically, it is the persistence of archaeal IPLs relative to bacterial IPLs that is of particular importance. This requires further study of the turnover of both Bacteria and Archaea in sediments, especially with regard to specific rates of synthesis, alteration and degradation of lipids. For microbes, the boundary between alive and dead is fuzzy, and the extent to which any category of biomolecule can define it remains unclear. Progress on all fronts of culture-independent and culture-dependent techniques will be necessary to tackle these uncertainties. ■

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