

## Extremophiles: There's More to Life

Philip Hendry<sup>A</sup>

<sup>A</sup> CSIRO Molecular and Health Technologies, PO Box 184, North Ryde, NSW 1670, Australia.  
Email: phil.hendry@csiro.au

Extremophiles, organisms that thrive under extreme conditions, are widespread and have played a critical role in shaping the Earth as we know it today. Fifty years ago, the idea that an organism would prefer to grow at pH 1, 110°C or under 800 atmospheres of pressure would have seemed ridiculous. However, as our ability to detect microbes has improved, the range of environments supporting life has expanded to the point now where it is no longer surprising to discover organisms 3 km underground, in acid streams at pH 0.5 or surrounding 'black smokers' 2 km under the sea. This Research Front on extremophiles takes us into the laboratories of some groups working in this intriguing and fast-developing field. This introductory essay is intended to give a flavour of the field of extremophile research and briefly introduce the diversity of extremophiles, the role they play in the environment and their utility in biotechnology.

The term 'extremophile' was introduced by MacElroy<sup>[1]</sup> in 1974, although the more specific terms acidophile, halophile and thermophile have been used in the literature since about the turn of the 20th century. Extremophiles can be grouped according to the conditions in which they thrive. Thus, there are thermophiles and psychrophiles (which grow at the extremes of temperature ranges, barophiles or piezophiles—high pressures), halophiles (high salt), acidophiles and alkaliphiles (extremes of pH). Some workers broaden the definition of extremophiles to include organisms tolerant of high concentrations of heavy metals or ionising radiation. This is not a universal practice because, by and large, those organisms grow at least equally well in the absence of those conditions. Other terms encountered in the field include xerophile, or, more correctly, xerotolerant, to describe organisms that tolerate very low water activity whether they are endoliths (inside rocks), halophiles or simply inhabit extremely dry environments. Some adventurous organisms are extreme in more than one dimension.

**Table 1. Current limits for extremophiles**

| Group        | Organism                           | Limit for growth | Reference |
|--------------|------------------------------------|------------------|-----------|
| Thermophile  | Archaeal 'Strain 121'              | 121°C            | [2]       |
| Psychrophile | Himalayan Midge                    | −18°C            | [3]       |
| Acidophile   | <i>Ferroplasma acidarmanus</i>     | pH 0             | [4]       |
| Alkaliphile  | <i>Alkaliphilus transvaalensis</i> | pH 12.5          | [5]       |
| Barophile    | <i>Moritella</i> spp.              | 80 MPa           | [6]       |

For example, members of the *Sulfolobus* genus are known with optimal growth parameters around 85°C and pH 2.0.<sup>[7]</sup> The current described limits for the extremophiles are given in Table 1.

The domain of life most strongly associated with extremophilia is the Archaea, which although morphologically similar to the Bacteria, were shown in the 1970s<sup>[8]</sup> by DNA sequence analysis to be distinct from both the Bacteria and Eukarya. However, there are representatives of all kingdoms in the extremophiles. The Himalayan Midge<sup>[3]</sup> is active at −18°C and many Bacteria and algae are happy growing at less than pH 1.0, while Bacteria are described that grow at pH 12.5.<sup>[5]</sup>

Many have speculated that the over-representation of the Archaea in the extremophiles (particularly the hyperthermophiles) reflects their phylogenetic proximity to LUCA (Last Universal Common Ancestor), and the conditions on Earth during the origins of life more than 4 billion years ago. The continued discovery of life in extreme environments has provided significant encouragement to the field of astrobiology, the search for life beyond Earth. The conditions required to sustain life even as we currently understand it are quite broad; temperatures  $\sim 50 \pm 70^\circ\text{C}$ , a relatively small range of elements, (liquid) water, and an energy source, conditions that almost certainly occur frequently throughout the universe.



Phil Hendry graduated with a Ph.D. in biomimetic inorganic chemistry from the Australian National University in 1986. This was followed by various research positions in chemistry and molecular biology before taking a post with CSIRO in Sydney where he worked on the kinetics, mechanism and evolution of catalytic RNAs. Phil is currently project leader of a group focussed on industrial microbiology. In this context, his group studied sulfur oxidising bacteria and hyperthermophilic archaea for sulfide leaching applications, isolated alkane and fatty-acid-oxidising soil bacteria, identified organisms from petroleum reservoirs and coal seams.

The roles that microbial extremophiles play in the environment have been underestimated for some time. This is partly because of the sluggishness of many of the processes and partly our ignorance of their existence. It is thought that microbes were responsible for the precipitation of many valuable concentrated ore deposits, for example, by sulfate or metal ion reduction. The devastation caused to waterways by acid mine drainage demonstrates the importance of oxidising acidophiles to the sulfur and many metal geochemical cycles. Thermophiles deep in the biosphere cause the degradation of petroleum deposits, while in coal deposits, consortia of Bacteria and Archaea cooperate to produce methane.

Biotechnologists have exploited extremophiles as a source of enzymes or whole organisms to catalyse processes under extreme conditions. The most celebrated example being the use of thermostable DNA polymerase from *Thermus aquaticus* to practically enable the polymerase chain reaction (PCR), which involves repeated cycling of the reactants between ~50 and 95°C.<sup>[9]</sup> Appropriately, PCR has now become one of the critical tools in the study of extremophiles. Although thermophiles have been the source of many 'extremozymes', many useful enzymes have also come from psychrophiles (proteases and lipases active at low temperature as detergent additives), acidophiles (amylases for acidic starch degradation), alkaliphiles (proteases for detergent applications), and halophiles (enzymes active in non-aqueous solvents). Acidophiles and alkaliphiles typically maintain their cytoplasm at relatively neutral pH and only the secreted enzymes tend to be extremophilic. In contrast, halophiles adapt to the osmotic stress by two basic mechanisms, by maintaining a high salt concentration in the cytoplasm or lowering the osmotic pressure by accumulating high levels of low molecular weight neutral organic species. In the former method, cytoplasmic proteins adapt to that environment (which tends to precipitate proteins) by accumulating more anionic amino acids at the surface, which also aids their stability and activity in non-aqueous solvents.

This Research Front gives an insight into a few topics in this fast-developing field. It begins with a review of the extremophiles and an examination of the chemistry of adaptation to extreme conditions.<sup>[10]</sup> It continues with a paper on difficulty of reliably extracting DNA from the acidic soils of Mount Hood.<sup>[11]</sup> This is important, because it is variously stated that between 90 and 99% of the microbial content of any soil sample is not able to be cultured. Therefore, any survey of bacterial populations in environmental samples relies heavily on culture-independent methods, many of which require reliable DNA recovery. There are two papers on various aspects of halophilic organisms. One provides an overview of the 'compatible-solute' method of coping with osmotic stress and goes on to discuss possible mechanisms for bacteria to sense osmotic pressure and regulate the internal concentration of those solutes.<sup>[12]</sup> Interestingly,

compatible solutes like ectoine are now becoming sought after as additives in cosmetics because of their 'ability to protect biological structures from stress'. The final paper studies the response of various fungi from hypersaline waters and polar glacial ice to changes in salinity.<sup>[13]</sup> In particular, the level of mycosporines was found to increase when some of the fungi were grown in the presence of increasing salt levels. Mycosporines are low molecular weight compounds with strong UV-absorbing properties found in a range of aquatic organisms and widely accepted to have a role in protection from UV radiation. The authors pose the further question: are mycosporines also acting as compatible solutes?

The contents of this Research Front give us a brief glimpse into the diversity of this rapidly expanding field. Over the next decades we will continue to uncover new extremophiles, with novel ways of dealing with extreme conditions and new extremozymes will be put to work. The major revolution, however, will come from the explosion of genomic information. It will soon be routine to sequence a microbial genome in a few hours for a few thousand dollars. That will be dramatic enough, but the price of sequencing will mean that it becomes feasible for researchers to sequence entire uncultured communities.<sup>[14]</sup> This will herald a new era in environmental microbiology when we can impute functions to and interactions between organisms that we have never seen or cultured.

## References

- [1] R. D. MacElroy, *Biosystems* **1974**, *6*, 74. doi:10.1016/0303-2647(74)90026-4
- [2] K. Kashefi, D. R. Lovely, *Science* **2003**, *301*, 934. doi:10.1126/SCIENCE.1086823
- [3] S. Kohshima, *Nature* **1984**, *310*, 225. doi:10.1038/310225A0
- [4] K. J. Edwards, P. L. Bond, T. M. Gihring, J. F. Banfield, *Science* **2000**, *287*, 1796. doi:10.1126/SCIENCE.287.5459.1796
- [5] K. Takai, D. P. Moser, C. Onstott, N. Spoelstra, S. M. Pfiffner, A. Dohnalkova, J. K. Fredrickson, *Int. J. Sys. Evol. Microbiol.* **2001**, *51*, 1245.
- [6] C. Kato, L. Li, Y. Nogi, Y. Nakamura, J. Tamaoka, K. Horikoshi, *Appl. Environ. Microbiol.* **1998**, *64*, 1510.
- [7] J. J. Plumb, B. Gibbs, M. B. Stott, W. J. Robertson, J. A. E. Gibson, P. D. Nichols, H. R. Watling, P. D. Franzmann, *Miner. Eng.* **2002**, *15*, 787. doi:10.1016/S0892-6875(02)00117-6
- [8] C. R. Woese, G. E. Fox, *Proc. Natl. Acad. Sci. USA* **1977**, *74*, 5088.
- [9] K. Mullis, F. Faloona, S. Scharf, R. Saiki, G. Horn, H. Erlich, *Biotechnology (N. Y.)* **1992**, *24*, 17.
- [10] A. A. H. Pakchung, P. J. L. Simpson, R. Codd, *Environ. Chem.* **2006**, *3*, 77.
- [11] R. M. Henneberger, M. R. Walter, R. P. Anitori, *Environ. Chem.* **2006**, *3*, 100.
- [12] H. J. Kunte, *Environ. Chem.* **2006**, *3*, 94.
- [13] T. Kogej, C. Gostincar, M. Volkmann, A. A. Gorbushina, N. Gunde-Cimerman, *Environ. Chem.* **2006**, *3*, 105.
- [14] J. C. Venter, K. Remington, J. F. Heidelberg, A. L. Halpern, D. Rusch, J. A. Eisen, D. Wu, I. Paulsen, et al., *Science* **2004**, *304*, 66. doi:10.1126/SCIENCE.1093857