EXTREMOPHILES: BASIC CONCEPTS

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Summary

Extremophiles are organisms which permanently experience environmental conditions which may be considered as extreme in comparison to the physico-chemical characteristics of the normal environment of human cells: the latter belonging to the mesophile or temperate world. Some eukaryotic organisms such as fishes, invertebrates, yeasts, fungi, and plants have partially colonized extreme habitats characterized by low temperature and/or of elevated hydrostatic pressure. In general, however, the organisms capable of thriving at the limits of temperature, pH, salt concentration and hydrostatic pressure, are prokaryotic. In fact, some organisms depend on these extreme conditions for survival and have therefore developed unique adaptations, especially at the level of their membranes and macromolecules, and affecting proteins and nucleic acids in particular. The molecular bases of the various adaptations are beginning to be understood and are briefly described. The study of the extremophile world has contributed greatly to defining, in more precise terms, fundamental concepts such as macromolecule stability and protein folding. In addition extremophiles offer unique and

unlimited potential in development of biotechnological tools, which are only now beginning to be exploited.

1. Introduction

The word “extremophile” refers to organisms that survive, and indeed proliferate, under physico-chemical conditions to some extent removed from those characterizing a suitable environment for human beings. This anthropocentric view is quite useful in defining what is an “extreme property” since the possible deleterious effect of an unsuitable parameter, physical or chemical, on human cell integrity is easily appreciated. Environments considered as harmful include those characterized by extreme acidity or alkalinity, very low or elevated temperatures, or by pressures remote from so-called atmospheric pressure. To these, one can add any environment in which our eukaryotic cells will suffer from the presence of various xenobiotic substances, such as heavy metals and high salt concentrations, or from the absence of oxygen. The cells, mainly prokaryotic, capable of supporting one or several of these extreme conditions, have been grouped into families on the basis of the main characteristic of their environment. One can nowadays therefore distinguish seven families of extremophiles:

1. *Thermophiles*, adapted to temperatures generally exceeding 60 °C.
2. *Psychrophiles*, which can readily grow at temperatures often below 0 °C.
3. *Halophiles*, which can be completely dependent on salt concentrations exceeding the salinity of seawater by more than a factor of ten.
4. *Alkaliphiles*, which can easily thrive at pH values exceeding 10.
5. *Acidophiles*, which can easily thrive at pH values close to 0.
6. *Metallophiles*, which can survive in high concentrations of heavy metals.
7. *Barophiles*, now more commonly named *piezophiles*, which can be successful in environments exposed to hydrostatic pressures as high as about 1,000 atmospheres—the conditions prevailing in the deepest ocean regions such as the Mariana Trench (–10,897m). By analogy, organisms having colonized ecosystems deprived of oxygen are known as *anoxiphiles*.

In 1974, MacElroy proposed the name “extremophile” to designate any organism able to support environmental conditions usually fatal to most eukaryotic cells. This extremophily has pertinently been compared to the eccentricity characterizing human beings displaying behavior considered marginal by normal citizens. In contrast, the term *mesophile* is applied to organisms adapted to non-stressing environmental conditions such as those experienced by normal eukaryotic cells.

Eukaryotic cells and multicellular organisms are usually very sensitive to unusual physical or chemical environmental conditions. For instance, the temperature limit for their survival does not exceed 60 °C, as in the case of some algae and fungi, whereas most other eukaryotic organisms do not tolerate temperatures higher than 50 °C. This is due to the particular sensitivity of some cellular components, the integrity of which is altered above a certain temperature threshold. This remark is, of course, also valid for the other extreme parameters mentioned above. The discovery of organisms resistant to temperatures higher than 60 °C only dates back to the late 1960s, when microorganisms capable of growing at temperatures greater than 70 °C were identified in hot springs at the Yellowstone National Park in the USA. Further investigation led to the identification of extraordinary microorganisms capable of growing at a temperature of 113 °C. These included *Pyrolobus fumarii*, isolated from a deep-sea hydrothermal vent and amazingly unable to thrive at temperature lower than 90 °C. In similar extreme contexts, microorganisms such as *Picrophilus oshimae*, discovered in acidic geothermally heated waters, can grow successfully in pH environments close to zero, equivalent to a 1M solution of Hydrochloric acid. Even more impressive are the organisms sustaining pressures equivalent to 1,000 atmospheres miraculously exhumed from the 10 km deep Mariana Trench, an elongated depression on the Pacific Ocean floor, in the early 1970s. These few examples testify to the remarkable adaptation processes which have taken place in order to render life possible in such extreme conditions. An irresistible current of curiosity has drawn numerous scientists into this field of investigations, not only to try to understand the molecular processes involved, but also to exploit the unique properties of these organisms and, in particular, of their enzymes.

Taking into account the origin of earth, resulting from a condensation process of cosmic dust about 4.5 billion years ago, one can assume that the primitive earth or initial crust amounted to a patchwork of hostile environments in which a cell prototype, having apparently some properties in common with the actual prokaryotes, was born. This crucial process took place 3.7–3.8 billion years, during the early stages of the Archaean: the first eon, corresponding to the period of time encompassing the origin of earth up until 2.5 billion years ago. This dating is supported by the fact that vestigial cells have been discovered in rocks 3.5 billion years old.

Two fundamental questions can now be addressed:

- Did the primordial cells originate from multiple streams or from a single ancestor? In other words, was early life already characterized by a certain degree of biodiversity?
- What were the characteristics of the proto-biotope(s) which has (have) enabled the establishment of cellular life?

These questions will be discussed in detail in other articles in EOLSS on-line, notably those by Michael Gross, Nicolas Glansdorff, and Patrick Forterre, but the idea has been formulated that perhaps the emergence of cellular organization was quickly succeeded by a rapid diversification, forced by the existence of environments displaying properties close to those defining the biotopes which are now known as extreme biotopes. Alternatively, perhaps the primordial cell or microorganism originated in a unique and specific environment and has seen slow and progressive modifications, these being modulated by the rapid global changes and the catastrophic processes responsible for the diversity which can be observed nowadays. However, the remarkable diversity in the present world system may represent only 0.1 percent of all species that ever inhabited the earth! The explosion of multicellular and skeleton-forming species succeeding the Ediacaran fauna and characterizing the most recent eon, the Phanerozoic, about 500 millions years ago, underlines the importance of global and rapid changes—like, for example, the emergence of high concentrations of oxygen in the atmosphere at that time—in the evolution and diversification of animal species. The fact that only 0.1

percent of “historical” overall diversity still exists nowadays does not mean that the biodiversity is decreasing globally. Although this is a matter of controversy, especially regarding the real number of species inhabiting our earth, there is some evidence that global diversity is continually increasing, the speciation rate apparently remaining higher than the extinction rate. The idea is supported in part by the fact that young fossil sites are generally richer in species than older ones.

One can assume that the wet environments, perhaps the cradle of primordial cells, were probably quite diversified in their hostile properties due to their tumultuous origin. It is quite possible that various extremophiles have been the earliest colonizers of our planet and that they were followed by more “normal” organisms, progressively adapted to environments evolving slowly towards biotopes displaying a set of relatively benign properties as defined earlier. If this scenario is correct, the extremophiles could therefore be considered as relics, still colonizing environments which have escaped the process of anthropomorphic normalization.

The somewhat controversial phylogenetic tree of microorganisms (see “Phylogeny of extremophile organisms,” EOLSS on-line, 2002) has been reconstituted from the analysis of sequence differences in ribosomal RNA. Based on the pioneering work of Carl Woese, it indicates that thermophilic species are very close, in terms of phylogenetic distances, to the point of separation between bacteria and archaea. This has given rise to the idea that the primordial cell was a thermophile.

2. Effects of Extreme Conditions on Cellular Components

Once again, we may take as reference points cellular components typical of human cells, or of microorganisms experiencing non-extreme conditions of temperature, pH, salinity, pressure, and concentration of xenobiotic substances. To analyze the adaptation processes one has to define the effects of certain physico-chemical parameters on various cellular compounds. It seems reasonable to focus primarily on the basic components of a cell, which are the membranes, the nucleic acids, and the proteins. We select these macromolecular and supramolecular structures as indicators of adaptation because they are stabilized by weak interactions, which are themselves very sensitive to the properties of the environment. Moreover, they are obviously the main actors of cellular life, controlling the exchanges between the intracellular space and the extracellular environments, cell division, cell structure, and cell chemistry.

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Biographical Sketch

Charles Gerday received a B.Sc. degree in Chemistry in 1961 from the University of Liège in Belgium and was appointed research assistant in the Department of Biochemistry, Faculty of Medicine in the same university. In 1964, he became lecturer at the Department of Biochemistry of the University of Montreal (Canada), and obtained a Ph.D. degree in 1967 with a thesis focusing on the synthesis of tritium labeled, optically active amino acids. Back in Belgium in 1967, he was appointed successively lecturer and senior lecturer in the Department of General Biology at the University of Liège where his research explored the regulation of muscle contraction by calcium-binding proteins. In 1972, he was a post-doctoral research fellow in the Laboratory of Molecular Biophysics at the University of Oxford. From 1981, his research activities were centered on the molecular adaptation of enzymes produced by cold-adapted organisms such as fish and bacteria originating from the Antarctic. He participated to several expeditions in the Antarctic. In 1988, he was appointed Professor of Biochemistry and Head of the Laboratory of Biochemistry at the University of Liège. He is the author of 160 original publications, and has been Chairman of the Belgian Biophysical Society since 1994.