Methods in ENZYMOLOGY

Volume 331

Hyperthermophilis Ensymes Part B

Bafford by

Michael W. W. Adams Robert M. Kelly



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Volume 331

Hyperthermophilic Enzymes Part B

EDITED BY

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Preface

More than thirty years ago, the pioneering work of Thomas Brock of the University of Wisconsin on the microbiology of hot springs in Yellowstone National Park alerted the scientific community to the existence of microorganisms with optimal growth temperatures of 70°C and even higher. In the early 1980s, the known thermal limits of life were expanded by the seminal work of Karl Stetter and colleagues at the University of Regensburg, who isolated from a marine volcanic vent the first microorganisms that could grow at, and even above, the normal boiling point of water. Subsequent work by Stetter and several other groups have led to the discovery in a variety of geothermal biotopes of more than twenty different genera that can grow optimally at or above 80°C. Such organisms are now termed hyperthermophiles.

Initial efforts to explore the enzymology of hyperthermophiles were impeded by the difficulty of culturing the organisms on a scale large enough to allow the purification of specific proteins in sufficient quantities for characterization. This often meant processing hundreds of liters of nearly boiling fermentation media under anaerobic conditions. In addition, relatively low biomass yields were typically obtained. Nevertheless, the first "hyperthermophilic enzymes" were purified in the late 1980s. It was demonstrated that they are, indeed, extremely stable at high temperatures, that this is an intrinsic property, and that they exhibit no or very low activity at temperatures below the growth conditions of the organism from which they were obtained. At that time it was difficult to imagine how quickly the tools of molecular biology would make such a dramatic impact on the world of hyperthermophiles. In fact, it was unexpected that the recombinant forms of hyperthermophilic enzymes would, to a large extent, correctly achieve their active conformation in mesophilic hosts grown some 70°C below the enzyme's source organism's normal growth temperature. This approach provided a much-needed alternative to large-scale hyperthermophile cultivation. With the ever-expanding list of genomes from hyperthermophiles that have been or are being sequenced, molecular biology provides universal access to a treasure chest of known and putative proteins endowed with unprecedented levels of thermostability.

In Volumes 330, 331, and 334 of *Methods in Enzymology*, a set of protocols has been assembled that for the first time describe the methods involved in studying the biochemistry and biophysics of enzymes and proteins from hyperthermophilic microorganisms. As is evident from the various chapters, hyperthermophilic counterparts to a range of previously stud-

ied but less thermostable enzymes exist. In addition, the volumes include descriptions of many novel enzymes that were first identified and, in most cases, are still limited to, hyperthermophilic organisms. Also included in these volumes are genomic analyses from selected hyperthermophiles that provide some perspective on what remains to be investigated in terms of hyperthermophilic enzymology. Specific chapters address the basis for extreme levels of thermostability and special considerations that must be taken into account in defining experimentally the biochemical and biophysical features of hyperthermophilic enzymes.

There are many individuals whose pioneering efforts laid the basis for the work discussed in these volumes. None was more important than the late Holger Jannasch of Woods Hole Oceanographic Institute. His innovation and inspiration opened a new field of microbiology in deep-sea hydrothermal vents and provided the research world access to a biotope of great scientific and technological promise. Holger will be remembered in many ways, and it is a fitting tribute that the first genome of a hyperthermophile to be sequenced should bear his name: *Methanocaldococcus jannaschii*. We wish to recognize Holger's pioneering efforts by dedicating these volumes to him.

MICHAEL W. W. ADAMS ROBERT M. KELLY

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