

# Early Days in Yellowstone Microbiology

Microbiology in Yellowstone at first focused on the basic science and ecology but gradually has expanded in scope

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y research in Yellowstone National Park began in the early 1960s as part of a larger interest in basic microbial ecology (Fig. 1). At the time, I was interested in simple ecosystems where I could study microorganisms in their natural environments. In particular, the hot springs in Yellowstone seemed to be steady-state systems containing large and readily accessible microbial populations. These "experiments in nature" soon led me to discover they are a habitat for bacteria that grow at high temperatures.

This Yellowstone experience provides a good example of the value of basic research, a fact that officials in the White House highlighted in 1991. During budget discussions with members of the House of Representatives that year, D. Allan Bromley, the director of the Office of Science and Technology Policy and the chief science advisor to President George Bush, made the following statement:

"Different kinds of research and development tend to have different kinds of returns. With basic research—the majority of which is done by individual scientists and small groups of scientists at universities-it is very difficult to predict when, where, and to whom the returns will eventually accrue. Yet even work that can seem highly abstract can have surprisingly immediate impacts. To take just one example, in 1968 Thomas Brock, a microbiologist at the University of Wisconsin, discovered a form of bacteria in the thermal vents of Yellowstone that can survive at very high temperatures. From these bacteria an enzyme was extracted that is stable at near-boiling temperatures. Nearly two decades later this enzyme proved

to be vital in the process known as the polymerase chain reaction, which is . . . the basis of a multimillion dollar business with applications ranging from the rapid diagnosis of disease to forensic medicine."

Because pressures to shift research funds from basic into applied research remain strong, those of us who agree with Bromley about basic research must repeatedly emphasize his ideas to federal officials and the Congress.

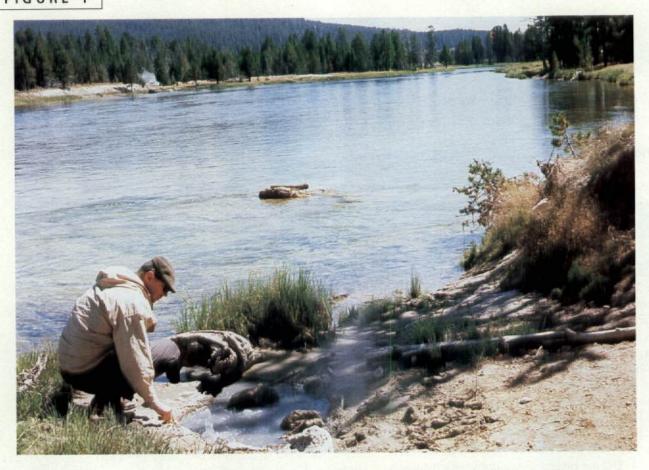
# Yellowstone Research Led from Photosynthesizers to Thermophiles

My early work in Yellowstone dealt primarily with the distribution and activity of photosynthetic microbes—primarily cyanobacteria—along the thermal gradients of hot spring outflows. Although the upper temperature limit for such phototrophs is about 70–73°C, many other nonphototrophic bacteria live at much higher temperatures in these same habitats.

In 1965, along the outflow channel of Octopus Spring, I first noticed masses of pink filamentous bacteria flourishing in water at 85– 88°C. Repeated attempts to culture these pink bacteria continue to prove unsuccessful. However, from a sample taken farther downstream, at 73°C, my colleague Hudson Freeze and I isolated an organism that we called *Thermus aquaticus*. For some years, *T. aquaticus* was the most extreme thermophile available in pure culture.

At first, I studied only those high-temperature bacteria that form macroscopic accumulations in the outflow channels of boiling springs. However, by 1967 I began using a simple immersion slide technique to reveal bacteria at the micro-

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Early Yellowstone microbiology: T. D. Brock sampling a small hot spring along the Yellowstone river, 22 August 1964.

scopic level in numerous boiling and superheated pools in Yellowstone.

With this same technique, Thomas Bott in 1968 began directly measuring growth rates of these microorganisms in their natural habitats. With doubling times of 2 to 6 hours in boiling water, these bacteria were not struggling to survive but were thriving at these high temperatures. This idea was confirmed by radioisotope studies. The fact that these organisms are optimally adapted to the temperatures at which they live has significant evolutionary implications.

# Practical Implications of Thermophile Discoveries

As this research progressed, I and others came to realize the broader implications of what we were doing. After one of our reports on the Yellowstone findings appeared as a lead article in Science in 1967, a number of biochemists from universities and industries asked us if they could use these organisms as sources of thermostable enzymes.

Although I sent out many cultures of *T. aquaticus*, the focus of most researchers in industry was then on extracellular enzymes, mainly proteases. However, *T. aquaticus* is not an appropriate organism for such enzymes. Later, the focus of research on *T. aquaticus* shifted to enzymes that act on DNA, such as restriction endonucleases and DNA polymerases. After the polymerase chain reaction (PCR) was discovered, and the value of *T. aquaticus* DNA polymerase came to be appreciated, interest boomed.

In the 22 December 1989 issue of Science magazine, the editors established a new award,

called "The Molecule of the Year" award. *Taq* polymerase from *T. aquaticus* was designated the first awardee.

# Additional Reasons for Studying Thermophiles

There are several strong justifications for conducting research on organisms that grow at high temperatures: (i) insights into basic microbial ecology, (ii) discoveries of unusual taxonomic interest and about microbial evolution, (iii) a means for modelling environmental pollution, and (iv) useful applications for industrial enzymology.

The evolutionary and taxonomic aspects of this research on thermophilic microorganisms led me in another direction. Among the interesting Yellowstone habitats are those that are both hot and very acidic, with high levels of sulfuric acid. Extensive microbial populations are found in such environments. Moreover, it is easy to obtain pure cultures from them.

In 1970-71, we described two new organisms from such hot, acidic environments, Thermoplasma acidophilum and Sulfolobus acidocaldarius. For some years, the main interest in these organisms was physiological and biochemical. However, in the late 1970s, Carl Woese of the University of Illinois reported that these two microorganisms are related to the methanogenic bacteria, thus greatly broadening his archaebacteria concept. Soon interest in

thermoacidophiles broadened significantly, with other researchers, particularly Wolfgang Zillig and Karl Stetter in Germany, exploring other hot, acidic habitats for additional examples of archaebacteria.

This work was exciting, but my own focus remained ecological, since the hot springs in which these organisms lived are ideal habitats for studying basic questions in microbial ecology. In the early and middle 1970s, we made direct measurements of the growth rates of these thermoacidophiles in their natural environ-

ments, something quite difficult to do with most microbes. These organisms grow surprisingly rapidly under these extreme environmental conditions.

# Deep-Sea Vents Change Outlook on Thermophiles

Although my early studies at Yellowstone may have struck many microbiologists as being somewhat "exotic," the discoveries of unusual microorganisms associated with the deep sea vents has helped to changes those attitudes. Compared to the exceedingly expensive and complex research conducted on microorganisms from thermal vents, studies at Yellowstone remain simple and inexpensive. Techniques developed in Yellowstone could be used with modifications on the vents. Further, the vents with temperatures much higher than 100°C can be used to assess the upper temperature limits for life. Pure cultures that grow well at temperatures significantly higher than 100°C are now available from such vents.

It was not until the advent of PCR that widespread attention really focused on "extremophiles." Not only has the biotechnology industry discovered Yellowstone National Park, but the park has discovered biotechnology. Never before has an industry profited directly from living creatures taken from a national park, and the Yellowstone administrators are interested in tapping into other forms of this naturally derived largesse.

Yellowstone, of course, has no monopoly on thermophiles. *T. aquaticus*, for instance, can be found in hot water heaters of many buildings. However, the park provides an accessible location with a wide variety of readily available

thermal habitats.

I do not object if commercial enterprises agree to develop products and pay royalties for use of materials that they derive from places such as Yellowstone National Park. My principal concern is that a focus on money might damage or undermine basic research efforts there. Important fundamental questions about microbial ecology and evolution in high-temperature environments can only be studied in such settings. Regardless of any commercially related activity in this same setting, such research should continue to be actively encouraged.

How can officials responsible for overseeing Yellowstone encourage basic research? When administrators were developing a "master plan" for the park 25 years ago, they asked me to suggest

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thermal areas that might be set aside permanently for scientific research. They were seeking to preserve some areas from development and retain them for use by researchers without the fear that road construction or tourist development would disrupt or destroy special habitats. As far as I know, that master plan was never completed (see Feature, p. 147). Nonetheless, something should be done to set aside suitable areas in Yellowstone National Park for long-term basic research on biology in high-temperature habitats.

#### SUGGESTED READING

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