ISSN 0026-2617, Microbiology, 2009, Vol. 78, No. 3, pp. 381–383. © Pleiades Publishing, Ltd., 2009.
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CRITICISM

A Review of the Monograph by Yu.A. Trotsenko and V.N. Khmelenina *Extermofil'nye metanotrofy* (Extremophilic Methanotrophs) (Pushchino: ONTI PNTs RAN, 2008, 206 pp.)

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DOI: 10.1134/S0026261709030199

The history of the investigation of methanotrophic bacteria, which are unique prokaryotic microorganisms capable of utilizing methane as the sole source of carbon and energy, spans more than a hundred years. Accumulation of the knowledge about methanotrophs passed through successive stages, including investigation of the morphology and ultrastructure of their cells, study of the key enzymes and metabolic pathways, evaluation of biotechnological potential, study of their ecology and activity in natural and anthropogenic ecosystems, and, finally, analysis of genomes and deciphering of the genetic potential of these organisms.

At the boundary of two centuries, in 2001, the Russian readers got the opportunity to acquaint themselves with the splendidly illustrated monograph by V.F. Gal'chenko *Metanotrofnye bakterii* (Methanotrophic Bacteria); this book summarized the voluminous fundamental knowledge accumulated about methanotrophic bacteria in the 20th century. The major part of this knowledge was obtained in studies of neutrophilic mesophilic methanotrophs. Some thermophilic, psychrophilic, and alkaliphilic representatives of the group were already known at that time. However, a breakthrough in their investigation occurred in the last decade. The focus of scientific interest shifted toward environments with extreme physicochemical characteristics, which became an inexhaustible source of a novel taxonomic, physiological, and metabolic diversity of methanotrophic bacteria. The boundaries of the growth conditions of known methanotrophs have been moved apart considerably, and currently correspond to the temperature range of 0 to 65°C, the pH range of 0.8 to 10.5, and the salinity range of 0 to 15%. Moreover, new methanotrophic organisms discovered in extreme ecosystems display a unique cell ultrastructure, specific metabolite pools, and novel mechanisms of adaptation. Detailed study of these peculiarities has been promoted by the application of the newest methods of genomics and proteomics. Molecular data is expanding that indicates the existence in nature of unknown forms of methanotrophs that continue to elude laboratory cultivation attempts. The pool of information accumulated over the last decade changes considerably our view of the diversity and biology of methane-oxidizing microorganisms and requires a new generalization. This task is fulfilled by the book *Extermofil'nye metanotrofy* (Extremophilic Methanotrophs), which has accumulated and systematized newest data on the ecophysiology, taxonomic, and structural–functional diversity of aerobic methanotrophic bacteria of various extreme ecosystems. The task taken on by the authors also included consideration of the mechanisms of adaptation and survival of methanotrophs under the impact of high and low temperatures, pH, and osmolarity.

The book consists of five chapters. The first chapter serves as an introduction and acquaints the reader with the miles tones in the investigation of methanotrophy and with the taxonomic diversity and specific metabolic features of methanotrophic bacteria. In spite of the introductory character of this chapter, it is meant for a prepared reader who already has basic knowledge about the key enzymes of methanotrophs and the fundamentals of their catabolism and anabolism. The chapter provides information on the important latest findings, including the unraveled transcription regulation mechanisms of membrane-bound and soluble methane monooxygenases (MMOs), discovery of two *iso*-forms of membrane-bound MMO differing in their affinity to methane in type II methanotrophs, deciphering of the three-dimensional structure of the enzymatic supercomplex formed by the membrane-bound MMO and methanol dehydrogenase of *Methylococcus capsulatus* Bath, and discovery of tetrahydrofolate- and tetrahydromethanopterin-dependent pathways of formaldehyde oxidation in methanotrophs. The first chapter also summarizes the current views on the nature of obligate methanotrophy and the biochemical basis of this phenomenon. In spite of the data from the analysis of the genomes of *Methylococcus capsulatus* Bath and *Methylomonas* sp. 16A, which became available several years ago, no final answer has yet been given as to the causes underlying obligate methanotrophy. Probably, the solution will be found in the course of comparative analysis of the genomes of the first known facultative methanotroph *Methylocella silvestris* BL2 and its close phylogenetic relative, the obligate methanotroph *Methylocapsa acidiphila* B2.

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The second chapter is devoted to thermophilic and thermotolerant methanotrophs, their diversity, thermoadaptation mechanisms, and systems of antioxidant defense. This ecophysiological group of methanotrophs is long known, since the description in 1966 of the moderately thermophilic *Methylococcus capsulatus* strain Texas, and it is constantly supplemented with new taxa. Over the last decade, microbiologists managed to obtain in a pure culture and validate the earlier described but permanently elusive hot spring inhabitant *Methylothermus thermalis.* An exiting event was the recent description by three independently working research teams of the first extremely thermoacidophilic methanotrophs, belonging to the phylum *Verrucomicrobia.* The entire framework that has been constructed to describe methanotrophy originated from the study of the *Proteobacteria* representatives; therefore, extension of the research field into another phylogenetic branch of the domain *Bacteria* marks the beginning of a new stage of investigation of this unique process and the microorganisms driving it. A separate subdivision of chapter 2 is devoted to comparative consideration of genomic predictions and reconstructions of metabolisms of the thermophilic methanotrophs belonging to the *Proteobacteria* and *Verrucomicrobia*: *Methylococcus capsulatus* Bath and '*Methylacidiphilum infernorum*'. The final part of the chapter provides detailed analysis of the experimental data obtained by the authors of the book: data on cytobiochemical rearrangements in cells of thermophilic and thermotolerant methanotrophs, on the role of sucrose in thermoadaptation, on the synthesis of a melanin pigment in *Methylocaldum szegediense*, and on antioxidant defense systems.

Having considered thermophiles, the authors switch to a contrasting ecophysiological group, namely, to methanotrophs of the psychrosphere and cryolithosphere. These microorganisms are much less studied than thermophiles. Among the reasons for this fact are the low growth rates, which make obtaining pure cultures and conducting any experiments extremely labor- and time-consuming. As a result, the list of the described taxa of obligately psychrophilic methanotrophs is extended much more slowly than that of thermophilic methanotrophs. Today, this list is the same as a decade ago and includes only *Methylobacter psychrophilus* and *Methylosphaera hansonii.* Essential new data that could explain the reasons of preference of low temperatures by these organisms are also lacking. However, considerable advances have been made over the last decade in the investigation of psychroactive methanotrophs, which are active enough in the temperature range of 10–15°C. The book lists the neutrophilic methanotrophic bacteria from the soils of tundra, deep subsurface granite deposits of the Finnish–Scandinavian shelf, and permafrost soil of the Kolyma Lowland, as well as moderately acidophilic methanotrophs of the northern wetlands. It should be noted that the role of representatives of this ecophysiological group in the biosphere is much greater than the role of thermophiles, since it is the cold ecosystems that are the main sources of the methane emission. However, the metabolic peculiarities and activity regulation mechanisms of the methanotrophs of cold ecosystems remain poorly studied, representing a vast field for future investigations.

The fourth chapter is, in a sense, the culmination of the authors' narration on the history of investigations on extremophilic methanotrophs and the beloved creation of the authors. It is devoted to microbial agents of methane oxidation inhabiting (hyper)saline and alkaline ecosystems. The mere existence of methanotrophic organisms adequately adapted to the conditions in these habitats was for a long time an open question. Only the use of the highly sensitive radioisotopic method made it possible to prove the occurrence of methane oxidation processes in saline and alkaline water bodies. The subsequent isolation of halo- and alkaliphilic methanotrophs from samples of bottom sediments and water of these ecosystems provided the authors the opportunity to study the adaptation mechanisms of these unique bacteria, and this opportunity was excellently used by them in a series of many-year studies. The fourth chapter of the reviewed monograph provides the reader with an overview of the results of the authors' studies on the peculiarities of metabolism of halo- and alkaliphilic methane-oxidizing bacteria, their osmoadaptation mechanisms, structure of the S-layers revealed on the outer surface of their cell walls, and phospholipid and fatty acid composition. Very well written is the section devoted to osmoprotectants of halophilic and halotolerant methanotrophs. Not only does it list the compounds playing the osmoprotective role, it also contains detailed analysis of the pathways of biosynthesis of these compounds in the cells of methanotrophs. Particular attention is given to ectoine, the pathways of its biosynthesis, deciphering of the structure and organization of the *ect* genes in methanotrophs, and to regulatory aspects of the synthesis of this osmoprotectant. Special attention is given by the authors to the bioenergetic aspects of osmoadaptation in methanotrophs. On the whole, the chapter makes a strong impression on the reader, since the presented current picture of the biology of halo- and alkaliphilic methanotrophs is detailed and integral, and this is primarily due to the research conducted by the authors of the book.

In response to the realities and demands of the present day, the fifth chapter provides a comprehensive list of potential fields of application of extremophilic methanotrophic bacteria, of the compounds produced by them, or of the knowledge about mechanisms of methane oxidation. Even the most critically minded reader standing up for the priority of the development of applied sciences will agree, upon reading this chapter, that all areas of practical application of the unique metabolism of extremophilic methanotrophs have been found due to most detailed fundamental investigations.

The Supplement to the book provides, for convenience, short descriptions of the new taxa of extremo-

philic and extremotolerant methanotrophs. The reader should however be aware that detailed analysis of the entire diversity of cultured and uncultured methanotrophs, as well as of the ecology of methanotrophs and their functioning in natural ecosystems, was not among the tasks of the reviewed book. A more complete knowledge on these issues is to be gained from publications in periodical scientific journals.

On the whole, the book is the result of a giant work on the generalization of the latest achievements in the studies on the biology and metabolism of methanotrophic bacteria, primarily, their extremophilic representatives. The book is particularly valuable due to the fact that a considerable part of the scientific data it presents are first-hand data obtained by the authors, who over many years conducted studies with all, without exception, ecophysiological groups of methanotrophs described in the book. This is the best guarantee of professional in-depth analysis of the presented data. The book is well illustrated. It is to attract the interest of both professional biochemists and microbiologists and those who strive to join their ranks: students, Ph.D. students, and junior scientists.