OUTER STRUCTURES OF BACTERIA

Edited by

WOUTERA VAN ITERSON

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A Hutchinson Ross Benchmark® Book

VNR VAN NOSTRAND REINHOLD COMPANY

Benchmark Papers in Microbiology, Volume 18
Library of Congress Catalog Card Number: 83-10800
ISBN: 0-442-28832-8

Manufactured in the United States of America.

Published by Van Nostrand Reinhold Company Inc.
135 West 50th Street
New York, New York 10020

Van Nostrand Reinhold Company Limited
Molly Millars Lane
Wokingham, Berkshire RG11 2PY, England

Van Nostrand Reinhold
480 Latrobe Street
Melbourne, Victoria 3000, Australia

Macmillan of Canada
Division of Gage Publishing Limited
164 Commander Boulevard
Agincourt, Ontario MIS 3C7, Canada

15 14 13 12 11 10 9 8 7 6 5 4 3 2 1

Library of Congress Cataloging in Publication Data
Main entry under title:
Outer structures of bacteria.
(Benchmark papers in microbiology ; 18)
Includes indexes.
1. Bacterial cell walls—Addresses, essays, lectures.
2. Bacteria—Morphology—Addresses, essays, lectures. I. Iterson, Woutera van, 1914– . II. Series.
[DNLM: 1. Bacteria—Cytology—Collected works. 2. Cell wall—Collected works. 3. Pili, Bacterial—Collected works. 4. Flagella—Collected works. W1 BE517M v.18 / QW 51 094]
QR77.3.097 1984 589.9'0875 83-10800
ISBN 0-442-28832-8

SERIES EDITOR'S FOREWORD

In the 30-odd years since the electron microscope came into general use, there has been a profound change in our concepts of both the inner and outer parts of the bacterial cell. An entirely new understanding of the organization of the bacterial cell resulted from improvements in the electron microscope itself and especially from the introduction of thin sections and positive and negative staining techniques and the introduction of the entirely different freeze-etch technique, which occurred in the late 1960s and early 1970s. In addition, the development of physical fractionation techniques followed by gentler methods permitted a new approach, making it possible to relate structure to function.

Woutera van Iterson. In many papers on cytology it is the selection of the typical photograph that constitutes the prime evidence for the structure reported. To reproduce lengthy papers containing many photographs of the relevant structures would require many volumes to cover the important observed structures. Further, not all bacteria are alike and the structures found in one kind may be completely absent in another. Consequently, the selection of data (consisting almost entirely of electron micrographs) is critical and requires vast experience in the artifacts that can be produced and in the kind of information that can be derived from such micrographs. If, however, one selects only the outstanding micrographs of structures, then increased discussion is also required.

Dr. van Iterson has excerpted many more papers than is usual in a Benchmark volume and because of the nature of the subject she has included a review of an enormous amount of related literature. Indeed, she has produced a monographic discussion of each of the observed structures and has reviewed the literature associated with each throughout the volumes, covering not only cellular cytology but biochemistry and physiology as well. These volumes represent a masterpiece treatment by a recognized world authority of the subject and will stand as benchmarks for years to come.

WAYNE W. UMBREIT

PREFACE

All information is imperfect.
We have to treat it with humility.
That is the human condition . . . .

J. Bronowski, *The Ascent of Man*

In this volume, *Outer Structures of Bacteria,* and its companion volume, *Inner Structures of Bacteria,* I have attempted to review the ideas on the ultrastructure of bacteria as they have developed during the more than 35 years that the electron microscope has been fruitfully applied to structure analysis. During its development, structure analysis proved to be a challenge to biochemistry and stimulated the impressive expansion of molecular biology. Consequently, much more is known about the biochemical or molecular biological aspects of the subjects treated in these volumes than about their ultrastructure.

It is not always possible to concentrate on ultrastructure without alluding to some of the biochemical findings. These findings are presented in a condensed form without giving due tribute to the host of difficult and sophisticated biochemical studies on which the information is based. The first few sections of this volume, therefore, may produce an unsatisfactory or even somewhat distorted picture. For instance, more progress was made during the biochemical dissection of the cell wall layers than during the study of the integrated wall structure at considerable resolution of the electron microscope. To obtain a well-balanced picture of these bacterial cell wall layers, some readers may find greater satisfaction in studying a work such as *Microbial Cell Walls and Membranes* (Rogers, Perkins, and Ward, 1980).

My aim has been to give a picture of the bacterium as an integrated whole, starting from its inside—the knot of the DNA thread in the cell's interior—and progressing gradually to the cell's outside. Because not all bacteria are alike, some peculiarities are discussed as well.

These two volumes certainly are not always illustrated with the most beautiful electron micrographs available. I have chosen those illustrations and parts of published papers that in some way portray steps in the advancement of knowledge.

ACKNOWLEDGMENTS

I would like to acknowledge Professor Wayne W. Umbreit's encouragement throughout this work, and further, the fact that several scientists have helpfully read through various chapters. (I hesitate to mention them by name because of all the alterations and rewritings I did afterwards.) Dr. H. L. van Vierssen Trip in Ottawa patiently typed and retyped and improved the style of the manuscripts. The language has been further brushed up by the staff of Van Nostrand Reinhold and partly by Professor Irene Manton, FRS, University of Leeds. Where my ways of expression remain outlandish, they must be attributed to my ecological niche on the North Sea shore. Librarians at several university libraries have been of immeasurable help to me throughout the many years of reading.

To all of those who helped me so much, I would like to express my appreciation and thankfulness.

WOUTERA VAN ITERSON

REFERENCE

Rogers, H. J., H. R. Perkins, and J. B. Ward, 1980, *Microbial Cell Walls and Membranes,* (Chapman and Hall, London and New York, 564p.

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All bacterial species have a fixed shape characterized by three basic cell forms: spherical, rod-shaped, or helical (spirillum- or comma-shaped). The degree of rigidity has not been accurately established yet. During movements, in particular gliding and flexing movements, some bacteria seem to display a certain amount of flexibility.

The bacterial cell envelope consists of two components: a protective cell wall and an osmotically sensitive cytoplasmic membrane. When the cell wall is absent, as for instance after removal by means of lysozyme, the cell contents, including both cytoplasm and nuclear material, are liable to swell or shrink and to change shape. The cytoplasmic membrane, then, seems to adjust itself passively to the

new cell shape. The protective cell wall, besides preserving the cell shape, prevents rupture of the cytoplasmic membrane by excessive extension in an environment of low osmotic pressure. The wall, indeed, contains a shape-preserving component, the so-called rigid layer, that consists of peptidoglycan (murein). On its outer surface, the wall may carry a surface coat. Usually it has various structures to interact with the environment: It may be covered by material to adhere to surfaces, it has receptors for bacteriophages, and antigenic components that may contribute to the bacterium's virulence.

DISTINCTION BETWEEN CELL WALL AND CYTOPLASMIC SURFACE

Distinctions between cell wall and cytoplasmic surface were demonstrated early by Knaysi (1930) in plasmolyzed cells and later by Murray and Robinow (1952) in crushed cells. Robinow and Murray (1953) devised methods for distinguishing the cell wall from the cytoplasmic membrane in plasmolyzed gram-positive bacteria by means of the light microscope.

In the electron microscope, the separateness of cell wall and cytoplasm was studied in broken and plasmolyzed cells. Thus, Dawson (1949) illustrated detached cell walls of *Staphylococcus aureus* after breakage by shaking with glass beads in a Mickle disintegrator. Mickle disintegration, with similar results, was applied by Salton and Horne (1951*b*) to *Escherichia coli, Streptococcus,* and *Salmonella,* supplementing earlier work by the same authors (Salton and Horne, 1951*a*) in which heated bacteria had been ruptured by sudden exposure to distilled water. In sections and other more refined preparations for the electron microscope, the wall and cytoplasmic membrane could clearly be seen, and, as we shall see, many important details have been analyzed.

Emptied cell envelopes, as well as autolyzed cells, often retain the shape of the bacterium, thereby indicating that the cell wall must be a major determinant of cell shape.

DISTINCTION BETWEEN GRAM-POSITIVE AND GRAM-NEGATIVE CELLS

Long before the existence of the cell wall was ascertained by visual or chemical means, one of its characteristics was used in the classification of bacteria. The gram-reaction, introduced by Christian Gram in 1884, is based on differences in permeability of cell walls (Salton, 1964). After coloring with a dye-iodine complex, an organic