



SECOND EDITION

# *Diagnostic microbiology*

A textbook for the isolation and identification  
of pathogenic microorganisms

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## *Diagnostic microbiology*

## Preface to second edition

This, the second edition of *Diagnostic Microbiology*, represents a substantial revision of the first edition; yet the same purpose—the provision of a current reference text for medical microbiologists and a course text for college students—still remains.

We are highly appreciative of the constructive criticism and the letters of encouragement received since publication of the 1962 edition. It has not been possible, however, to follow the suggestion that the book be enlarged to allow comprehensive coverage in virology and to introduce parasitology. The reasons for this are twofold: first, the book would lose its present identity; second, many good books covering these areas are available.

All chapters covering the isolation and identification of the pathogenic microorganisms have been virtually rewritten to include current concepts and recommended methods. The chapter on the mycobacteria in particular has been expanded considerably.

The extensively revised section on the mycoses now includes the dermatophytes and gives far greater coverage and depth

to the pathogenic fungi. The sketches of the main species discussed are new and have been prepared by us from photomicrographs. It is hoped that these illustrations will assist trained microbiologists as well as those who are in training in recognizing these increasingly important pathogens.

The revised chapter on antibiotic susceptibility testing provides the new and effective methods in use. A short chapter on fluorescence microscopy has been added to the serological procedures, and the section on media and reagents includes several important additions.

We acknowledge the many kindnesses and invaluable technical help of Dr. George Kubica, Dr. Fritz Blank, Dr. Harriet Vera, Miss Elizabeth King, Dr. Max Moody, and numerous others, all of whom have been most generous and stimulating.

We further wish to extend to our wives our sincere appreciation for their periodic stenographic assistance, their indulgence, and their encouragement.

W. Robert Bailey  
Elvyn G. Scott

## Preface to first edition

**D**iagnostic *Microbiology* is the first edition of a new series and not a revision of the former publication *Diagnostic Bacteriology*, the latest edition of which we revised (1958). This new title derives in part from the fact that the new volume includes microorganisms other than bacteria. The reader will note, for example, that the former *Society of American Bacteriologists* has now become the *American Society for Microbiology*.

Since this book is designed to be used as a reference text in medical bacteriology laboratories and as a textbook for courses in diagnostic bacteriology at the college level, the material has been consolidated and placed in separate parts and chapters. The selected sequence will be commensurate with the needs of both the diagnostician and the student.

For purposes of orientation in taxonomy and ready reference, an outline of bacterial classification has been included. For the student beginning diagnostic work, some pertinent background information is presented on the cultivation of microorganisms, the microscopic examination of microorganisms, and the proper methods for collecting and handling specimens.

A number of chapters include recommended procedures for the cultivation of both the common and the rare pathogens isolated from clinical material and should

serve to familiarize the microbiologist with the wide variety of pathogens that may be encountered. An additional chapter has been devoted to the methods employed in the microbiological examination of surgical tissue and autopsy material.

To effect further consolidation of the book's content, one part has been devoted to a series of chapters which cover the various groups of bacteria of medical importance—their taxonomic position, general characteristics, and procedures for their identification. The chapter on the enteric bacteria introduces the new classification of the family Enterobacteriaceae, outlines the group biochemical characteristics, and discusses the serological aspects. The chapter on the mycobacteria includes a discussion of the increasingly important unclassified (anonymous) acid-fast bacilli, giving the methods for their identification, certain cytochemical tests, and animal inoculation procedures.

The chapter on laboratory diagnosis of viral and rickettsial diseases includes a guide to the collection of specimens and offers recommendations for the appropriate time of collection. In the chapter on laboratory diagnosis of systemic mycotic infections, the biochemical approach in identifying the pathogenic fungi is brought to the reader's attention.

The remainder of the book includes

prescribed tests for the susceptibility of bacteria to antibiotics, serological procedures on microorganisms and patients' sera, and a technical section on culture media, stains, reagents, and tests, each in alphabetical sequence.

We would like to express our gratitude to Mrs. Isabelle Schaub and to Sister Marie Judith for committing the continuation of

the original publication, *Diagnostic Bacteriology*, to our care and responsibility. We also acknowledge the many kindnesses extended by a number of microbiologists and clinicians in permitting the use of published and unpublished materials.

W. Robert Bailey  
Elvyn G. Scott



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# Classification of the plant kingdom

## CLASSIFICATION OF THE PLANT KINGDOM\*

<b>Division</b>	<b>I</b>	<i>Protophyta</i> —Primitive plants
<b>Division</b>	<b>II</b>	<i>Thallophyta</i> —Molds and yeasts
<b>Division</b>	<b>III</b>	<i>Bryophyta</i> —Mosses
<b>Division</b>	<b>IV</b>	<i>Pteridophyta</i> —Ferns
<b>Division</b>	<b>V</b>	<i>Spermatophyta</i> —Seed plants

## CLASSIFICATION OF THE PRIMITIVE PLANTS

(Including the bacteria, related forms, the rickettsiae, and the viruses)

<b>Division</b>	<b>I</b>	<i>Protophyta</i>
<b>Class</b>	<b>I</b>	<i>Schizophyceae</i> —Blue-green algae
<b>Class</b>	<b>II</b>	<i>Schizomycetes</i> —Bacteria and related forms
<b>Order</b>	<b>I</b>	<i>Pseudomonadales</i> —Gram-negative, rod-shaped bacteria; usually motile with polar flagella; soil and water forms; some pathogenic species
<b>Suborder</b>	<b>I</b>	<i>Rhodobacteriineae</i> —Contain photosynthetic pigments; cells spherical, spiral, or rod-shaped
<b>Family</b>	<b>I</b>	<i>Thiorhodaceae</i> —Sulfur purple bacteria
<b>Family</b>	<b>II</b>	<i>Athiorhodaceae</i> —Nonsulfur purple and brown bacteria
<b>Family</b>	<b>III</b>	<i>Chlorobacteriaceae</i> —Green sulfur bacteria
<b>Suborder</b>	<b>II</b>	<i>Pseudomonadineae</i> —No photosynthetic pigments, but may produce water-soluble pigments; coccoid, rod-shaped, vibrio-like, and spiral forms
<b>Family</b>	<b>I</b>	<i>Nitrobacteraceae</i> —Autotrophic; oxidize ammonia to nitrites or nitrites to nitrates
<b>Family</b>	<b>II</b>	<i>Methanomonadaceae</i> —Autotrophic; oxidize methane, hydrogen, or carbon dioxide
<b>Family</b>	<b>III</b>	<i>Thiobacteriaceae</i> —Oxidize sulfur compounds; colorless sulfur bacteria

\*Based on the outline given in *Bergey's manual of determinative bacteriology*, ed. 7, Baltimore, 1957, The Williams & Wilkins Co.

**Division I**—cont'd

**Class II**—cont'd

**Order 1**—cont'd

- Family IV** *Pseudomonadaceae*—Usually heterotrophic; frequently oxidative; many plant and a few animal pathogens
- Family V** *Caulobacteraceae*—Cells attached to substrate by means of a stalk
- Family VI** *Siderocapsaceae*—Cells free-floating or attached to substrate by capsular material
- Family VII** *Spirillaceae*—Cells curved, vibrio-like to spiral-shaped
- Order II** *Chlamydobacteriales*—Colorless, gram-negative, algalike bacteria, usually ensheathed and often containing deposits of iron oxide; occur in trichomes; reproduction by swarm spores or by conidia; freshwater and saltwater forms
- Family I** *Chlamydobacteriaceae*—Motile conidia, trichomes often branched
- Family II** *Peloplocaceae*—Trichomes unbranched
- Family III** *Crenotrichaceae*—Nonmotile conidia
- Order III** *Hyphomicrobiales*—Gram-negative forms which may be ovoid, ellipsoidal, spherical, or pyriform; multiplication by budding or by budding and longitudinal fission; mud and freshwater forms; some parasitic
- Family I** *Hyphomicrobiaceae*—Buds borne on filaments
- Family II** *Pasteuriaceae*—Buds sessile
- Order IV** *Eubacteriales*—The true bacteria; gram-positive and gram-negative rods and cocci; motile forms possess peritrichous flagella; reproduction by transverse binary fission; wide distribution; saprophytes, parasites; many pathogens
- Family I** *Azotobacteraceae*—Gram-negative rods; soil forms; nonsymbiotic nitrogen fixers
- Family II** *Rhizobiaceae*—Gram-negative rods; symbiotic nitrogen fixers; soil forms; heterotrophic
- Family III** *Achromobacteraceae*—Gram-negative rods; water and soil forms; some chromogens
- Family IV** *Enterobacteriaceae*—Gram-negative rods; glucose fermented by all members; many species live in the intestines of man and other animals; many animal pathogens, some plant pathogens
- Tribe I** *Escherichieae*—Lactose fermented, usually in 48 hours, but delayed in some cases; protopectinase not produced
- Tribe II** *Erwinieae*—May produce protopectinase; plant pathogens
- Tribe III** *Serratieae*—Chromogenic forms producing prodigiosin
- Tribe IV** *Proteeae*—Lactose rarely fermented; urease produced
- Tribe V** *Salmonelleae*—Lactose rarely fermented; urease not produced
- Family V** *Brucellaceae*—Small gram-negative rods; often require growth factors on original isolation; CO<sub>2</sub> sometimes required on isolation, its presence usually enhances growth; parasitic and pathogenic for warm-blooded animals
- Family VI** *Bacteroidaceae*—Gram-negative rods with rounded or pointed ends; pleomorphic; found in intestinal tract and mucous membranes of warm-blooded animals; some pathogens
- Family VII** *Micrococcaceae*—Gram-positive cocci; gram-variability not infrequent; saprophytes and parasites, some pathogens; found on the skin, throat, and nasopharynx; some soil and marine forms

**Division I—cont'd**

**Class II—cont'd**

**Order IV—cont'd**

- |               |             |   |
|---------------|-------------|---|
| <b>Family</b> | <b>VIII</b> | <i>Neisseriaceae</i> —Gram-negative cocci occurring in pairs or masses; parasitic   |
| <b>Family</b> | <b>IX</b>   | <i>Brevibacteriaceae</i> —Gram-positive rods; varying from coccoid to long straight forms; found in dairy products, soil, and water   |
| <b>Family</b> | <b>X</b>    | <i>Lactobacillaceae</i> —Gram-positive cocci and rods; the lactic acid bacteria; found in the mouth and intestinal tract of animals, including man, also in foods and fermenting vegetable juices; some highly pathogenic                 |
| <b>Tribe</b>  | <b>I</b>    | <i>Streptococceae</i> —Gram-positive cocci; catalase-negative; some pathogenic forms; some require enriched media   |
| <b>Tribe</b>  | <b>II</b>   | <i>Lactobacilleae</i> —Gram-positive nonsporulating rods; some pathogens  |
| <b>Family</b> | <b>XI</b>   | <i>Propionibacteriaceae</i> —Gram-positive, irregularly shaped rods; active in carbohydrates; found in animal intestinal tract  |
| <b>Family</b> | <b>XII</b>  | <i>Corynebacteriaceae</i> —Gram-positive rods, frequently showing banding or beading with metachromatic granules; marked pleomorphism in some species; some chromogens; animal and plant pathogens; also found in soil and dairy products |
| <b>Family</b> | <b>XIII</b> | <i>Bacillaceae</i> —Gram-positive, spore-forming rods; aerobic to anaerobic; catalase-variable; mostly saprophytic; some species highly pathogenic; usually found in the soil   |
| <b>Order</b>  | <b>V</b>    | <i>Caryophanales</i> —Occurring in trichomes or shorter structures; found in water, decomposing organic matter, and the intestines of arthropods and vertebrates  |
| <b>Family</b> | <b>I</b>    | <i>Caryophanaceae</i> —Large trichomes and bacillary forms; nonsporulating; motile and nonmotile  |
| <b>Family</b> | <b>II</b>   | <i>Oscillospiraceae</i> —Trichomes vary in length; actively motile; parasitic forms   |
| <b>Family</b> | <b>III</b>  | <i>Arthromitaceae</i> —Nonmotile, sporulating forms   |
| <b>Order</b>  | <b>VI</b>   | <i>Actinomycetales</i> —The members form elongated cells with a tendency to branch; spores produced by some species; some species acid-fast; some are pathogenic; the majority are soil forms   |
| <b>Family</b> | <b>I</b>    | <i>Mycobacteriaceae</i> —Gram-positive rods and cocci; nonsporulating; one acid-fast genus; some animal pathogens; found in soil and dairy products   |
| <b>Family</b> | <b>II</b>   | <i>Actinomycetaceae</i> —Mycelium nonseptate, changing to septate and fragmenting; sporulating variously; some parasites; some acid-fast forms  |
| <b>Family</b> | <b>III</b>  | <i>Streptomyetaceae</i> —Vegetative nonfragmenting mycelium; conidia produced; mainly soil forms; several antibiotic-producing species; a few parasitic forms   |
| <b>Family</b> | <b>IV</b>   | <i>Actinoplanaceae</i> —Inconspicuous vegetative mycelium; reproduction by spores, some of which are motile; found in soil and fresh water  |
| <b>Order</b>  | <b>VII</b>  | <i>Beggiatoales</i> —Occurring mostly in trichomes; gliding forms; nonflagellated; do not possess chlorophyll; freshwater and saltwater forms; also found in soil and decomposing organic matter  |
| <b>Family</b> | <b>I</b>    | <i>Beggiatoaceae</i> —Individual cells in the trichomes not visible without staining  |
| <b>Family</b> | <b>II</b>   | <i>Vitreoscillaceae</i> —Colorless trichomes; gram-negative, gliding forms  |

**Division I—cont'd**

**Class II—cont'd**

**Order VII—cont'd**

**Family III** *Leucotrichaceae*—Long trichomes of short cylindrical cells; nonmotile; multiplication by gonidia

**Family IV** *Achromatiaceae*—Large spherical or oval unicellular organisms, containing sulfur droplets

**Order VIII** *Myxobacterales*—The slime bacteria; flexible rods, reproducing by binary fission; resting cells and fruiting bodies; mainly soil forms; some aquatic forms; some parasites

**Family I** *Cytophagaceae*—Flexible rods; no fruiting bodies or resting cells

**Family II** *Archangiaceae*—Resting cells are short rods; fruiting bodies irregularly shaped

**Family III** *Sorangiaceae*—Fruiting bodies in cysts

**Family IV** *Polyangiaceae*—Rod-shaped resting cells in cysts

**Family V** *Myxococcaceae*—Rods develop into spherical or ellipsoidal microcysts on fruiting

**Order IX** *Spirochaetales*—Slender, flexuous, spiral-shaped cells; no flagella; motility is by a flexing movement or whirling and spinning about the long axis; stained only with special stains; free-living, saprophytic and parasitic forms; reproduction by transverse fission

**Family I** *Spirochaetaceae*—Possess obvious protoplasmic structures; found in stagnant water and in intestinal tract of some molluscs

**Family II** *Treponemataceae*—Possessing no obvious protoplasmic structure; mostly parasitic in vertebrates; some pathogens

**Order X** *Mycoplasmatales*—Gram-negative, pleomorphic organisms; nonmotile; filaments break up into coccoid, filterable forms; nutritionally demanding; some pathogens

**Family I** *Mycoplasmataceae*—Aerobic to facultatively anaerobic; mostly human and animal sources; contains the pleuropneumonia-like organisms (PPLO)

**Class III** *Microtobiotes*—The smallest living things; obligate parasites; mostly intracellular; mostly filterable forms; require special methods of culture

**Order I** *Rickettsiales*

**Family I** *Rickettsiaceae*

**Family II** *Chlamydiaceae*

**Family III** *Bartonellaceae*

**Family IV** *Anaplasmataceae*

**Order II** *Virales*

**REFERENCE**

Breed, R. S., Murray, E. G. D., and Smith, N. R.: *Bergey's manual of determinative bacteriology*, ed. 7, Baltimore, 1957, The Williams & Wilkins Co.

## Part II

### Laboratory methods



