

LABORATORY MANUAL  
AND  
WORKBOOK FOR

**MICROBIOLOGY  
IN  
HEALTH  
AND  
DISEASE**

*Sixth Edition*

**ROBERT FUERST, B.S., M.A., Ph.D.**



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Laboratory Manual and Workbook for  
MICROBIOLOGY IN HEALTH AND DISEASE

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# PREFACE

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This laboratory manual has been updated from the earlier edition and all errors have been carefully removed; my apologies to those users who may find new errors resulting from the revision. The fourteenth edition of *Frobisher and Fuerst's Microbiology in Health and Disease*, revised by Robert Fuerst, is the text from which this manual has been prepared. However, the effectiveness of the manual is not limited to users of any specific text. The organization of the laboratory manual proceeds from introductory material on the microscope, morphology and staining of microorganisms to disinfection and sterilization, sanitation, immunity, and pathogenic microorganisms. This is a common sequence in teaching microbiology in many schools. The section on pathogenic microorganisms has been presented from the viewpoint of methods of transmission rather than by using morphologic characteristics as the primary organizational pattern. In preparing this manual the author has relied on his own experience in teaching this material as well as on the seemingly successful fifth edition which predated this revision. For clarity, each laboratory exercise is divided into Key Steps and Important Points.

The author believes that laboratory work in microbiology can be taught without exposing the students to highly pathogenic microorganisms, but that all cultures of microorganisms should be handled with caution. It is his belief that beginning students in microbiology have not developed their aseptic technique sufficiently to warrant the hazard of having them handle virulent cultures. The author also feels that one of the primary outcomes of laboratory experience in microbiology is the realistic development of aseptic technique. Therefore, the instructors should insist on rigid adherence to principles and practice of asepsis in the laboratory. There should be good carryover of the techniques learned in the laboratory to clinical situations involving the health professions.

The author is grateful to many co-workers who have helped develop these laboratory exercises by their comments and suggestions, to the innumerable students who have made criticisms over a period of years, and to the instructors who have used the previous edition and forwarded suggestions and comments to the publisher. Special appreciation is due to the originator of this manual, Lucille Sommermeyer, who regrettably chose not to participate in the preparation of this and the last two editions.

The format of the allergy clinic scratch test has been provided through the courtesy of Dr. Ben Fisch, whose encouragement and personal friendship have been invaluable to the author.

Thanks are extended also to Miss Becky Russo, who conducted some of the experiments.

The author also acknowledges with sincere gratitude the outstanding work by the staff of the W. B. Saunders Company, who were always ready to help and cooperate, and without whose expert workmanship this laboratory manual would not have been possible.

ROBERT FUERST

# NOTE TO INSTRUCTORS

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This laboratory manual has been prepared to help you in your busy program. To use it best, you must be thoroughly familiar with each experiment. To utilize the time allotted for laboratory periods to its maximum advantage, it is recommended that all supplies and equipment should be available and ready for use before the laboratory period begins. In some situations assistants will prepare and check required materials. In other situations students may be assigned certain tasks of preparing equipment. Laboratory instruction in microbiology requires a great deal of expert planning and organization in order that the students may utilize the time provided for maximum learning. The author has found it highly profitable to prepare a schedule before beginning the course. This schedule includes the days when media should be prepared for use by the students, days when media should be inoculated with cultures, when transfers of stock cultures should be made, and so on. Such a schedule, if carefully thought through, reduces the possibility of forgetting vital materials.

This revision presents some new illustrations which may or may not meet your approval as far as techniques in the laboratory are concerned; no illustrations were deleted from the last edition. These illustrations were included here because the author has taught many students how to inoculate, pipette, streak plates, etc., by these methods. They were found to be helpful tools in learning to *avoid contamination*. If your methods are slightly different from those shown in the illustrations they are certainly not wrong. Perhaps the student can learn how to do things in more than one way, but most important *learn what not to do* in the laboratory or in the hospital situation. It is hoped that other new illustrations and tables added throughout the manual will be of help to students.

Exercise 40 (Staphylococci and Streptococci) and Exercise 41 (Growth of Anaerobes) are entirely new. The importance of anaerobic infections in the health field can hardly be overemphasized. You will find Exercise 41 easy to perform, and the results are very rewarding.

Although this laboratory manual is too long and detailed for some courses in microbiology, the exercises are written so that if omission is necessary the continuity will not be lost. For the short courses in microbiology, parts of experiments might be omitted or some of the following exercises might be completely deleted: Exercises 8, 10-12,

vi / NOTE TO INSTRUCTORS

14, 15, 17-19, 26, 27, 30 and 31. Some of the work in Unit IV  
used as out-of-class assignments. The author believes that thi  
is comprehensive enough to meet the needs of the most extensi  
in microbiology given in schools of nursing and allied health  
as well as in many colleges and universities.

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course  
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# NOTE TO STUDENTS

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All laboratory manuals are only as good as the time, thought, and effort that is put into them by students. This laboratory manual has been arranged for your convenience. Space has been provided for recording results. All tables which could have been preconstructed have been prepared for you. Although it would be possible to complete this laboratory manual from available source materials, your knowledge of microbiology will grow into a useful, practical body of information only if you carry out each experiment as assigned, make your own observations, and record the results you observe. This entire laboratory manual has been prepared and selected from a wealth of available materials, and selection has been made of those materials which are essential to your practice in the health professions. The latest terminology has been applied throughout, and if it has changed since the last edition a footnote to this effect gives the former usage as well. Microbiology is a practical science, and the information learned in the course, and especially in the laboratory, will be important in the daily care of patients.



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# Unit 1

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INTRODUCTION  
TO THE USE OF  
THE MICROSCOPE

MORPHOLOGY OF  
BACTERIA,  
MOLDS, YEASTS  
AND PROTOZOA

STERILE  
TECHNIQUES  
AND THE  
PREPARATION OF  
MEDIA

STAINING  
TECHNIQUES

OTHER  
TECHNIQUES IN  
THE STUDY OF  
MICROORGANISMS

**TOPIC: GENERAL DIRECTIONS FOR WORK IN A MICROBIOLOGY LABORATORY**

- OBJECTIVES:**
1. To help the student understand the work and the functions of the microbiology laboratory.
  2. To present certain basic guide lines for laboratory activity.

KEY STEPS	IMPORTANT POINTS
1. Protect your health and the health of others.	<p>1.1. The reason you are taking this course and laboratory is first of all to learn to protect your health and the health of others against pathogenic microorganisms <i>as long as you shall live</i>.</p> <p>1.2. Every microorganism you are using in the laboratory must be considered a potential pathogen; you will learn that many are indeed pathogenic.</p> <p>1.3. You never eat, drink or smoke in the laboratory, nor do you bring any food or soft drinks into the laboratory.</p>
2. Keep the laboratory as neat as possible at all times.	<p>2.1. Because working space is limited and work in microbiology requires a certain amount of "elbow" room, you should bring only essential articles to the laboratory (laboratory manual, textbooks, pencils, pen). If possible, your overcoat should remain in the hall.</p> <p>2.2. Ask your laboratory instructor for a proper place for your purse, satchel, books, and so forth.</p> <p>2.3. A cluttered mess of test tubes, culture plates and slides not only is unsightly but also usually indicates that the worker is confused and disorganized. Therefore, you should discard materials you no longer need in the place designated by the instructor.</p>
3. Protect your clothing.	<p>3.1. Stains are used in microbiology and occasionally they are spilled accidentally. These will stain your clothes permanently. Therefore, you should wear a laboratory coat, smock or long plastic apron while you are in the laboratory. Lately, paper aprons (fireproof) have become available; they can even be washed and will wear long enough for one laboratory semester.</p>

## KEY STEPS

## IMPORTANT POINTS

4. Wash your hands and the laboratory desk with a 2% Lysol or Cresol solution, or at least with soap and water, at the beginning and the end of each laboratory period. For some students with sensitive skin, the use of a more expensive, but milder, disinfectant may be recommended for hand washing.

5. At the end of each laboratory period check and arrange neatly all equipment assigned to you.

6. Moisten the labels and pencils with water from the tap, kept in some convenient container on the working table—but *never* with your tongue.

7. Keep your hands away from the region of your face and head during a laboratory period.

8. Keep your hair under control.

9. Avoid setting things on fire.

10. Report all accidents to the instructor. (This includes minor cuts, abrasions, pricks and burns, as well as major injuries.)

4.1. This removes dust, prevents unnecessary contamination and helps keep the laboratory tidy.

4.2. pHisoDerm, a milder disinfectant, has replaced pHisoHex, often used for washing hands.

5.1. In order to assure that you will have your allocated equipment, it is important to replace missing and broken materials at the end of each laboratory period.

6.1. Placing any articles, or even your fingers, in your mouth is a method of transfer of microorganisms.

6.2. Your laboratory instructor can help you to break the bad and dangerous habit of putting your pencil into your mouth by saying "fomite" every time you do it.

7.1. Your hands may be soiled, stained or contaminated.

8.1. Long, uncontrolled hair and non-fireproof clothing are fire hazards in the laboratory where Bunsen burners will be lit. When you smell burning hair, it usually is in contact with or too close to the open flame.

9.1. Fires will occur in every microbiology laboratory. It's usually cotton that burns. Stay cool; don't blow at the fire. Putting your hand over the cotton will help, or step on it—smother the fire.

9.2. Most important, the laboratory instructor will demonstrate use of the fire extinguisher in your laboratory to you.

10.1. Minor injuries can be treated adequately with first-aid measures. If these injuries are untreated, they may result in much more serious conditions.

10.2. Burns are best treated with ice and cold water, applied immediately. Sodium bicarbonate (baking soda) liberally applied to the burn will reduce the pain.



11. Leave all teaching materials in the laboratory.

12. Discard all solid waste material in containers provided for this purpose.

13. Check with your instructor about disposal of unused sterile materials and the return of special equipment.

14. Before leaving the laboratory, wash your hands thoroughly with 2% Lysol solution, Cresol, pHisoDerm, other mild disinfectants, or at least soap and running water.

10.3. More serious injuries should be referred immediately to the medical center.

11.1. There is no excuse for storing cultures and specimens in your room.

11.2. Cultures, specimens and illustrative material need to be shared with other students in the class.

12.1. Materials like paper, cotton and media will clog the plumbing if these substances are put in the sink.

13.1. These materials may be needed for other students.

14.1. Hands may be a method of transferring microorganisms from the laboratory to your nose and mouth and to others.