EIGHTEENTH EDITION

Zinsser Microbiology

Joklik, Willett, Amos

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Preface

With each passing year the term *Microbiology* becomes a less satisfactory umbrella for the many disciplines that it attempts to cover. Bacteriology, immunology, virology, mycology, and parasitology have each long since become separate and independent disciplines. They are together in a single text simply because they deal with the agents of infectious disease in humans and with the mechanisms employed by the host in his defense.

In spite of the undeniable triumphs of antibiotic chemotherapy, which has revolutionized the practice of medicine and very likely represents the greatest single triumph of biomedical science, "microbes" are by no means "conquered"; they continue to cause infections that demand a large amount of the physician's time. In fact, new infectious agents, unsuspected properties of known agents, additional mechanisms for the genesis and persistence of infections, and advances in our understanding of the behavior of infectious agents at the molecular, cellular, and organismal levels are constantly being reported. As a result, the scope and complexity of the material to be presented to students expand rapidly, and although the literature abounds with excellent papers and reviews, the compilation of a comprehensive textbook of manageable size becomes increasingly difficult.

This new edition of *Zinsser Microbiology*, the 18th, is designed for use by medical students experiencing their first exposure to medical microbiology. To that end, there is presented not only a description of the pathogenic infectious agents and the diseases that they cause, but also a discussion of the basic principles of bacterial physiology and genetics, of molecular and cellular immunology, and of molecular virology, the purpose of which is to provide a firm basis for growth with the field during the remainder of the student's future professional career. By the same token, the book will also fulfill the needs of advanced undergraduates who plan careers in medicine or biomedical research. The book is also designed as a reference source for instructors; to that end each chapter is supplemented with a selection of both reviews and important original papers that will permit a rapid entrée to any specialized topic that may require further study.

The 18th Edition represents a very extensive revision of the 17th Edition; all portions of the text that were not completely rewritten have been thoroughly updated. Section VII, Medical Parasitology, has been completely rewritten by a new contributor, Dr. Norman F. Weatherly; he replaces Dr. John E. Larsh, Jr., who contributed this section to the last six editions and who has now retired. Dr. Weatherly brings fresh approaches and new viewpoints without sacrificing authority. The Clinical Virology section also has several new contributors, who contributed completely new chapters on herpesviruses, papovaviruses, viruses in gastrointestinal tract infections, and hepatitis viruses. In the Basic Virology and Bacterial Physiology sections many chapters, particularly those on the molecular aspects of virus multiplication cycles, on tumor viruses, and on the molecular basis of genetics have been completely rewritten; these are areas in which exciting discoveries are constantly being made, many of them changing fundamentally our views of the arrangement of genetic material and how it is expressed. The same applies to the Immunology section where new chapters on immunopathology, immune responses to infection, and immunity to tumors and to pregnancy have been provided. This section provides a comprehensive account of both basic and clinical immunology, organized so as to highlight topics currently deemed of maximum relevance to medical students. Finally, the Medical Bacteriology and Medical Mycology sections have been brought up-to-date, with new material added on new diseases such as Legionnaire's disease, the toxic shock syndrome, the acquired immune deficiency syndrome (AIDS), and Lyme disease. In these sections, which like all other sections have been carefully edited by a single author so as to ensure a uniform format, emphasis is again placed on correlating the basic and clinical aspects of each infectious agent so that the student may acquire an appreciation of how fundamental research may be used in unravelling the complexities of host–parasite relationships. Each chapter consists of (1) an introduction to the important biologic properties of the organism, (2) a description of the clinical infection in humans, including a discussion of mechanisms of pathogenicity, (3) a section on laboratory diagnosis that provides information on modern culture and immunological procedures, and (4) a discussion of the currently recommended treatment.

With regard to the bibliography, we have again elected not to reference specific statements in the text but to append to each chapter a list of recent reviews and key

original papers. The former will quickly guide the reader to any specific aspect of microbiology and immunology that he wishes to pursue; the latter makes available the detailed considerations and circumstances that have gone into the genesis of the most important discoveries. Many of the papers that are cited already are, or no doubt will soon become, "classics."

We have tried not to increase the size of the book—no easy task in view of the enormous amount of new information that has accumulated since publication of the last edition in 1980. Obviously, this has entailed the omission of a certain amount of older material; however, we are confident that there are no major gaps and that in our presentation of the newest advances we have not sacrificed careful and logical explanations of fundamental principles.

The list of individuals who have helped to produce this volume extends far beyond the circle of our colleagues who contributed textual material and to whom we are profoundly indebted. We would especially like to thank our many colleagues who permitted us to use illustrative material and who almost invariably supplied us with original photographs, and the many publishers who allowed us to reproduce previously published material. We would also like to thank the artists who did a superb job in drawing the innumerable charts and diagrams, and the many secretaries who cheerfully massaged the text innumerable times on their word processors. Finally, we wish to express our appreciation to the staff of Appleton-Century-Crofts for their efficient cooperation in producing this new edition.

Wolfgang K. Joklik
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Preface to the First Edition

The volume here presented is primarily a treatise on the fundamental laws and technic of bacteriology, as illustrated by their application to the study of pathogenic bacteria.

So ubiquitous are the bacteria and so manifold their activities that bacteriology, although one of the youngest of sciences, has already been divided into special fields—medical, sanitary, agricultural, and industrial—having little in common, except problems of general bacterial physiology and certain fundamental technical procedures.

From no other point of approach, however, is such a breadth of conception attainable, as through the study of bacteria in their relation to disease processes in man and animals. Through such a study one must become familiar not only with the growth characteristics and products of the bacteria apart from the animal body, thus gaining a knowledge of methods and procedures common to the study of pathogenic and nonpathogenic organisms, but also with those complicated reactions taking place between the bacteria and their products on the one hand and the cells and fluids of the animal body on the other—reactions which often manifest themselves as symptoms and lesions of disease or by visible changes in the test tube.

Through a study and comprehension of the processes underlying these reactions, our knowledge of cell physiology has been broadened, and facts of inestimable value have been discovered, which have thrown light upon some of the most obscure problems of infection and immunity and have led to hitherto unsuspected methods of treatment and diagnosis. Thus, through medical bacteriology—that highly specialized offshoot of general biology and pathology—have been given back to the parent sciences and to medicine in general methods and knowledge of the widest application.

It has been our endeavor, therefore, to present this phase of our subject in as broad and critical a manner as possible in the sections dealing with infection and immunity and with methods of biological diagnosis and treatment of disease, so that the student and practitioner of medicine, by becoming familiar with underlying laws and principles, may not only be in a position to realize the meaning and scope of some of these newer discoveries and methods, but may be in a better position to decide for themselves their proper application and limitation.

We have not hesitated, whenever necessary for a proper understanding of processes of bacterial nutrition or physiology, or for breadth of view in considering problems of the relation of bacteria to our food supply and environment, to make free use of illustrations from the more special fields of agricultural and sanitary bacteriology, and some special methods of the bacteriology of sanitation are given in the last division of the book, dealing with the bacteria in relation to our food and environment.

In conclusion it may be said that the scope and arrangement of subjects treated in this book are the direct outcome of many years of experience in the instruction of students in medical and in advanced university courses in bacteriology, and that it is our hope that this volume may not only meet the needs of such students but may prove of value to the practitioner of medicine for whom it has also been written.

It is a pleasure to acknowledge the courtesy of those who furnished us with illustrations for use in the text, and our indebtedness to Dr. Gardner Hopkins and Professor Francis Carter Wood for a number of the photomicrographs taken especially for this work.

P. H. Hiss, Jr.
H. Zinsser

Contents

SECTION I

BACTERIAL PHYSIOLOGY

CHAPTER 1

The Historical Development of Medical Microbiology

The history of the many concepts now embodied in the doctrines of microbiology is an account of attempts to solve the problems of the origin of life, the putrefaction of dead organic materials, and the nature of communicable changes in the bodies of living humans and animals. The visible aspects of these phenomena were as apparent and interesting to ancient observers as they are to modern biologists. In the past, notions of ultimate causes were derived from the available factual knowledge colored by the theologic and philosophic tenets of the time. The early history of what has become the science of microbiology is to be found, therefore, in the writings of the priests, philosophers, and scientists who studied and pondered these basic biologic problems.

Infection and Contagion

Among ancient peoples, epidemic and even endemic diseases were regarded as supernatural in origin and sent by the gods as punishment for the sins of man. The treatment and, more important, the prevention of these diseases were sought by sacrifices and lustrations to appease the anger of the gods. Since man is willful, wanton, and sinful by nature, there was never any difficulty in finding a particular set of sins to justify a specific epidemic.

The concept of contagion and the practice of hygiene were, however, not entirely unknown to ancient man. The Old Testament is often quoted as indicating the belief that leprosy was contagious and could be transmitted by contact. The principle of contagion by invisible creatures was later recorded by Varro in the second century BC. and the concept was familiar to Greek, Roman, and Arabic writers. Roger Bacon, in the thirteenth century, more than a millennium later, postulated that invisible living creatures produced disease. The Venetian, Fracastorius, in 1546, wrote from a knowledge of syphilis that communicable disease was transmitted by living germs, "seminaria morbi," through direct contact or by intermediary inanimate fomites and through air "ad distans." Fracastorius expressed the opinion that the seeds of disease, passing from one infected individual to another, caused the same disease in the recipient as in the donor. This clear expression of the germ theory of disease was three centuries ahead of its time.

First Observation of Bacteria

Direct observation of microorganisms had to await the development of the microscope. The human eye cannot see objects smaller than 30 μm ($^1/_{1000}$ inch) in diameter, and

although knowledge of magnifying lenses reaches back to the time of Archimedes, the science of optics was not clarified until the thirteenth century by the Franciscan monk, Roger Bacon. The telescope was invented by Galileo in 1608, followed by the microscope in the same century. The first person known to have made glass lenses powerful enough to observe and describe bacteria was the amateur lens grinder, Anton van Leeuwenhoek (1632–1723), of Delft, Holland. In letters to the experimentalist group, The Royal Society of London, Leeuwenhoek described many "animalcules," including the three major morphologic forms of bacteria (rod, sphere, and spiral), various free-living and parasitic protozoa from human and animal feces, filamentous fungi, and globular bodies we now know as yeasts, and he discovered spermatozoa. In his lifetime he made some 250 single-lensed microscopes. He searched everywhere in this new microcosmic world he had discovered. In letters from 1676 to 1683, he described the sizes, shapes, and even the motility of bacteria (Fig. 1-1), using simple single biconvex lens microscopes, such as illustrated in Figure 1-2. There is no doubt that he saw the most common forms of bacteria, the cocci, bacilli, and spirochetes. His observational reports were enthusiastic and accurate and developed some interest at the time, but unfortunately Leeuwenhoek did all this as a hobby and left no students to continue his work. However, in 1678, Robert Hooke, who developed the compound microscope, confirmed Leeuwenhoek's discoveries. Microorganisms were then occasionally studied by those primarily interested in classifying the various life forms observable with the microscope. These observations lay dormant and were not exploited by those interested in disease. The following 125 to 150 years witnessed the gradual development of knowledge and acceptance of the experimental method, which slowly disseminated throughout the expanding learned centers of the world. Improved microscopes became generally available only in the 1800s as a result of the Industrial Revolution, which allowed rapid technologic advances. Even then, no notable advance in microbiology was accomplished until after the attention of the scientific world was focused on microbes and their role in the controversies of the doctrine of spontaneous generation and the associated phenomenon of fermentation.

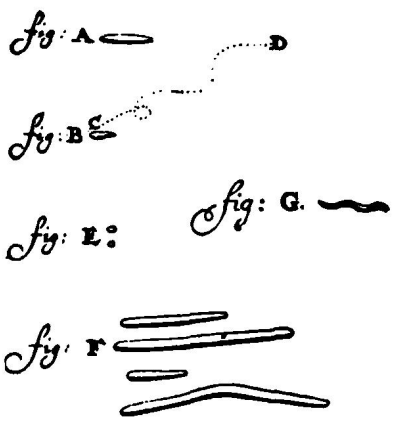

Figure 1-1. Leeuwenhoek's picture of bacteria from the human mouth. Dobell's identifications are as follows: **A.** A motile *Bacillus.* **B.** *Selenomonas sputigena.* **E.** Micrococci. **F.** *Leptothrix buccans.* **G.** Probably *Spirochaeta buccalis. (From Dobell: Anton van Leeuwenhoek and His "Little Animals," 1932. New York, Harcourt, Brace and Co.)*

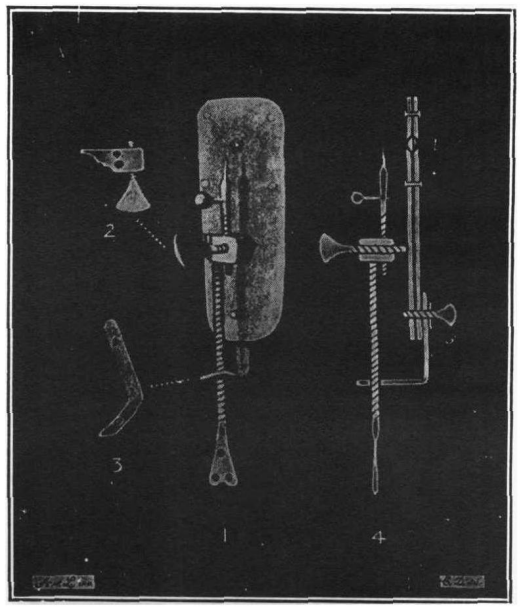

Figure 1-2. Leeuwenhoek's "Microscope." *(From Dobell: Anton van Leeuwenhoek and His "Little Animals," 1932. New York, Harcourt, Brace and Co.)*

Spontaneous Generation

The controversy over human ability to create life carried over from Greek mythology. Even Aristotle (384–322 BC) thought animals could originate from the soil. Samson, in the Bible (Old Testament), and again Virgil, about 40 BC, described recipes for producing bees from honey, and for centuries it was believed that maggots could be produced by exposing meat to warmth in the air. This was not refuted until Francesco Redi (1626–1697) proved that gauze placed over a jar containing meat prevented maggots forming in the meat. Redi also observed that adult flies, attracted by the odor of meat through the gauze, laid eggs on the cloth, and maggots developed from the eggs. Recipes for producing mice and other similar life forms in litter and refuse were gradually disproved and discarded in similar fashion. However, the question was not settled in all minds. When microbes were discovered, their association with putrefaction and fermentations again raised the question of spontaneous generation. John Needham, in 1749, observed the appearance of microorganisms in putrefying meat and interpreted this as spontaneous generation. Spallanzani, however, boiled beef broth for an hour, sealed the flasks, and observed no formation of microbes. Needham and, 100 years later, Pouchet (1859) argued that

access of air was necessary for the spontaneous generation of microscopic living beings. Disproof came from several lines of evidence. Franz Schulze (1815–1873) passed air through strong acids and then into boiled broth, while Theodor Schwann (1810–1882) passed air through red hot tubes and observed no growth. About 1850, Schroeder and von Dusch filtered air through cotton filters into broth and observed no growth. Pasteur was able to filter microorganisms from the air and concluded that this was the source of contamination. He developed an aseptic technique, using heat, in order to tranfer and work with his microbes and, finally, in 1859, in public controversy with Pouchet, prepared boiled broths in flasks with long narrow gooseneck tubes that were open to the air. Air could pass but microorganisms settled in the gooseneck, and no growth developed in any of the flasks. Finally, a British physicist, John Tyndall (1820–1893), proved that dust carried the germs, and the story was complete. Tyndall also found that bacterial spores could be killed by successive heating, a process now known as tyndallization.

The Germ Theory of Disease

Empirical Observations. A firm basis for the causal nature of infectious disease was established only in the latter half of the nineteenth century. One of the first proofs came from Agostino Bassi who, in the early 1800s, proved that a fungus, later named *Botrytis bassiana* in his honor, caused a disease of silkworms called "muscardine" in France and "mal segno" in Italy. In 1839, Schoenlein found the causative fungus in lesions of favus, and, in 1846, Eichstedt noted the contagiousness of pityriasis versicolor and discovered a fungus in skin scrapings from patients.

In the 1840s, the American poet-physician, Oliver Wendell Holmes, wrote "The Contagiousness of Puerperal Fever," in which he suggested that disease was caused by germs carried from one new mother to another. In 1861, Ignaz Semmelweis, who had drastically decreased childbirth deaths by antiseptic techniques and practices, published his extremely important "The Cause, Concept and Prophylaxis of Childbed Fever." However, the importance of antiseptics in reducing contagious disease was not fully realized until the late 1870s, when Joseph Lister demonstrated the value of spraying operating rooms with aqueous phenol.

Lessons Learned from Fermentations. Further emphasis on microbial activities came from the work of Louis Pasteur from the 1850s to the 1880s. In studies on the diseases of wine, Pasteur demonstrated that alcoholic fermentation of grapes, fruit, and grains was caused by microbes, then called "ferments." In good wine batches certain types of "ferments" existed in the vats, while in poor or bad fermentations other types of microbes were found, some of which Pasteur found to be capable of growing anaerobically. He suggested eliminating the bad types of "ferments" from fresh juices by heating at 63C for one-half hour, then cooling and reinoculating with a culture from the satisfactory vats. Pasteur's success with the problems of the wine industry led the French government to request that he study a disease, pébrine, which was ruining the silkworm industry in Southern France. Pasteur struggled with this problem for several years before he isolated the causative organism and showed that farmers could eliminate the problem by using healthy, noninfected breeding stock.

Observations and Experiments with Animals. In 1850, Rayer and Davaine observed rod-shaped microorganisms in the blood of animals that had died of anthrax. Rayer recalled the experiments in 1825 of Barthélemy who had shown that anthrax was transmissible by inoculation in series in sheep, and by 1863, Davaine had experimentally transmitted anthrax by blood containing these rods but not by normal blood from which rods were absent. In 1872, Obermeier discovered the relationship of a spirillum to relapsing fever and demonstrated for the first time the presence of a pathogenic microorganism in the blood of a human being.

Importance of Pure Culture Techniques. Through all this time, etiologic research was not based on pure culture work. Pure cultures were obtained largely by accident, and investigators had no way, except by crude morphologic microscopic examination, of knowing when contaminants were present. This resulted in much equivocal thinking and work that hindered progress.

The first pure or axenic culture technique was developed by Joseph Lister in 1878. Lister used a syringe to make serial dilutions in liquid media to obtain pure cultures of a simple type of organism, which he named *Bacterium (Lactobacillus) lactis.* Meanwhile, Koch, as a student of Henle who insisted on proof that an organism caused disease, was also developing and refining techniques for the isolation of pure cultures. From the work of others, notably Ehrlich, Koch learned methods of staining bacteria on glass with aniline dyes for microscopic observation. In his early work on anthrax, Koch used sterile aqueous humor of the eyes of animals as a growth medium. But, having seen the advantages of older, solid but opaque media, such as potato, beets, starch, bread, egg white, and meat, Koch developed a transparent solid medium by mixing gelatin with Löffler's peptone solution. The gelatin mixture liquefied on warming, could be heat-sterilized and aseptically poured into plates, and upon cooling, it solidified. Microorganisms streaked upon it developed into macroscopic colonies as the result of the growth of a single invisible cell. However, gelatin liquefies at a relatively low temperature (26C), and Koch later switched to agar, the transparent red seaweed extract that solidifies below 43C.

Etiologic Proof of Infectious Agents. Koch was able to isolate the anthrax organism in pure culture by streaking on his solid media and found that even after many transfers, the organism could still cause the same symptoms and disease when inoculated into animals. On the ba-

sis of his experiences, Koch formulated criteria that provided proof that a specific bacterium caused a disease. We now call these Koch's postulates.

1. The organism must always be found in the diseased animal but not in healthy ones.
2. The organism must be isolated from diseased animals and grown in pure culture away from the animal.
3. The organism isolated in pure culture must initiate and reproduce the disease when reinoculated into susceptible animals.
4. The organism should be reisolated from the experimentally infected animals.

Koch's work thus provided impetus and means for proof of the germ theory of disease.

The 20-year period following Koch's work was the Golden Age of Bacteriology. By 1900, almost all the major bacterial disease organisms had been described. The list included anthrax (*Bacillus anthracis*), diphtheria (*Corynebacterium diphtheriae*), typhoid fever (*Salmonella typhi*), gonorrhea (*Neisseria gonorrhoeae*), gas gangrene (*Clostridium perfringens*), tetanus or lockjaw (*Clostridium tetani*), dysentery (*Shigella dysenteriae*), syphilis (*Treponema pallidum*), and others.

Viruses

Only with advances in technique and improvement in apparatus is it possible to make fundamental advances through new ideas and observations. The development of bacteriologic filters and the discovery of viruses is a case in point.

Bacteriologic Filters. As an alternate to heat sterilization, unsuccessful efforts to remove bacteria from solutions by filtration through paper and similar materials led Chamberland and Pasteur to test and develop unglazed porcelain as the first successful bacterial filter (1871–1884). The Berkefeld filter of Kieselguhr (diatomaceous earth) was developed shortly thereafter in 1891. Synthetic polymer filters of cellulose nitrate, cellulose acetate, polyester, and so forth have come into common use only in the last two or three decades because of technical advances allowing quality control of pore size. It is of interest to note that these are essentially space-age products developed in part for the rapid removal of microorganisms from jet and rocket fuels.

Discovery of Viruses. The tobacco mosaic disease agent was discovered by Iwanowski in 1892 in bacteria-free filtrates of diseased plant leaf juices. This finding, confirmed by Beijerinck in 1899, marked the beginning of studies on the so-called filterable agents.

The filterable agent causing foot-and-mouth disease in cattle, the first described animal virus, was discovered by Löffler and Frosch in 1898. The yellow fever virus of humans was discovered in 1900 by Walter Reed and his coworkers. Bacterial viruses, or bacteriophages, were discovered in 1915 by Twort in England and d'Herelle in France.

Viruses could not be grown in artificial media, and Koch's criteria could not be specifically applied. Because these pathogens require a living host for propagation, rapid progress in their study developed only in recent years. Again, as in the Golden Age of Bacteriology, technology had to be developed. Outstanding were the development of the electron microscope, of the ultracentrifuge, and of tissue culture, and the application of sophisticated microchemical and biochemical techniques.

It is of interest to note that some filterable agents first thought to be viruses, such as the *Mycoplasma*, *Rickettsia*, and *Chlamydia*, all of which are extremely fastidious in their growth requirements and almost all of which require living host cells, were subsequently shown to be bacteria.

Immunity

Ancient peoples immunized themselves against venomous snakes by introducing small amounts of venom into scratches in the skin. The Chinese used variolization with dried material from dermic smallpox lesions for 20 centuries. This practice spread through Asia by trade routes and was well accepted in the Middle East. Later, Edward Jenner (1749–1823) noticed that milkmaids who developed cowpox were immune to smallpox and found that he was able to protect susceptible individuals by vaccinating them with cowpox. Pasteur developed a chicken cholera vaccine in 1877; he inoculated chickens with old attenuated cultures so that a mild disease rendered the chickens immune to virulent organisms. He called this "vaccination," after Jenner's procedure. Shortly afterward, in 1881, applying the same concept, Pasteur prepared temperature-attenuated anthrax grown at 42 to 43C and protected sheep by first injecting them with these bacteria before challenging them with virulent anthrax grown at lower temperatures. Salmon and Smith, in 1884–1886, used heat-killed cultures of hog cholera bacillus to develop resistance or immunity in swine against challenge by live virulent organisms. Pasteur developed rabies vaccine in 1886, again using the idea of injecting an attenuated living disease agent. In this case, Pasteur used dried animal spinal cords without, apparently, recognizing the viral form of the disease agent.

Two schools of thought arose in explanation of the increased resistance following vaccination. Metchnikoff developed, in the 1880s, the cellular theory of protection; Bordet and others proposed the humoral, or specific, antibody concept of immunity. There is now evidence that both theories are correct. The last two decades have resulted in the isolation and, in large measure, the structural description of the major humoral immune proteins, the immunoglobulins. These are now commonly referred to as IgA, IgG, IgD, IgM, and IgE. The functions of these various immunoglobulins are currently being intensively studied. Much work is also being devoted to the mechanisms of cellular interactions in immune reactions that occur not only in infectious disease caused by bacteria, viruses, fun-

gi, and parasites but also in rejection reactions of tissue and organ transplants, and of cancer cells.

Antimetabolites

Many antimetabolites, which were pioneered in concept by Ehrlich in the mid to late 1800s, are now accepted household words, e.g., penicillin. The modern era of antibiotics developed only after Domagk reported in 1935 that Prontosil had a dramatic effect on streptococcal infections. It was soon discovered that Prontosil was converted in the body to sulfanilamide, the active chemical agent, which is an analog of the vitamin *p*-aminobenzoic acid. In the 1940s, as the result of the stimulus of World War II, Florey and Chain and their associates reinvestigated Fleming's penicillin, isolated and characterized it, and demonstrated its practical clinical value. As a result of millions of tests with thousands of organisms, we now have numerous other antibiotics active against almost all types of bacteria.

With the recognition of the metabolic and structural differences, at the molecular level, between pathogenic microorganisms and human or animal cells, the rationale for developing new chemotherapeutic compounds is now often based on exploiting these differences. There is every reason to believe that newer, more specific and potent drugs will be discovered. However, chemotherapy has created new problems. Many previously susceptible organisms are now resistant to therapeutic levels of many widely used drugs. In addition, drug sensitization reactions or allergies occasionally develop, clinical syndromes are modified, and the normal ecologic flora of the body is disturbed.

Impact of Microbiology on Genetics and Biochemistry, and the Development of Molecular Biology

The enormous advantages of the availability of homogeneous populations of cells for every conceivable type of investigation were soon realized. As a result, many of the epoch-making advances during the last century in cell physiology, biochemistry, and genetics have resulted from studies with microorganisms or materials isolated from them. During the last two or three decades, these advances have led to a precise way of investigating the structure and function of nucleic acids and proteins, which has become known as "molecular biology." For example, the demonstration of the central role of DNA as the repository of genetic information resulted from the studies of Griffith in the 1920s that pneumococci could be transformed from one capsular type to another, followed by the demonstration by Avery and associates during the 1940s that the transforming factor was DNA. Final proof beyond doubt was provided by the demonstration of Hershey and Chase in 1952 that viral nucleic acid itself contained all the information necessary for virus multiplication. At the same time, Watson and Crick developed the double-helix model of DNA, which led them to suggest that one of the complementary DNA strands could serve as the template for the synthesis of the other, thus providing a description of self-perpetuating gene replication and continuity.

Demonstration of the transcription from DNA of information in the form of messenger RNA synthesized in complementary sequence to DNA soon followed, again in a microbial system. Messenger RNA was then found to be translated into polypeptides on ribosomes. By the early 1960s Nirenberg, Ochoa, and others had worked out the nature of the triplet RNA base sequences corresponding to the codon signals for each amino acid.

More recently, attention has focused on the arrangement of genetic material, including the nature of genes, and on the mechanisms that control its expression. While much of this research work is still being carried out with microbial systems, cells of higher organisms, including mammalian cells, are also being used very extensively nowadays. A very important factor in this connection has been the development of the technique of tissue culture, which permits animal cells to be grown, cloned, and passaged like microorganisms. New concepts concerning the regulation of gene expression in mammalian cells are being developed rapidly. As a result, it should be possible to develop, within the foreseeable future, a rational system of antiviral chemotherapy, so that virus-caused diseases may be brought under effective control, just as antibiotics control bacteria-caused diseases. Further, there is every hope that the fundamental control mechanisms that operate in both normal and abnormal cell differentiation, including cancer, will become apparent before too long.

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