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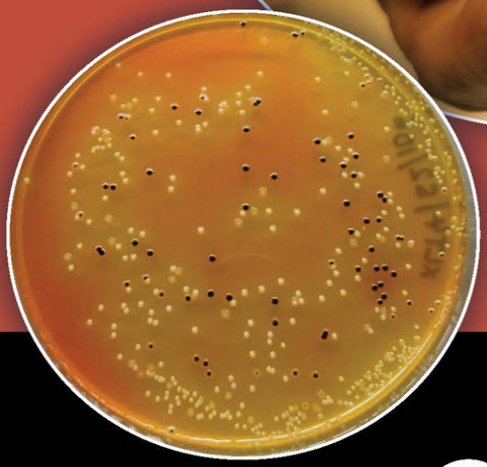
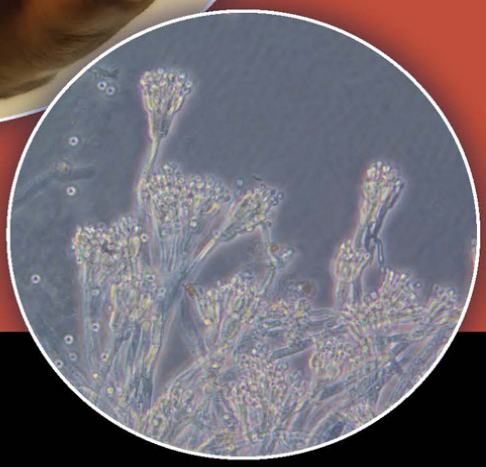
Fundamental Food Microbiology

Bibek Ray and Arun Bhunia





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Fundamental Food Microbiology

Food microbiology has seen enormous growth in the last decade, fueled by the global pandemic of COVID-19 and continual routine outbreaks with traditional foodborne pathogens. In addition, climate change and global warming also affect agriculture and food production, in turn shifting microbial ecology. Such changes will affect pathogen behavior, spoilage, and microbial growth, impacting food safety and quality. Health-conscious consumers are also looking for foods with alternative protein sources from plants and insects, such as fermented, antioxidant, and micronutrient-rich superfoods. All three areas of food microbiology—beneficial, spoilage, and pathogenic microbiology—are expanding and progressing incredibly. What was once a simple process of counting colonies has become a sophisticated process of sequencing complete genomes, gene editing, and biotechnology for starter cultures and probiotics improvement and application of sophisticated analytical tools for microbial analysis.

Fundamental Food Microbiology, Sixth Edition captures these developments and broadens coverage of foodborne disease mechanisms, spoilage microbes, and microbial inactivation strategies. Written by experts with approximately 60 years of combined experience, the book provides an in-depth understanding of how to reduce microbial food spoilage, improve intervention technologies, and develop effective control methods for different types of foods.

See What's New in the Sixth Edition:

- Condensed chapter descriptions with illustrations
- CRISPR/Cas system for gene editing
- Novel food processing technologies, including plasma and micro/nanobubble technologies
- Food radiation and hurdle concept chapters merged and overhauled
- Comprehensive list of mycotoxins and seafood-related toxins
- Updates on several new antimicrobial compounds from animal and plant sources

Maintaining the high standard set by the previous bestselling editions, and based on feedback from students and professors, this new edition includes even more easy-to-follow figures and illustrations. The chapters are presented logically, connecting the information and allowing students to understand and retain the concepts easily. These features make this a comprehensive introductory text for undergraduates and a valuable reference for graduate-level and working professionals in food microbiology, food safety, or food technology.

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Fundamental Food Microbiology

Sixth Edition

Bibek Ray and Arun Bhunia

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Dedication

Bibek Ray dedicates this book to his parents, Hem and Kiron Ray

Arun Bhunia dedicates this book to his parents,
Nandalal and Bangabala Bhunia,
and his family: Banashri, Arni, and Irene Bhunia

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Preface to the Sixth Edition

In the sixth edition, we aimed to capture scientific advancements related to foodborne pathogens, the impact of climate change on foodborne diseases, food spoilage, and sustainability. We reorganized several chapters and merged the appendix sections for better presentation and clarity. New topics included in this edition are a brief description of the CRISPR/Cas system for gene editing, plasma technology, micro/nanobubble technology for food processing, plant- and animal-derived antimicrobial peptides, bioengineering of probiotics, and some key mycotoxins. All chapters in this sixth edition have been thoroughly revised and updated with recent reference articles, illustrations, and figures when possible.

I am indebted to my wife, Banashri Bhunia, my son, Arni, and my daughter, Irene, for their inspiration and patience while I worked late hours, weekends, and holidays for several months on this book. I also want to thank my colleagues, Drs. Un-Joong Oh and Carlos Corvalan, for providing updated information on the content. I want to thank my postdoc, Atul Singh, and graduate students, especially Nicholas Gallina, for generating photographs of microorganisms, and other past and present postdocs and students for their continued support.

I sincerely thank Prof. Bibek Ray for trusting me to continue this project and update the content with the most recent developments in food microbiology. We thank CRC Press, especially Mr. Stephen M. Zollo, Senior Editor, for his continued support and leadership. We sincerely hope that the sixth edition will continue to generate interest as a trusted source of knowledge and information for students and professionals around the globe.

Bibek Ray
Arun Bhunia
June 2024

Authors

Bibek Ray, PhD, currently Professor Emeritus, was a professor of food microbiology in the Department of Animal Science at the University of Wyoming, Laramie. Prof. Ray earned BS and MS degrees in veterinary science from the University of Calcutta and the University of Madras, in India, respectively. He received his PhD in food science from the University of Minnesota in 1970 and joined the faculty in the Department of Food Science, North Carolina State University, and then the Department of Biology at Shaw University, both at Raleigh, North Carolina. He joined the University of Wyoming in 1981. There, he expanded his research to intestinal beneficial bacteria, bacteriocins of Gram-positive bacteria, and high hydrostatic pressure preservation of food along with his previous research activities in the area of microbial sublethal injury. He also taught courses in food microbiology, food fermentation, food safety, and a course titled, "Safety of Our Food" to nonscience undergraduates. His laboratory was involved in extensive studies in both basic and applied areas of the bacteriocin pediocin AcH from *Pediococcus acidilactici* H. In addition, his group studied various aspects of bacteriocins produced by *Lactococcus*, *Leuconostoc*, *Lactobacillus*, *Pediococcus*, and *Bacillus* and *Staphylococcus* spp. He received research funding from the National Science Foundation, American Public Health Association, National Live Stock and Meat Board, United States Department of Agriculture, United States Army Research, North Atlantic Treaty Organization (with Turkey) and Binational Agriculture Research Development Agency (with Israel), Wyoming Development Fund, and the industry. Before retirement, he was studying the combined effects of bacteriocins, ultrahigh hydrostatic pressure, and pulse field electricity and sublethal injury on the destruction of microbial cells and spores and its applications in food preservation. In addition, Dr. Ray established collaborative research programs with research institutes and universities in Turkey, Israel, India, Indonesia, and France. He also participated in symposiums, offered short courses, presented research activities, and visited research laboratories in Japan, Indonesia, Thailand, Australia, New Zealand, India, Turkey, France, Spain, Germany, Norway, Belgium, England, Denmark, Netherlands, Hungary, Austria, Greece, Italy, Mexico, Canada, and Argentina.

Prof. Ray has published over 100 research articles, reviews, book chapters, proceedings articles, and popular articles on food microbiology. He has also edited four books: *Injured Index and Pathogenic Bacteria* (1989) and *Food Biopreservatives of Microbial Origin* (1992, with Dr. M.A. Daeschel), both published by CRC Press, Boca Raton, FL; *Lactic Acid Bacteria: Current Advances in Metabolism, Genetics, and Applications* (1996, with Dr. Faruk Bozoglu); and *Novel Processing and Control Technologies in the Food Industry* (1999, with Dr. Faruk Bozoglu and Tibor Deak). He was a member of the American Society for Microbiology and the Institute of Food Technologists and a fellow of the American Academy of Microbiology. He also served on the editorial boards of the *Journal of Food Protection*, *Applied and Environmental Microbiology*, and the *Indian Journal of Microbiology.* In 1994, Prof. Ray was awarded the University of Wyoming Presidential Achievement Award in recognition of his excellence in academic performance. He retired from the University of Wyoming in 2002. Then he, with his wife, Purnima, a molecular biologist, moved to Dr. Ray's village of birth and started a nonprofit organization (Gohaldanga Ramkrishna Sarada Vivekananda Sevakendra) to improve the education, health, and economy of the underprivileged villagers. Since April 2024, the organization has been fully accepted by the Ramkrishna Mission Ashram Narendrapur: Gohaldanga Campus (a branch of the renowned Ramkrishna Mission).

Arun Bhunia, BVSc, PhD, is a professor of molecular food microbiology in the Department of Food Science and the Department of Veterinary Comparative Pathobiology (by courtesy) at Purdue University, West Lafayette, Indiana. He is also affiliated with the Purdue Institute of Immunology, Inflammation and Infectious Disease and the Purdue University Interdisciplinary Life Sciences program (PULSe).

Prof. Bhunia received his Bachelor of Veterinary Medicine degree (1984) from Bidhan Chandra Krishi Viswa Vidyalaya (currently West Bengal University of Fisheries and Animal Sciences), West Bengal, India. He received his PhD (1989) from the University of Wyoming under the mentorship of Prof. Bibek Ray and postdoctoral training (1995) from the University of Arkansas, Fayetteville, under the mentorship of Prof. Michael G. Johnson. He then joined Alabama A&M University (Huntsville) as an assistant professor in 1995. In 1998, he joined the Department of Food Science at Purdue University. He teaches four graduate-level courses: Food Microbiology; Microbial Foodborne Pathogens (major topics: mechanism of the pathogenesis of foodborne pathogens and toxins, and host–parasite interactions); Microbial Techniques for Food Pathogens (laboratory course that incorporates rapid methods employing immunoassays, and genetic and biosensor tools), and co-teaches a graduate-level journal club, Intestinal Microbiology and Immunology. In addition, he routinely lectures in courses such as Principles of Public Health (both undergraduate and graduate levels) on foodborne diseases and Veterinary Microbiology and Mycology on zoonotic foodborne pathogens. Prof. Bhunia's laboratory maintains a balanced research program (basic and applied) to address problems related to foodborne pathogens in the following areas: microbial pathogenesis (host–pathogen interaction), immunology, probiotic bioengineering, and foodborne pathogen detection. His research has been funded by grants from the U.S. Department of Agriculture, the National Science Foundation, the National Cattleman's Beef Association, the National Institute of Health, and Feed Company.

Prof. Bhunia has published 226 refereed and 33 proceedings articles and 204 research abstracts, written 23 book chapters, and delivered over 170 invited presentations in national and international meetings or institutions. He has also written a graduate-level textbook (*Microbial Foodborne Pathogens: Mechanisms and Pathogenesis. Second Edition*, 2018) and coedited four books (*Foodborne Pathogens: Microbiology and Molecular Biology*, 2005; *The Science of Homeland Security,* 2006; *High Throughput Screening for Food Safety Assessment: Biosensor Technologies, Hyperspectral Imaging and Practical Applications*, 2015; and *Sensors for Food Safety and Quality*, 2016).

He is a member of the American Society for Microbiology (ASM), the Institute of Food Technologists (IFT), and the International Association for Food Protection (IAFP), and was a past member of the Society for Photo-Optical Engineers and the American Association for the Advancement of Science. Prof. Bhunia was a member of the National Advisory Committee on Microbiological Criteria for Foods (NACMCF: 2013–2017). He serves as the editor-in-chief of *Foods* (2021–present) and an associate editor for *Frontiers in Microbiology* (2019–present), *PLoS One* (2018–present), *Frontiers in Sustainable Food Systems* (2020–present), *BMC Microbiology* (2020–present), *Indian Journal of Microbiology* (2008–2012), and is an editorial board member for the *Journal of Food Protection* (2002–present), *Indian Journal of Microbiology* (2007–present), and *Frontiers in Cellular and Infection Microbiology* (2021–present), and is Advisory Board Member of *Indian Journal of Animal Health* (2024–present).

In 2003, Prof. Bhunia received a prestigious Purdue Agricultural Research Award; in 2005, he was selected as a University Faculty Scholar; his research team received a "2006 Agriculture Team Award" from Purdue University, an R&D Award from IFT (2009), an Outstanding Graduate Educator Award from the Department of Food Science (2010–2011) and College of Agriculture (2013) at Purdue University; Fulbright Specialist (2016–2023); IAFP Maurice Weber Laboratorian Award (2017); and Fellow IAFP-2023 (2023).

PART I

Introduction to Microbes in Foods

Microorganisms are living entities of microscopic size and include bacteria, archaea, viruses, yeasts and molds (together designated as fungi), algae, and protozoa. For a long time, bacteria have been classified as prokaryotes (cells without definite nuclei), and fungi, algae, and protozoa as eukaryotes (cells with nuclei); viruses do not have regular cell structures and are classified separately. In the 1990s, this classification changed and will be briefly mentioned in Chapter 2. Microorganisms are ubiquitous on Earth, including in humans, animals, plants and other living creatures, soil, water, and the atmosphere, and they can multiply everywhere except in the atmosphere. Together, their numbers far exceed all other living cells on this planet. They were the first live cells to inhabit the Earth more than 3 billion years ago and since then have played important roles, many of which are beneficial to other living systems.

Among the microorganisms, some molds, yeasts, bacteria, and viruses have both desirable and undesirable roles in our food. In this section, the importance of microorganisms in food, predominant microorganisms associated with food, sources from which they get into the food, and the microbiological quality of food under normal conditions are presented in the following chapters:

Chapter 1: History and Development of Food Microbiology
Chapter 2: Characteristics of Predominant Microorganisms in Food
Chapter 3: Sources of Microorganisms in Food
Chapter 4: Microbiological Quality of Foods and Its Significance

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CHAPTER 1

History and Development of Food Microbiology

INTRODUCTION

Except for a few sterile foods, all foods harbor one or more types of microorganisms. Some have desirable roles in food, such as producing naturally fermented food, whereas others cause food spoilage and foodborne diseases. To study the role of microorganisms in food and to control them, it is essential to isolate them in pure culture and learn their morphological, physiological, biochemical, and genetic characteristics. Some of the most straightforward techniques today for these studies were developed over the past 300 years; a brief description is included here.

DISCOVERY OF MICROORGANISMS

The discovery of microorganisms[1–3] ran parallel with the invention and improvement of the microscope. Around 1658, Athanasius Kircher observed minute living worms in putrid meat and milk under a microscope. The magnification power of his microscope was so low that he could not have seen bacteria. In 1664, Robert Hooke described the structure of molds. However, Antony van Leeuwenhoek was the first to visualize different microorganisms. He observed bacteria in saliva, rainwater, vinegar, and other materials; sketched the three morphological groups (spheroids or cocci, cylindrical rods or bacilli, and spirals or spirilla); and described some as motile. He called them **animalcules**, and between 1676 and 1683, he reported his observations to the newly formed leading scientific organization, The Royal Society of London, where his observations were read with fascination. Without much advancement in the microscope in the next 100 years, most scientists only confirmed Leeuwenhoek's observations. In the nineteenth century, the Industrial Revolution helped build more modern microscopes. By 1838, Ehrenberg (who introduced the term *bacteria*) had proposed at least 16 species in 4 genera; by 1875, Ferdinand Cohn had developed the preliminary classification system of bacteria. Cohn discovered that some bacteria produce spores. The existence of submicroscopic viruses was recognized in the mid-nineteenth century after the invention of the electron microscope in the 1940s.

WHERE ARE THEY COMING FROM?

Following Leeuwenhoek's discovery, some scientific minds were curious to determine the source of the animalcules.[1–3] Society had just emerged from the Renaissance period, and science,

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known as experimental philosophy, was in its infancy. Many influential followers among the educated and elite classes accepted the theory of spontaneous generation. Since the time of the Greeks, the emergence of maggots from dead bodies and decaying flesh was thought to be due to spontaneous generation. However, in ca. 1665, Redi disproved that theory by showing that the maggots in rotten meat and fish could only appear if flies were allowed to contaminate them. The advocates of the spontaneous generation theory argued that the animalcules could not regenerate themselves (biogenesis), but they were present in different things only through abiogenesis (spontaneous generation). In 1749, Turbevill Needham showed that boiled meat and meat broth, following storage in covered flasks, could contain animalcules within a short time. This was used to prove the appearance of these animalcules by spontaneous generation. Lazzaro Spallanzani (1765) showed that boiling the meat infusion broth in a flask and immediately sealing the flask prevented these microscopic organisms' appearance, disproving Needham's theory.

Later, Antoine Laurent Lavoisier and his coworkers showed the need for oxygen for life. The believers in abiogenesis rejected Spallanzani's observation, suggesting that there was not enough vital force (oxygen) present in the sealed flask for animalcules to appear through spontaneous generation. Later, Schulze (1830, by passing air through acid), Theodore Schwann (1838, by passing air through red-hot tubes), and Schröeder (1854, by passing air through cotton) showed that bacteria failed to appear in boiled meat infusion even in the presence of air. Finally, in 1861, Louis Pasteur demonstrated that, in boiled infusions, bacteria could grow only if the infusions were contaminated with bacteria carried by dust particles in the air.[1,4] His careful and controlled studies proved that bacteria could reproduce (biogenesis) and that life could not originate by spontaneous generation. John Tyndall, in 1870, showed that boiled infusion could be stored in dust-free air in a box without microbial growth.

WHAT ARE THEIR FUNCTIONS?

The involvement of invisible organisms in many human diseases was suspected as early as the thirteenth century by **Roger Bacon**. In the sixteenth century, **Girolamo Fracastoro** of Verona suggested that small creatures transmitted many human diseases from person to person. This was also indicated by **Kircher** in 1658. In 1762, **von Plenciz** of Vienna suggested that different invisible organisms were responsible for various diseases. **Theodore Schwann** (1837) and **Hermann Helmholtz** (1843) proposed that putrefaction and fermentation were connected with the presence of organisms derived from the air. Finally, **Pasteur**, in 1875, showed that microorganisms caused the fermentation of grapes and souring of wine. He also proved that spoilage of meat and milk was associated with the growth of microorganisms. Later, he showed the association of microorganisms with several diseases in humans, cattle, and sheep, and he also developed vaccines against a few human and animal diseases caused by microorganisms, including rabies. In Germany (1880s and 1890s), **Robert Koch** isolated pure cultures of bacteria responsible for anthrax, cholera, and tuberculosis. He also developed the famous "Koch Postulate" to identify a specific bacterium as a causative agent for a disease. Along with his associates, he also developed techniques of agar plating methods to isolate bacteria in pure cultures and to determine microbial numbers in a sample, the Petri dish (by Julius Richard Petri in 1887, Petri in his laboratory),[5] staining methods for improved microscopic observation of bacteria, and the use of steam to sterilize materials to grow bacteria.[1,6]

With time, the importance of microorganisms in human and animal diseases, soil fertility, plant diseases, fermentation, food spoilage and foodborne diseases, and other areas was recognized, and microbiology was developed as a specific discipline. Later, it was divided into several subdisciplines: medical microbiology, industrial microbiology, soil microbiology, plant pathology, and food microbiology.

DEVELOPMENT OF EARLY FOOD MICROBIOLOGY (BEFORE A.D. 1900)

It is logical to comprehend that our early Homo ancestors, the hunters and gatherers, were aware of food spoilage and foodborne diseases. Even without perceiving the causative agents, they used ice and fire to preserve foods and make them safe. Around 8000 B.C., as agriculture and animal husbandry were adopted by the early civilizations, food supply, especially agricultural produce, became abundant during the growing seasons. Food preservation became necessary for a constant supply of food around the year. Between 8000 and 1000 B.C., many food preservation methods, such as drying, cooking, baking, smoking, salting, sugaring (with honey), low-temperature storage (in ice), storage without air (in pits), fermentation (with fruits, grains, and milk), pickling, and spicing, were used, probably mainly to reduce spoilage. However, one cannot be sure whether society had recognized the implications of diseases transmitted through food. In the later periods, however, the scriptural injunctions laid out by many religions suggest that societies recognized an association of diseases with some foods. Some of the regulations, such as not eating meat from a diseased animal or an animal killed by a scavenger or not eating food that appeared unnatural or had been handled by an unclean person, were developed to safeguard the health of citizens against foodborne diseases. Fermentation was used extensively by many societies not only to preserve foods but also as a method to produce various types of desirable foods from milk, meat, fish, eggs, grains, fruits, and vegetables.

Following the discovery of the ubiquitous existence of microorganisms (mainly bacteria and yeasts) by Leeuwenhoek around the 1670s, some individuals started associating the possible role of these organisms with food spoilage, food fermentation, and foodborne diseases. The major developments of ideas on the possible roles of microorganisms in foods and their scientific proof were initiated by Pasteur in the 1870s, followed by many other scientists before the end of the nineteenth century. This paved the way for establishing early food microbiology in the twentieth century. Some of the major developments in the nineteenth century are briefly listed here.[1,7,8]

Food fermentation

1822	C.J. Person named the microscopic organism found on the surface of wine during vinegar production *Mycoderma mesentericum*. Pasteur, in 1868, proved that this organism was associated with converting alcohol to acetic acid and named it *Mycoderma aceti*. In 1898, Martinus Beijerinck renamed it *Acetobacter aceti*.
1837	Theodor Schwann named the organism involved in sugar fermentation *Saccharomyces* (sugar fungus).
1838	Charles Cogniard-Latour suggested that the growth of yeasts was associated with alcohol fermentation.
1860	Louis Pasteur showed that the fermentation of lactic acid and alcohol from sugar resulted from the growth of specific bacteria and yeasts, respectively.
1883	Emil Christian Hansen used pure cultures of yeasts to ferment beer.

Food spoilage

1804	Francois Nicolas Appert developed methods to preserve foods in sealed glass bottles by heat in boiling water. He credited this process to Lazzaro Spallanzani (1765), who first used the technique to disprove the spontaneous generation theory.
1819	Peter Durand developed canning, the preservation of foods in steel cans. Charles Mitchell introduced the tin lining of metal cans in 1839.
1870	Louis Pasteur recommended heating wine at 145 °F (62.7 °C) for 30 min to destroy souring bacteria. Franz von Soxhlet advanced boiling milk for 35 min to kill

	contaminating bacteria. Later, this method was modified to kill mainly vegetative pathogens and many spoilage bacteria and named "pasteurization."
1895	Harry Russell showed that gaseous swelling with a foul odor in canned peas was due to the growth of heat-resistant bacteria (spores).

Foodborne diseases

1820	Justin Kerner described food poisoning from eating blood sausage (due to botulism). Fatal disease from eating blood sausage was recognized as early as A.D. 900.
1835	Christian Gottfried Ehrenberg described a soil-borne organism, *Vibrio subtilis*, and later, in 1872, Ferdinand Julius Cohn renamed it *Bacillus subtilis*.
1849	John Snow suggested the spread of cholera through drinking water contaminated with sewage. In 1854, Filippo Facini named the cholera bacilli *Vibrio cholerae*, and these were isolated in pure form by Robert Koch in 1884.
1856	William Budd suggested that water contamination with feces from infected persons spread typhoid fever and advocated using chlorine in the water supply to overcome the problem. In 1800, G. de Morveau and W. Cruikshank approved the use of chlorine to sanitize potable water.
1885	Theodor Escherich isolated *Bacterium coli* (later named *Escherichia coli*) from feces and suggested that some strains were associated with infant diarrhea.
1855	Theobald Smith reported the isolation of *Salmonella* from pig intestines and named it after his mentor, Daniel E. Salmon, a veterinary pathologist. The organism was later renamed *Salmonella enterica* serovar Choleraesuis.
1871	Von Recklinghausen isolated cocci from a diseased kidney and called them "micrococci." Later, in 1880, Sir Alexander Ogston, credited with discovering the pathogen, coined the name "Staphylococcus."
1880	Karl Joseph Eberth isolated *Salmonella* from a patient suffering from typhoid, and the organism was renamed *Salmonella enterica* serovar Typhi.
1886	Theodor Escherich was also credited with discovering a curved rod-shaped bacterium; in 1913, McFayeden and Stockman renamed this *Campylobacter.*
1891	William L. Welch isolated *Clostridium perfringens* from human autopsy samples.
1894	J. Denys associated pyogenic *Staphylococcus* with the death of a person who ate meat prepared from a diseased cow.
1894	Alexandre Yersin, a French scientist, first reported a bacillus as the causative agent for plague, and it was later named *Yersinia pestis. Yer. pestis* can cause foodborne diseases. *Yer. enterocolitica* and *Yer. pseudotuberculosis* are two members of the genus *Yersinia* and are known foodborne pathogens.
1895	Emil von Ermengem isolated *Bacillus botulinus* (*Clostridium botulinum*) from contaminated meat and proved that it caused botulism.
1926	E.G.D. Murry isolated *Bacterium monocytogenes* from rabbits, and it was later named *Listeria monocytogenes.*
1981	Stanley Prusiner characterized a prion as a causative agent for bovine spongiform encephalopathy or "Mad Cow Disease."

Microbiology techniques

1854	Heinrich Schröder and Theodore von Dusch used cotton to seal tubes and flasks to prevent microbial contamination in heated culture broths.
1876	Carl Weigert used methylene blue (a synthetic dye) to stain bacteria in aqueous suspensions.
1877	Ferdinand Cohn showed heat resistance of *Bacillus subtilis* endospores.

1878	Joseph Lister isolated *Streptococcus* (now *Lactococcus*) *lactis* from sour milk.
1880s	Robert Koch and his associates, including Julius Richard Petri, introduced many important methods that are used in all branches of microbiology, such as solid media (first gelatin, then agar) to purify and enumerate bacteria, the Petri dish, flagellar staining, steam sterilization of media above 100 °C, and photography of cells and spores.
1884	Hans Christian Gram developed Gram staining of bacterial cells.

FOOD MICROBIOLOGY: CURRENT STATUS

In the early twentieth century, the association and importance of microorganisms, especially pathogenic bacteria, in food were well established. Specific methods were developed for their isolation and identification. The importance of sanitation in food handling to reduce contamination by microorganisms was recognized. Particular methods were developed to prevent growth and destroy spoilage and pathogenic bacteria. Interest in the isolation of beneficial bacteria from fermented dairy products had begun. However, after the 1950s, food microbiology entered a new era. The availability of basic information on the physiological, biochemical, and biological characteristics of diverse types of food, microbial interactions in food environments, and microbial physiology, biochemistry, genetics, and immunology has helped open new frontiers in food microbiology.[1,7–11] Now, understanding microbial community and microbial interaction in complex food environments through microbiome analysis, whole genome sequencing, and the development of novel and natural intervention technologies are at the forefront of food microbiology.

Food Fermentation/Probiotics

- Development of strains with desirable metabolic activities by genetic transfer among strains
- Development of bacteriophage-resistant lactic acid bacteria
- Metabolic engineering of strains for overproduction of desirable metabolites
- Development of methods to use lactic acid bacteria to deliver biologically relevant proteins or vaccines
- Sequencing genomes of crucial lactic acid bacteria and bacteriophages for a better understanding of their characteristics
- Food biopreservation using food-grade microbes and their antimicrobial metabolites
- Knowledge of essential characteristics of probiotic bacteria and development of desirable strains
- Effective methods to produce starter cultures for direct use in food processing
- Bioengineered probiotics for health promotion and disease control

Food Spoilage

- Identification and control of new spoilage bacteria associated with the current changes in food processing and preservation methods
- Spoilage of frozen and refrigerated foods with an extended shelf life due to bacterial enzymes
- Development of molecular methods, including nanotechnology, to identify metabolites of spoilage bacteria and predict the potential shelf life of foods
- Importance of environmental stress for the resistance of spoilage bacteria to antimicrobial preservatives
- Microbial community, ecology, and quorum sensing in food

Foodborne Diseases

- Methods to detect emerging foodborne pathogenic bacteria from contaminated foods
- Application of molecular biology techniques, including nanotechnology and biotechnology, for rapid detection of pathogenic bacteria in food and environment
- Effective detection and control methods for foodborne pathogenic viruses
- Transmission potential of prion diseases from food animals to humans
- Importance of environmental stress for the detection and destruction of pathogens
- Factors associated with the increase in antibiotic-resistant pathogens in food
- Biofilm formation, microbial quorum sensing, and attachment of foodborne pathogens on food and equipment surfaces
- Mechanisms of pathogenicity of foodborne pathogens
- Effect of food or environment-related stress on gene regulation, pathogenicity, and survival
- Effective methods for epidemiological study of foodborne diseases: Genome-based approaches to study epidemiology, including whole genome sequencing and molecular fingerprinting techniques
- Application of bacteriophages and other natural antimicrobials for the control of foodborne pathogens
- Control of pathogenic parasites in food
- Mold and mycotoxin detection and control
- Fish and shellfish toxin detection and control
- Foodborne disease control strategies at pre-harvest (in the farm) and post-harvest food production
- The potential emergence of infectious and foodborne diseases from bushmeat, wildlife, and alternative protein sources, such as insects

Miscellaneous

- Application of hazard analysis of critical control points (HACCP) in food production, processing, and preservation
- Hazard analysis and risk-based preventive controls for human food
- Novel food-processing technologies
- Microbiology of unprocessed and low-heat-processed ready-to-eat foods
- Microbial control of foods from farm to table (total quality management)
- Food safety legislation, including the Food Safety Modernization Act of 2011

FOOD MICROBIOLOGY AND FOOD MICROBIOLOGISTS

From the above discussion, it is apparent what food microbiology as a discipline has to offer. Before the 1970s, food microbiology was regarded as an applied science mainly involved in the microbiological quality control of food. Since then, the technologies used in food production, processing, distribution, and retailing, and food consumption patterns, have changed dramatically. These changes have introduced new problems that can no longer be solved using applied knowledge. Thus, modern-day food microbiology must include basic science to understand and effectively solve the microbiological issues associated with food. The discipline consists of microbial aspects of food spoilage and foodborne diseases, effective control and bioprocessing, and essential information on microbial ecology, physiology, metabolism, and genetics. This information is helping to develop methods for rapid and effective detection of spoilage and pathogenic bacteria, to develop desirable microbial strains by recombinant DNA technology, to produce fermented foods of better quality and health benefits, to develop thermostable enzymes for the enzyme processing of food and food additives, to develop methods to remove bacteria from food and equipment surfaces, and to combine multiple strategies for the effective control of spoilage and pathogenic microorganisms in food.

An individual who has completed courses in food microbiology (both lecture and laboratory) should gain knowledge in the following areas:

- Determine the microbiological quality of foods and food ingredients by using appropriate techniques
- Determine the microbial types involved in spoilage and health hazards and identify the sources
- Understand factors contributing to the emergence of new pathogens or the reemergence of old pathogens
- Understand the primary mechanism of pathogenesis of foodborne microbes
- Design corrective procedures to control the spoilage and pathogenic microorganisms in food
- Learn rapid methods to isolate and identify pathogens and spoilage bacteria from food and the environment
- Identify how new and novel technologies adapted to food processing can have specific microbiological problems and design methods to overcome the problems
- Design effective sanitation procedures to control spoilage and pathogen problems in food-processing facilities
- Effectively use desirable microorganisms to produce fermented foods
- Design methods to improve starter cultures for use in fermented foods and probiotics
- Know about food regulations (state, federal, and international)
- Understand the microbiological problems of imported foods

To be effective, in addition to the knowledge gained, one has to communicate with different groups of people about the subject (food microbiology and its relation to food science, food processing, food biotechnology, and biomedical sciences). An individual with good common sense is always better positioned to sense a problem and correct it quickly.

CONCLUSION

Human civilization began when hunters and gatherers adopted not only the production but also the preservation of foods. Thus, long before the existence of microorganisms was discovered, their importance for food spoilage and health hazards was conceived by our early ancestors. Once their association and importance in food were proven, efforts were made to understand the basic principles associated with food and microbial interactions. This knowledge was used to control undesirable microbes and effectively use the desirable types. Current investigations are directed toward understanding microbes at the molecular level. A food microbiologist should understand the current developments in food microbiology and the characteristics of microorganisms important in food. The latter aspect is discussed in Chapter 2.

QUESTIONS

1. Describe briefly the contributions of the following scientists in the development of microbiology and food microbiology: (a) Leeuwenhoek, (b) Spallanzani, (c) Pasteur, (d) Koch, (e) Hess, (f) Cohn, (g) Lister, (h) Soxhlet, (i) Gartner, and (j) Appert.
2. Why did Needham's experiments fail to disprove spontaneous generation for microbes, but Pasteur succeeded in disproving that theory?
3. List three critical areas (each) of current studies in food biotechnology, food spoilage, and foodborne diseases.
4. Briefly explain the significant differences in understanding microorganisms' importance in foods before and after the 1900s.
5. List the pathogens that were proven to be associated with foodborne diseases before 1900.
6. Briefly describe what a food microbiology student is expected to know.

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