

Microbiology

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THE HISTORY OF Microbiology (or Bacteriology, as it was originally known) at the Massachusetts General Hospital (MGH) is a complex one. Although Dr. Reginald Heber Fitz's pioneering work established the pathogenesis of appendicitis in 1886 (15, 41; and see chapter 2), the laboratory specialty of microbiology owes its origins to Dr. James Homer Wright, who was appointed Pathologist at MGH in 1896. Wright was involved in a number of landmark studies in the etiology of parasitic and other infections, established the first diagnostic bacteriology laboratory functions at the MGH, and hired several physicians to undertake bacteriological work in the department. In 1925, just before the end of Dr. Wright's tenure as chief, the official position of Bacteriologist was established in the Department of Pathology, and this was held in succession by Drs. George Lawson and Merrill King over the next several years. In 1930 Dr. Louis Dienes was appointed Bacteriologist, a position he held until 1952. Under his direction the field of bacteriology began to flourish and the number of diagnostic studies carried out in the Clinical Bacteriology Laboratory increased dramatically. After Dr. Dienes's retirement in 1952, Thomas Fite Paine Jr. was appointed Chief of Bacteriology, and a separate department was established, beginning a nearly four-decade period (1952–1989) in which clinical microbiological studies were performed outside the Pathology department. Upon Paine's

abrupt departure the following year, Dr. Lawrence Kunz, who had come to the MGH as an Assistant Bacteriologist in 1952, assumed de facto responsibility for the laboratory until he was officially appointed chief in 1956. During his tenure the Department of Bacteriology developed close ties with the Infectious Diseases Division and the Department of Medicine. This was also a time during which a number of "boutique" clinical laboratories sprang up throughout the hospital. Microbiology and Infectious Diseases were no exception, and in rapid order independent laboratories of Antibiotic Blood Levels, Parasitology, and Virology were established in the Infectious Diseases Unit. All these had close relationships with the Bacteriology Laboratory as well. In 1989 the activities of the Bacteriology/Clinical Microbiology Laboratory were once again returned to Pathology under Clinical Pathology. With the consolidation of laboratories in the early 1990s, the activities in all these specialized infectious diseases laboratories were ultimately incorporated under Bacteriology to make it a true Clinical Microbiology Laboratory. Before this, however, Dr. Kunz had retired in 1982 and Dr. Mary Jane Ferraro had been appointed his successor. She has maintained leadership of the Clinical Microbiology Laboratories until the present time. Thus, in a period of just less than a century, Clinical Microbiology at the MGH evolved from a nascent discipline in Pathology to a separate

Department of Bacteriology, which ultimately became a full-fledged Division of Clinical Microbiology, and was returned to its original roots in Pathology, where it now resides as a flourishing clinical, research, and teaching operation.

During the first hundred years of Bacteriology/Clinical Microbiology at the MGH, the laboratory occupied a number of different venues. It began its existence under Dr. Wright in the Allen Street Building. By the 1930s bacteriology was carried out in a building adjacent to the Allen Street Building (see below). In the early 1950s the laboratory was relocated to the fourth floor of the Domestic Building, where it remained until it was moved to a temporary building erected on the lawn in front of the Bulfinch Building to enable the destruction of the Domestic Building and construction of the Gray Building. In 1968 the laboratory moved into new quarters on the fourth floor of the Gray Building, which had been made possible through a bequest from the estate of Mrs. Stephen S. Fitzgerald, in memory of her father, Francis Blake. Blake was the inventor of the Blake transmitter, which was developed for the Bell Telephone Company, and was once a Trustee of the MGH. The Francis Blake Bacteriology Laboratories provided much-needed space and facilities for the growing activities of the Bacteriology/Microbiology Laboratory.

THE WRIGHT ERA

Dr. James Homer Wright (chapter 4) served as Chief of the Department of Pathology at MGH from 1896 until 1926. In the early 1900s, Dr. Wright authored sections titled “Actinomycosis” and “Diseases Due to Vegetable Parasites Other Than Bacteria” for Dr. William Osler’s textbook *Modern Medicine*, evidence of his growing interest in infectious diseases (28). In 1903 he reported on the discovery of a protozoan in a tropical ulcer, but, although this was an independent observation, Dr. Wright was six months later than Charles Donovan in what turned out to be the discovery of the causative organism of

leishmaniasis. Dr. Wright’s monograph entitled “The Biology of the Microorganism of Actinomycosis” led to his receipt of the Samuel D. Gross Prize (53). During Dr. Wright’s tenure, laboratory testing for infectious diseases was established in Pathology, although the number of tests carried out was relatively modest by today’s standards. Initial tests involved studies of blood and urine but were expanded to include cerebrospinal fluid before Wright retired (1). After visiting a laboratory in Germany, Wright commissioned the building of the first water-jacketed incubator in the United States (figure 21.1), which was built by a Boston coppersmith named Peter Gray. This gas-powered incubator served to provide the appropriate environment for early bacterial cultures at the institution. It was placed on display in the MGH Ether Dome after its utility in the Bacteriology Laboratory ended.

A review of the annual reports of the Department of Pathology to the MGH Board of Trustees provides an interesting insight into the microbiology activities carried out in Pathology during the Wright era (1). As early as 1898, Dr. Wright reported that Pathology was involved in the “application of bacterial culture tests in cases of suspected diphtheria in the hospital wards” and “the testing of tissues and fluids for the presence of the bacillus of tuberculosis.” His 1899 report notes that Pathology had begun “examination of sputum for tuberculosis bacillus” and also observes that the laboratory was carrying out examinations of blood for the “malaria parasite.” He then went on to describe an interesting test for typhoid fever that was employed in 1899 but is not mentioned in subsequent reports: “In typhoid fever the blood test is of greatest value confirming the diagnosis. The test consisted of a drop of blood from earlobe or finger-tip mixed with a few drops of bouillon containing living typhoid bacilli, which caused the bacilli to gather together as clumps or groups.” He also noted that the laboratory was doing throat cultures for diphtheria and stated that the laboratory was beginning to study methods to cultivate



Figure 21.1 James Homer Wright (left) in his laboratory with the first water-jacketed incubator (far right) in the United States. The person standing at the bench at the back of the laboratory is most likely Louis Brown.

bacteria in the absence of oxygen “with special reference to the bacillus of tetanus or ‘lockjaw’”; and in 1900 he published a paper describing a rather crude but effective method for culturing anaerobic bacteria (57). It was at this time that he also began his own personal studies of actinomycosis (figure 21.2). These were remarkably pioneering efforts in early bacteriology, especially given the fact that they took place less than a quarter century after the establishment of the microbial basis for bacterial infections by Robert Koch and Louis Pasteur.

Wright’s diagnostic efforts and research studies in bacteriology were supported by several other physicians working in the Pathology department, including Drs. Mark Wyman Richardson and Albert E. Steele. Richardson did pioneering work on methods for diagnosing typhoid fever and was

prolific in this area, publishing eight articles on the subject in 1897 and 1898 alone. He went on to serve on the board of public health and published on inoculation against typhoid fever. Dr. Steele served as Assistant in Clinical Bacteriology from 1910 through 1926. He had trained at Boston City Hospital and at MGH. In MGH Pathology he played a major role in the therapeutic use of killed bacteria.

In 1907 the Pathology Department began to treat “certain bacterial diseases by subcutaneous injections of killed cultures of the infecting organism” (1). Hundreds of patients received this therapy on the wards and in two separate rooms in the Pathology Department. Indeed, 500 patients were treated in 1908, with “very satisfactory results.” It should be noted that this type of therapy was carried out in Pathology for

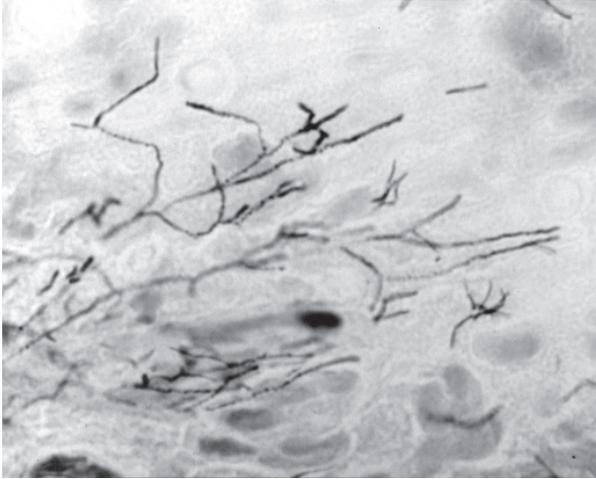


Figure 21.2 “Human Actinomycosis smear preparation from a ‘granule’ showing the branching filaments of the microorganism” by James Homer Wright (photograph by Louis S. Brown)

the next several decades (although in decreasing numbers), but no formal evaluation of the its results were ever given in the reports. In 1911 the laboratory began studies of the urinary excretion of Salvarsan (discovered by Paul Ehrlich in Germany only two years earlier) and began working on a modified serological test for syphilis. By 1912 Dr. Wright’s laboratories were carrying out more than 2,000 “bacterial evaluations for diphtheria, typhoid fever and other diseases” and had inoculated 300 guinea pigs for tuberculosis. The serological testing for syphilis had begun in earnest, and 2,262 Wassermann tests were performed in the laboratory that year. The number of bacterial examinations carried out in the laboratory remained essentially stable until near the end of Dr. Wright’s tenure, but the number of serological tests for syphilis had increased almost eightfold, to 16,291 in 1925. In 1926 the official position of Bacteriologist was established in the Department of Pathology, and this was filled in rapid succession by Dr. George Lawson and then by Dr. Merrill King. Lawson was appointed Bacteriologist in 1927, but in 1928 he left to accept a position as Associate Professor of Bacteriology at the University of Louisville (53). Dr. King was

appointed Bacteriologist in his place, but he left in 1930 after a brief tenure to enter a career in ophthalmology, which led to the appointment of Dr. Louis Dienes (53).

THE DIENES ERA

Dr. Louis Dienes (figure 21.3) was appointed Bacteriologist in the Department of Pathology in 1930, a position he held until his retirement in 1952 (53). Dienes was born in Tokay, Hungary, in 1885 and received a medical degree from the University of Budapest in 1908. In 1913 he began to work in bacteriology research in the laboratory of Professor Fred Neufeld at the Robert Koch Institute in Berlin. He then served as an Army field bacteriologist in Poland and Ukraine during World War I, which provided him with a vast experience in outbreaks of cholera, typhoid, and typhus fever. He developed a particularly valuable collaboration with Professor Edmund Weil in the study of epidemic typhus fever and his work on the Weil-Felix reaction began to focus his interest on the immune response in infectious diseases. In 1922 he went to the von Ruck Research Laboratory at a tuberculosis sanitarium in Asheville, North Carolina, where he began to study the immune response in tuberculosis. Through this research he was able to characterize the histological features of the tuberculin reaction and noted that these are distinctive reactions that differ from other types of inflammation and from immunologic reactions mediated by circulating antibodies—a phenomenon we now know as delayed-type hypersensitivity (47). He also noted that delayed-type hypersensitivity can be elucidated with other nonbacterial antigens and is not unique to infection with the tubercle bacillus. His work also paved the way for using an adjuvant to enhance the development of delayed-type hypersensitivity.

In 1930 Dr. Dienes moved to Boston to work with Hans Zinsser in the Department of Bacteriology and Immunology at Harvard Medical School (HMS), and he was appointed



Figure 21.3 Louis Dienes

Bacteriologist at the MGH, where he continued his studies of delayed-type hypersensitivity in close collaboration with Tracy B. Mallory until the mid-1930s (chapter 23). At that point he changed research interests and began studying the so-called L-forms derived from *Streptobacillus moniliformis*. His work defined the stages that led to the development of L-forms from the bacillary form of *S. moniliformis*, and he demonstrated the spontaneous development of L-forms from other pleomorphic organisms (7). He was able to show that penicillin and glycine could be used to induce the development of L-forms of the species in which they did not occur spontaneously and ultimately demonstrated through electron microscopy that L-forms lack a complete cell wall (8). His work with L-forms led to work with a morphologically similar pleuropneumonia-like organism, which he initially isolated from the genital tract of a patient in the late 1930s. These organisms, which are now known to be mycoplasmas, were first isolated by Dr. Dienes in pure culture from an abscess of a Bartholin's gland cyst.

During Dr. Dienes's early years at the hospital, office and laboratory space was primitive by today's standards. The building housing the Bacteriology Laboratory was over 100 years old and had formerly been the stable housing the

horses' wagons used for transport of patients to the hospital. The small building on Allen Street separated these important facilities from the rest of the hospital. Included among them were the Pathology Laboratories, the autopsy room, and the "cold room," where cadavers were held. A large amphitheater overlooked the autopsy area. Also present were the Bacteriology media-making room, with its stoves and autoclaves, and the "animal farm," which was a vital research facility. Despite these limitations, Dienes was able to carry out his important research and to oversee the growing clinical activities of the Bacteriology Laboratory.

Dr. Dienes continued the bacterial examinations for diphtheria, typhoid, and other organisms started by Dr. Wright. He also performed the serological tests for syphilis, which continued to grow in volume (1). In 1928 the laboratory switched from the Wassermann to the Hinton test after it was developed by William A. Hinton, a professor at HMS who had worked in MGH Pathology in the early 1920s (chapter 3). This test for syphilis was thought to be more accurate than the earlier Wassermann examination. In 1933 the laboratory began typing pneumococci, and the number of bacterial examinations carried out by the laboratory increased to 5,160; 20,773 Hinton tests were also performed in 1933. Although the number of patients treated with bacterial vaccines in Pathology persisted, the actual number of patients receiving this therapy continued to decrease, and by 1940 only 30 patients were treated with vaccines in Pathology. That year the number of bacterial cultures jumped dramatically, to 17,292, a direct result of the discovery of the sulfonamides and the beginning of the antibiotic era. Guinea pig inoculations for tuberculosis totaled 1,056, and 831 pneumococcal typing tests and 30,150 Hinton tests for syphilis were carried out in the laboratory. In 1951, just a year before Dr. Dienes retired, it was noted that the number of bacterial cultures had remained stable over the preceding decade, at just over 17,000 (1).

Louis Dienes retired as head of the Bacteriology Laboratory in 1952 but continued his research in a small laboratory at the MGH where, with the technical assistance of Sarabelle Madoff Annenberg, he collaborated with members of the Arthritis (now Rheumatology) Unit until 1970. He died on January 31, 1974. In 1976 his daughter, Dr. Priscilla D. Taft, Director of MGH Cytopathology, was among those present for the unveiling of the plaque dedicating the Louis Dienes Conference Room in the Francis Blake Bacteriology Laboratories.

THE KUNZ ERA

Upon the retirement of Louis Dienes, major changes occurred in the Bacteriology Laboratory. Thomas Fite Paine Jr. was named Chief of the Bacteriology Laboratory in 1952, and the Bacteriology Laboratory became a separate, independent department, severing its direct ties with Pathology (11). Paine's tenure, however, was short, and he left within a year to take a position at Vanderbilt University. At that point Dr. Lawrence J. Kunz, who had been serving as an Assistant Bacteriologist at the MGH, assumed responsibility for the laboratory; in 1956 he was made permanent Chief of the Department of Bacteriology (figure 21.4). Kunz was born in Brooklyn, New York, where he graduated from Brooklyn Preparatory School. He attended and graduated from St. Peter's College in New Jersey. Following service in the 15th Medical Corps in Italy during World War II, he earned a Ph.D. in bacteriology from Harvard University, working with the Nobel laureate Dr. John Enders.

Under Kunz's direction, the Bacteriology Laboratory was ushered into the modern era. By the late 1950s the volume of specimens for routine bacterial cultures, fungal cultures, mycobacterial cultures, and serology had skyrocketed as medicine entered the expanding antibiotic era, which allowed for specific treatment of infections with known etiology. The relocation of the laboratory to the new Francis Blake Bacteriology



Figure 21.4 Lawrence J. Kunz

Laboratories on Gray 4 (now Gray 5) served a number of important functions. It provided the space and facilities for rapid expansion of the laboratory and, because of its proximity to the Infectious Diseases Unit, it allowed many fruitful collaborations between Bacteriology and Infectious Diseases. Before the movement of the laboratories to Gray 4, the cramped space in the old Domestic Building and its relative isolation precluded many of the advantages realized after the move to the Gray Building. In the early 1960s there was very little if any contact between members of the MGH house staff and the laboratory. Infectious Diseases was in its infancy, Dr. Morton Swartz having been appointed chief of the nascent unit in 1956, which in the early 1960s consisted of Drs. Swartz, Frank Austen, Paul Black, and, subsequently, Arnold Weinberg. Drs. Kunz and Weinberg, who was also from Brooklyn, quickly became friends and close colleagues; Weinberg joked that although he didn't know Kunz as a youth, he strongly suspected that Kunz was one of the boys that used to throw stones at him as

he walked through his neighborhood on his way to school!

Many diagnostic activities were carried out or initiated outside the laboratory. In 1955 there were incubators on Burnham 6, on Bulfinch 2, and in the Emergency Ward so that house staff and physicians could plate their own cultures when the Bacteriology Laboratory was not open nights and weekends. A memorandum from Dr. Kunz in 1956 reminded the house staff that "plated out cultures which were submitted to the laboratory must be streaked out on the surface of the agar with an inoculating needle. Specimens streaked out with a swab are unsatisfactory." The laboratories in the Emergency Ward and on all the major clinical units provided materials for house staff to carry out their own Gram stains, buffy coat smears, routine urinalysis, and blood counts. As a result, members of the medical house staff in particular were well trained in carrying out and interpreting Gram stains of sputum, urine, and other specimens.

In 1955 Massachusetts experienced a major polio epidemic. Because of the large number of patients with respiratory difficulties and the shortage of professional personnel on the wards, as well as the need for repeated bacteriological cultures, particularly of the respiratory tract, a special section of the Bacteriology Laboratory was temporarily set up on White 9 in the charge of a second-year medical student, John Livingstone. Fortunately, Livingstone had been working in the Bacteriology Laboratory during the summer, and that made it possible for him to take charge of this special unit, which monitored the flora of patients who were at continual risk of contracting bacterial infections. Livingstone's work not only provided immense benefit to the patients stricken with polio, but it resulted in a 1957 publication in the *New England Journal of Medicine* (29).

Dr. Kunz began a number of research studies in the laboratory, beginning with work in collaboration with William Ewing at the Centers for

Disease Control (CDC) on the taxonomy of the so-called paracolony bacilli, which includes many of the currently recognized species of *Enterobacteriaceae* (13). He also was among the first to make effective utilization of the Bacteriology Laboratory for the identification of nosocomial infections. Thus, with Dr. Orjan T. C. Ouchterlony, a Swedish physician and bacteriologist who was a visiting professor at MGH in 1954, he and his colleagues discovered that brewers yeast contained three different types of *Salmonella* as contaminants that were causing infections in hospitalized patients (25). One of the most intriguing clinical investigations involved the solving of an outbreak of nosocomial infections at the MGH with a rare species of *Salmonella*, *Salmonella cubana*, which was ultimately traced to carmine red dye that was used in the 1960s as a measure of gastrointestinal motility (27). The detective work carried out by Kunz, his colleague Dr. David Lang, and a laboratory technologist, Harriet Provine, served as the basis for a number of scientific papers and generated a *New Yorker* article by Berton Rouché entitled "The Santa Claus Culture." With closer collaboration between the Bacteriology Laboratory and members of the Infectious Diseases Unit, a number of important clinical investigations were carried out and published. These included major studies of anaerobic infections, extrapulmonary streptococcal infections, salmonellosis, listeriosis, endocarditis due to group D streptococci or enterococci, and infections due to *Acinetobacter calcoaceticus* (2, 9, 17).

In 1968, while serving as a research fellow in Infectious Diseases at the MGH, Dr. Robert C. Moellering Jr. began a series of collaborations with Dr. Kunz in the Bacteriology Laboratory. By using novel sealed agar plates provided by Dr. Kunz, Dr. Moellering was able to carry out a number of cultures under jungle conditions in the Solomon Islands (34). He used this method to obtain soil cultures from the environment as well as wound and stool cultures from the natives in an area on Malaita, where antibiotics had not

previously been used clinically. From these cultures Dr. Moellering and his colleagues isolated bacteria with transferable resistance to streptomycin and tetracycline, thus providing the first evidence that resistance genes existed in nature independent from the clinical use of antimicrobial agents (14). Dr. Moellering also provided soil samples to the Mycobacteriology section of the MGH Bacteriology Laboratory, from which were isolated a series of “atypical” mycobacteria that probably provided the source of positive skin tests for atypical mycobacteria among the Solomon Islanders. This work served to suggest strongly that exposure to “atypical” mycobacteria does not provide protective immunity to tuberculosis, which had recently been introduced into the Solomon Islands (32, 42).

Upon completion of his clinical and research fellowship in Infectious Diseases, Dr. Moellering joined the full-time staff in the Infectious Diseases Unit and established an independent research laboratory. He then received a small hospital stipend to serve as a formal Consultant to the Bacteriology Laboratory and collaborated with Dr. Kunz in a number of projects relating to antimicrobial susceptibility testing and laboratory automation. They developed a novel mechanical method for inoculating plated media used in antibiotic susceptibility tests (24) and began evaluation of a computer-associated electronic zone analyzer, which fed data on antimicrobial susceptibility testing directly into the laboratory computer (37). This proved to be a powerful tool for evaluating antimicrobial susceptibilities of a large number of organisms in the laboratory and yielded invaluable data for evaluation of new antimicrobial agents such as tobramycin and amikacin (35), for monitoring the development of antibiotic resistance, and for providing annual summaries of institutional-based data to guide initial empiric therapy (4, 36, 38, 39). By feeding data directly from susceptibility test results into the computer, it was also possible to develop a real-time quality assurance system that identified

errors in susceptibility test results or organism identification, based on “sensi-ID discrepancies” (37). These activities, which were unique to MGH at the time, have now become routine or mandated in today’s microbiology laboratories.

Clinical microbiology laboratories are among the few laboratories analyzing clinical specimens that have proven difficult to automate. Appropriate isolation and identification of bacteria, viruses, fungi, and other pathogens still require individual, subjective, technical judgment. Dr. Kunz, however, was among the first to realize the potential of the computer for the accurate collection and analyzing of data generated in the Bacteriology Laboratory and for transmission of the data to the clinicians caring for infected patients. He collaborated with members of the Laboratory of Computer Science at MGH, including James Poitras and Dr. G. Octo Barnett, to create a system that was uniquely suited to the needs of the hospital (26, 37). Although a number of commercial systems for microbiology laboratory computerization have been developed in subsequent years, none has proven more flexible, sophisticated, and user-friendly than the original program developed by Kunz and his colleagues. Not only did the system provide a valuable tool for analyzing large amounts of data and providing suitable reports on susceptibility patterns, frequency of isolation of various pathogens from specific body sites, and the like, but it also served to eliminate many of the transcription errors that occur when massive amounts of data are analyzed and transmitted manually.

During this period Dr. Barbara K. Watson was recruited to the laboratory, joining Dr. Theodore F. Medrek in a research investigation on streptococci. They carried out studies relating to streptococcal taxonomy in the Bacteriology Laboratory, and both collaborated with Moellering and other members of the Infectious Diseases Unit in studies relating to their clinical or research activities. Particularly useful among Dr. Watson’s studies were a number of projects defining more accurate

ways to identify streptococci to the species level (9, 54, 55). Dr. Watson retired in 1975. Eventually, Dr. Kathryn L. Ruoff, who joined the Bacteriology Laboratory in 1981 as an Assistant Bacteriologist, continued the research work on streptococci and produced some particularly valuable results on the pathologic significance of *S. bovis* and related species (46). Dr. Ruoff received her Ph.D. degree from Cornell University in 1975 and early in her professional career taught microbiology at Cornell and at Wheaton College in Massachusetts. She contributed greatly to teaching her discipline both in the MGH laboratory and at HMS, where she was promoted to Associate Professor of Pathology in 1997. In 2002 she left MGH to assume a position at Dartmouth-Hitchcock Medical Center and was appointed Associate Director of Microbiology in 2007.

THE BOUTIQUE INFECTIOUS DISEASES LABORATORIES

Because of a growing need for specialized clinical laboratory diagnostics beyond those of bacteriology and mycology, the Department of Medicine's Infectious Diseases Unit established three small, independent clinical laboratories during the 1970s. These laboratories were among a number of boutique laboratories that had sprung up over the years at the MGH and performed tests for Parasitology, Antibiotic Blood Levels, and Virology; the latter two arose from active research programs that found themselves also providing necessary but unfunded clinical services.

In the early 1970s Dr. Robert Moellering (figure 21.5) and Christine Wennersten began to carry out assays for concentrations of antimicrobial agents in patients' serum in Moellering's research laboratory. Initially these laboratory-developed bioassays were used to measure serum levels of aminoglycosides (gentamicin, tobramycin, and amikacin) and vancomycin and allowed for the study of appropriate dosage of aminoglycosides in specialized situations, such as children with extensive burns, and for the determination of

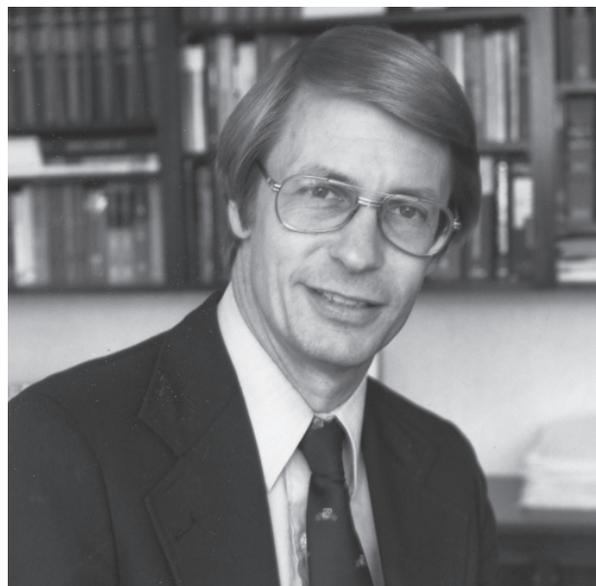


Figure 21.5 Robert C. Moellering Jr.

pharmacokinetic parameters and dosage recommendations that had previously not been available for drugs such as vancomycin (16, 23, 33). In the mid-1970s the research laboratory began to serve as a clinical laboratory for antibiotic assays, and fluorescent polarization enzyme immunoassays eventually became available commercially and replaced the labor-intensive bioassays. Shortly after Dr. Moellering's departure in 1981 to become Chair of the Department of Medicine at New England Deaconess Hospital, the activities of his former Antibiotic Blood Level Laboratory were consolidated into the Microbiology Laboratory. In December 1991 these assays were transferred to the Chemistry Laboratory, allowing for seven-day service, more automated enzyme immunoassay testing, and rapid reporting.

The Parasitology Laboratory was developed in the Infectious Diseases Unit at approximately the same time. Initially supervised by Drs. Gordon Moore, an infectious diseases fellow who had obtained training in parasitology at the CDC, and Dr. Morton Swartz, the laboratory was subsequently overseen by Dr. John Wolfson. Barbara Parkhurst and later Mary Ann Waldron served as the supervisors and major laboratory resources

for this operation. They assisted Dr. Wolfson in a series of annual lectures on the clinical and laboratory diagnosis of parasitic diseases, which were mandatory for infectious diseases fellows and offered to Pathology residents. Thick and thin smears for the laboratory diagnosis of malaria, one of the true medical emergencies in infectious diseases, were carried out by infectious diseases fellows during evenings, nights, and weekends, when the laboratory was not open. In 1991, following Dr. Wolfson's untimely death, the Parasitology Laboratory was incorporated into the Clinical Microbiology Laboratory, which allowed the eventual cross-training of technologists and incorporation of test results into the Microbiology Laboratory's computer-generated patient reports. Dr. Anthony Mattia assisted in the direction of this section of the laboratory from 1993 to 1994.

The need for a Diagnostic Virology Laboratory at MGH was recognized in the early 1970s. Improvements in culture techniques for a variety of viruses, early studies on antiviral therapy at MGH and elsewhere, and curtailment of state and federal diagnostic services combined to emphasize the need for an in-house laboratory. Until 1974, the few MGH diagnostic studies done were performed as part of therapeutic studies in herpes simplex encephalitis and varicella-zoster virus infections and were performed in the research laboratory of Dr. Martin Hirsch of the Infectious Diseases Unit.

Early in 1974 Dr. Hirsch (figure 21.6) initiated discussions with Dr. Lawrence Kunz, Director of the MGH Bacteriology Laboratory, regarding a joint effort to begin diagnostic virology at the hospital. Dr. Kunz generously offered space on Gray 5 for this purpose, and Drs. Hirsch and Morton Swartz, Chief of the Infectious Diseases Unit, prepared an application to the MGH Resource Allocation Board for approval of such a laboratory, with the active support of many other departments (e.g., Medicine, Pediatrics, and Surgery) that would benefit from such a service. This

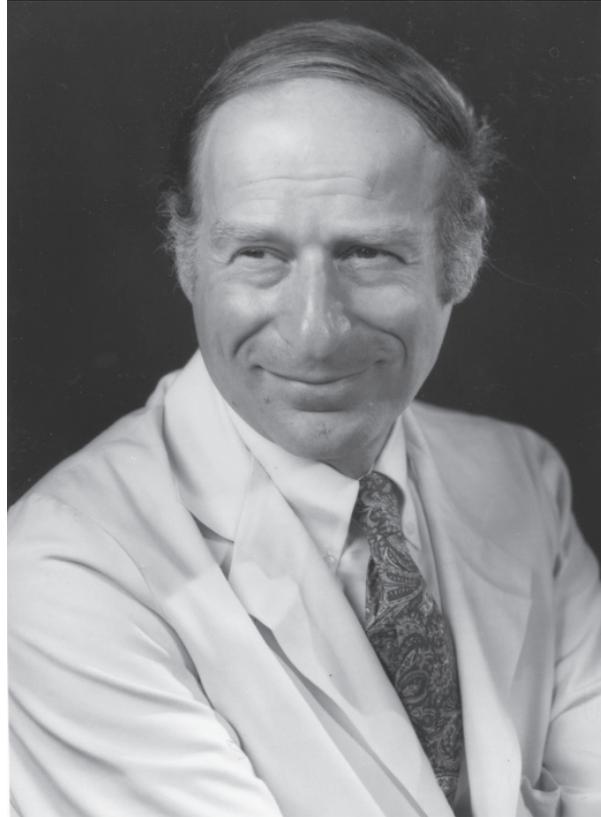


Figure 21.6 Martin S. Hirsch

application was approved, equipment was purchased, and the first laboratory personnel were hired in late 1974 and early 1975.

During the first year of operation, 771 specimens were received for virus isolation, of which 129 were positive, and 1,271 specimens were received for antibody studies. In 1977 the first attempts at rapid diagnosis (immunofluorescent detection of herpes group viruses) were begun, and in 1979 chlamydia diagnosis was added to the tests offered (10, 21, 51). The Virology Laboratory was incorporated into the Microbiology Laboratory in 1991 and subsequently directed by Drs. Mary Jane Ferraro and Angela Caliendo (see below). Dr. Hirsch, however, remained a valued consultant to Microbiology and continued to participate in important decisions regarding implementation of more rapid nonculture methods for viral diagnosis (31).

Each of these three laboratories in the

Infectious Diseases Unit had close ties with the Bacteriology Laboratory, and in the early 1990s, when administrative responsibility for the Microbiology Laboratory was returned to the Department of Pathology, they were incorporated into the clinical laboratory to form the modern-day Clinical Microbiology Laboratory. The establishment of these specialized clinical laboratories in the Infectious Diseases Unit and their ultimate consolidation into the Clinical Microbiology Laboratory in Pathology marked the continuation of a close and mutually beneficial association between the laboratory and the Infectious Diseases Unit. The association began informally with collaborative studies between Bacteriology and the newly formed Infectious Diseases Unit in the 1960s, as noted earlier. It was codified in 1973 when Dr. Moellering was named a formal Consultant in Bacteriology, a position he held until his departure in 1981. In 1982 Dr. Arnold Weinberg was named Consultant in Bacteriology and provided valuable assistance to the laboratory for several years. Ultimately, this position was incorporated more formally into the laboratory with

the direct Pathology appointments of several key members from the Infectious Diseases Unit as Assistant Directors, starting with Dr. Angela Caliendo in 1994 and Dr. Eric Rosenberg in 2002. In many ways this association has been a model for collaborative efforts between academic departments of Pathology and Medicine in joint research and training activities.

Dr. Lawrence Kunz retired from his position as Chief of the Clinical Microbiology Laboratory in December 1982. In 1993, 11 years after his retirement, Kunz was honored by having a newly classified bacterial species, *Helcococcus kunzii*, named after him (6) (figure 21.7). Appropriately, this organism had been discovered and characterized at the MGH by Dr. Kathryn Ruoff, who was instrumental in its naming. Dr. Kunz died on October 27, 1999.

THE PRESENT ERA

Dr. Mary Jane Ferraro took over direction of MGH Microbiology upon Dr. Kunz's retirement in 1982. Dr. Ferraro received her Ph.D. in Medical Sciences–Microbiology from Boston University



Figure 21.7 Reception in honor of the newly classified bacterial species *Helcococcus kunzii*. Left to right: Mary Jane Ferraro, Lawrence J. Kunz, Kathryn L. Ruoff, Arnold N. Weinberg, Katherine Kunz, Morton N. Swartz.

School of Medicine, where she worked with Dr. Sidney Cooperband. She then completed studies leading to an M.P.H. and postdoctoral training at the Harvard School of Public Health, which included internships at the Massachusetts Department of Public Health Laboratories and MGH. She was recruited to the MGH in 1976 as a junior staff member in Medicine and Assistant Director in the Bacteriology Laboratory. In the laboratory she began a number of research studies relating to automated, rapid, and cost-effective diagnostic methods, antimicrobial susceptibility testing, and other applications of clinical microbiology (3, 12, 18, 20, 22, 43, 45, 52). Her work has led to setting worldwide standards and guidelines for susceptibility testing of bacteria and other microorganisms (48, 49). She became Professor of Pathology in 2004 and Professor of Medicine in 2006, which places her among a handful of women ever to have held dual HMS professorships.

Under Dr. Ferraro's direction there was a rapid expansion in laboratory services, including, as noted earlier, the incorporation of parasitology and virology operations in the Clinical Microbiology Laboratory and the implementation of a night shift, thereby creating the first 24/7 microbiology service operation and thus eliminating the need for house staff and infectious diseases fellows to inoculate important cultures and perform Gram stains during off-hours. New approaches to nonculture-based laboratory testing were implemented, including direct antigen testing for some bacterial and fungal pathogens as well as nucleic acid-based tests for the detection of *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, and *Mycobacterium tuberculosis* (56). A same-day cytomegalovirus (CMV) antigenemia test replaced slower CMV blood cultures (31), and respiratory virus cultures were discontinued in favor of rapid direct fluorescent antibody (DFA) testing for respiratory viruses. Moreover, by posting the summaries of the DFA results of tests for respiratory viruses in real time, the laboratory

provided a valuable early warning system for the clinicians at the MGH and other local institutions concerning possible seasonal outbreaks of respiratory virus infections.

The appointment of Angela Caliendo as Assistant Microbiology Laboratory Director in 1994 marked the further introduction of molecular diagnostic methods in Microbiology, particularly those utilizing polymerase chain reaction (PCR). Dr. Caliendo was exceptionally well trained to develop this new service, having received her Ph.D. in biochemistry in 1983 from Case Western Reserve University, where she continued studies leading to her M.D. in 1987. Following her residency in internal medicine at Brigham and Women's Hospital, she completed a fellowship in infectious diseases at the MGH. Her initial efforts led to the development of a "home-grown" system for identifying herpes simplex virus in the cerebrospinal fluid (CSF) of patients with meningitis and encephalitis. Following this, the laboratory initiated HIV-1 viral load testing and made the test available for patient care long before it was FDA-cleared. This revolutionized the care of HIV-infected patients at MGH and provided the support for many seminal clinical studies of HIV in collaboration with members of the Infectious Diseases Unit (30, 40, 44, 50), including the definitive description of acute HIV infection as a mononucleosis-like syndrome by Dr. Eric Rosenberg during his infectious diseases fellowship. During the mid-1990s, Drs. Caliendo, Ferraro, and their colleagues also studied or incorporated a number of molecular diagnostics, including PCR for cytomegalovirus (19) and *Pneumocystis carinii* (5), and molecular tests for atypical respiratory pathogens such as *Mycoplasma pneumoniae* (the genera originally studied by Dr. Louis Dienes), and *Chlamydia pneumoniae*. Dr. Caliendo left MGH in 1999 to become Medical Director of Microbiology and Molecular Diagnostics at Emory University in Atlanta, and she was subsequently named Vice Chair for Clinical Pathology in the Department of Pathology



Figure 21.8 MGH Microbiology, 2010. Left to right: JiYeon Kim (fellow), Eric S. Rosenberg, Mary Jane Ferraro, John A. Branda.

and Laboratory Medicine. Shortly after, Dr. James Versalovic, who had just completed clinical pathology residency training at MGH, joined the Microbiology Laboratory as an Assistant Director with responsibility for molecular diagnostics. Dr. Versalovic left MGH in 2001 for Baylor University College of Medicine, where he became Chairman of the Pathology Department at Texas Children's Hospital in 2009. In 2002 Dr. Eric Rosenberg became Associate Director of the Microbiology Laboratories with responsibility for the molecular microbiology section. A 1991 graduate of Mount Sinai School of Medicine, Dr. Rosenberg trained in internal medicine at the University of North Carolina and completed his infectious diseases fellowship at the MGH. While still maintaining his research and clinical activities in infectious diseases, he increased the test menu to include PCR-based viral load tests for hepatitis viruses and genotyping.

In addition to its diagnostic and research activities, the Clinical Microbiology Laboratory continues its strong tradition of education. Several members of the laboratory staff devote significant effort to teaching of medical students in the clinical laboratory. Of particular note has been Dr. John Branda; an HMS graduate, he was appointed Assistant Director of Microbiology in 2004 after finishing his residency in clinical pathology at MGH, and he has been closely involved in the teaching of microbiology at HMS. Pathology residents and infectious diseases fellows have regular rotations in the laboratory, where they receive intensive immersion in all aspects of clinical microbiology. Regularly scheduled "plate rounds" are one of a number of joint teaching efforts of the Clinical Microbiology Laboratory faculty and the Infectious Diseases Unit. In 2010 a formal fellowship in clinical microbiology was started; it was approved by the ACGME in 2011.

In addition, medical or pediatric residents and infectious diseases fellows and attendings regularly stop by the laboratory to check on culture results and for informal training and consultation.

CONCLUSION

Clinical microbiology is now a full-service laboratory, with several faculty directors, a microbiology fellow (figure 21.8), and approximately 70 employees, that processes over 500,000 specimens per year. Included among these many tests are occasional requests for studies of the same type that James Homer Wright performed to assist in the diagnosis of diphtheria, tetanus, malaria, typhoid fever, and tuberculosis. These studies, however, are overwhelmed by a plethora of new tests that have been brought online in recent years.

The microbiology laboratories, over the more than 100 years of their existence, witnessed the first applications at the MGH of the discoveries of Pasteur, Koch, and others—research that provided the first rational basis for the diagnosis and ultimate treatment of infectious diseases. They participated, several generations later, when the discovery and clinical application of the sulfonamides and penicillin ushered in the golden age of the management of infectious diseases, making the activities of the clinical microbiology laboratory a key player in the battle against severe infections. The last half of the twentieth century and the dawn of the twenty-first have seen striking improvements in therapy for fungal and viral infections as well—all the more important as modern medicine and the HIV virus have markedly increased the numbers of immunocompromised patients with a wide variety of infections unimaginable by James Homer Wright and his colleagues. Throughout all these changes, the MGH microbiological resources have adapted to meet each new challenge. The battle is far from over, but it is certain that the activities of MGH Microbiology will continue to evolve and provide benefit for the patients at the MGH and elsewhere well into the future.

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