ACS
Chemistry for Life®

www.acs.org

A National Historic Chemical Landmark

The Development of Diagnostic Test Strips

May 1, 2010

American Chemical Society

"He [Al Free] said, 'Well, instead of doing it that way, we could get rid of the dropper if we just dipped the paper into the urine.' That's what started it."

— Helen Murray Free

ORIGINS: EARLY DIAGNOSTIC TESTS

In 1938 Dr. Walter Ames Compton joined Miles Laboratories in Elkhart, Indiana, a company best known for Alka-Seltzer®. Miles intended to pursue a "wonder" drug, allowing the firm to move into the lucrative prescription business. Compton had other ideas.

From his experience as an intern, Compton appreciated the inadequacy of existing tests for analyzing a patient's urine. Benedict's reagent was the primary test for glucose in urine, an indication of diabetes. The test required mixing urine with a reagent in a test tube, then heating it over a Bunsen burner. A color change indicated the presence and amount of sugar in the urine.

The procedure was slow and inaccurate. Compton, as head of R&D, pushed for the development of a better test. Building on Miles' experience with Alka-Seltzer, Compton and colleagues Jonas Kamlet and Maurice Treneer searched for a method to put reagents in an effervescent tablet that could determine the amount of sugar in urine. They succeeded, and in 1941 Miles introduced the effervescent tablet Clinitest®.

Free assumed that analytes in urine could be detected on a strip of paper containing reagents that produced color changes.

Clinitest tablets contained cupric sulfate, sodium hydroxide, and citric acid mixed with carbonate to make it fizz. Glucose could be measured by adding a few drops of urine to a tablet in a test tube and charting differences in color. Clinitest more accurately measured the amount of glucose than previous tests, making it an effective clinical diagnostic tool. Even though it was more expensive than Benedict's test, Clinitest quickly became a strong seller because it was more convenient and it did not require an external source of heat, so it easily could be performed and read in a doctor's office or hospital.

CLINISTIX®: THE FIRST DIP & READ TEST

In 1946 Alfred Free joined the Ames Division of Miles Laboratories to set up a biochemistry division. Free had a Ph.D. in biochemistry from Western Reserve University and additional research experience at the Cleveland Clinic. He assembled a research team, and a young woman working as a quality control chemist at Miles interviewed for a position. Free hired Helen Murray, and in 1947 they married, starting a long personal and professional relationship.

Free's team improved Clinitest by making it more sensitive, then turned to a second key test for diabetes, using nitroprusside to detect ketones. This resulted in Acetest®. After several more innovations, Free wondered if there were a better way to do the test. Helen Free remembers: "It was Al who said, 'You know, we ought to be able to make this easier and even more convenient than tablets, so no one would have to wash out test tubes and mess around with droppers.'"

Free assumed that analytes in urine could be detected on a strip of paper containing reagents that produced color changes. Free's team also knew that Clinitest detected the presence of any sugar, not just glucose. That diminished the utility of Clinitest for doctors who needed to measure specifically the presence of glucose in urine. The second challenge for the Free team was to embed the reagents on a filter paper strip. The result was dip-and-read Clinistix®, the first test specific for glucose, released in 1956. The researchers used a double sequential enzymatic reaction: glucose oxidase and peroxidase. The process was very labor intensive. Researchers cut the filter paper, dipped it into reagent solutions, and dried the paper in ovens.

MULTIPLE TESTS

In 1957 Miles introduced Albustix®, a dip-and-read test for protein in urine. The company now had diagnostic procedures for the two most common urine tests. Other tests followed. The development of additional diagnostic tests led to another breakthrough: combining reagents for two or more tests on one strip as a further convenience for the user.

Combining two reagents on one strip required creating a water-impervious barrier between the reagents on the paper to prevent the reagents from running together and compromising results. Uristix®, released in 1957, combined tests for glucose and protein. In the twenty years that followed, Miles developed and manufactured reagents to measure ketones, blood, bilirubin, urobilinogen, protein, nitrite, urinary leukocytes, and pH. The Frees became recognized experts in the field of urinalysis and published several texts and monographs.

Miles continued to add more tests to the strips. In 1981 the company added a specific gravity test and introduced the ten-reagent urine strip, Multistix® 10 SG. Miles also developed the Clinitek® instrument to facilitate the reading and interpretation of urine test results in physicans' offices and hospitals. Urine strip testing became a standard practice for patients seen in physicians' offices, medical clinics and hospitals as an initial indicator of metabolic, kidney, and liver disorders.

INSTRUMENT-BASED FINGERTIP BLOOD TESTING

Urine testing does not provide a real-time picture of blood glucose levels since glucose levels in urine lag behind those in blood. In 1964, Miles released Dextrostix®, reagent strips for testing blood glucose. Five years later Miles introduced the Ames Reflectance Meter® (ARM), invented by Anton

Clemens. Bulky and heavy by modern standards, and powered by a lead-acid battery, the analog device was the first portable blood glucose meter.

Although marketed for use by professionals, the ARM proved effective for patient self-testing. Later improvements by numerous companies included optically read test strips, electrochemical strips, the ability to test capillary blood from anatomical sites other than the fingertips, and continuous blood glucose monitors. Today, self-management of blood glucose is common clinical practice in the management of diabetes.

In the end, Miles Laboratories never discovered its "wonder" drug, but, as Helen Free says, "they sure went hog wild on diagnostics, and that's all Al's fault. He was the one who pushed diagnostics."

HELEN FREE

Helen Mae Murray was born in 1923 in Pittsburgh, the daughter of James Murray, a coal company salesman, and Daisy Piper Murray, who died in an influenza epidemic when Helen was six. The family moved to Youngstown, Ohio, when Helen was three. One of her earliest recollections is accompanying her father, "a real wonderful guy," on his rounds as he sold coal to dealers who in turn sold to homes.

Helen attended Youngstown public schools through the sixth grade, moving to the suburb of Poland for the seventh grade, where she finished elementary school and high school and where she received straight "A's." So did another female student, but because her father could afford to send her to college, the school arbitrarily designated the other student valedictorian and Helen salutatorian. "I didn't think it was very fair," she remembers, "but what could I do about it?"

Free had exposure to chemistry and physics in high school, but she intended to be a Latin and English teacher when she entered the College of Wooster in September 1941. That changed after Pearl Harbor, when the housemother announced that with all the men gone to war, the "girls" should take science. She turned to Free and said, "Helen, you're taking chemistry, aren't you? Why don't you switch [majors]?" Reflecting on her decision to agree with the housemother, Free observes, "Just like that! I think that was the most terrific thing that ever happened because I certainly wouldn't have done the things I've done in my lifetime."

Free took the requisite chemistry courses, and upon graduation landed an interview at Miles Laboratories. She went to Elkhart for the interview and remembers being crammed in a car with three or four men who were going to lunch at the Friday Club, which did not admit women, so they dropped her off at the YWCA. Though she did not get lunch, she was offered a job in the control laboratory testing ingredients for vitamins.

After a few years in the control laboratory, Helen joined her future husband's research team, a move that satisfied her wish to do research. Al and Helen, who married in 1947, became lifelong research partners as well.

Research at first turned out to be less than Helen Free had anticipated. It was, she remembers, "just as routine as quality control... I did bilirubins all day long, day in and day out." On the other hand, Free found it "kind of neat" at first, because the work aimed to discover a new antibiotic. Unfortunately, the effort failed.

Helen Free retired in 1982 but continued as a consultant with what is now Bayer HealthCare LLC through 2007. She has remained active as a champion of science education and outreach. Free chaired the National Chemistry Week task force of the American Chemical Society for five years, and in 1993 she was elected president of the Society, using her post to raise public awareness of the contributions of chemistry to modern life. The ACS created an award in her honor, the Helen M. Free Award in Public Outreach. In 2000, Helen and Al Free were inducted into the National Inventors Hall of Fame.

Al and Helen had six children together. Al Free died in 2000.

Helen Free

The ACS created an award in her honor, the Helen M. Free Award in Public Outreach.

National Historic Chemical Landmark

The American Chemical Society designated development of diagnostic test strips as a National Historic Chemical Landmark in a ceremony at ETHOS in Elkhart, Indiana, on May 1, 2010. The text of the plaque commemorating the development reads:

A Miles Laboratories research team led by Alfred and Helen Free developed the first diagnostic test strip, Clinistix®, for detecting glucose in urine. Reagent-impregnated strips changed color based on the concentration of glucose. This breakthrough led to additional dip-and-read tests for proteins and other substances. Subsequently, researchers devised a method to combine several tests on one strip to provide healthcare professionals with simple, immediate tools to aid in the detection of disease. These innovations, along with instrument-based measurement of glucose in fingertip blood, provided patients with inexpensive means to aid in the management of diabetes and kidney disease, significantly improving their quality of life.

About the National Historic Chemical Landmarks Program

The American Chemical Society, the world's largest scientific society with more than 161,000 members, has designated landmarks in the history of chemistry since 1993. The process begins at the local level. Members identify milestones in their cities or regions, document their importance, and nominate them for landmark designation. An international committee of chemists, chemical engineers, museum curators, and historians evaluates each nomination. For more information, please call the Office of Public Affairs at 202-872-6214 or 800-227-5558 ext. 6214, e-mail us at nhclp@acs.org, or visit our web site: www.acs.org/landmarks.

A nonprofit organization, the American Chemical Society publishes scientific journals and databases, convenes major research conferences, and provides educational, science policy, and career programs in chemistry. Its main offices are in Washington, DC, and Columbus, Ohio.

Acknowledgments

Written by Judah Ginsberg

Photo credits: Bern Harrison, Bayer HealthCare LLC

Quotations on the inside pages of this brochure are taken from the following: Helen Murray Free, interview by James J. Bohning in Elkhart, Indiana, 14 December 1998 (Philadelphia: Chemical Heritage Foundation, Oral History Transcript #0176).

The author wishes to thank Frankie Wood-Black, Maureen Chan, Janan Hayes, and Carmen Giunta of the National Historic Chemical Landmarks Committee and members of the site committee, all of whom read this brochure in draft form and whose suggestions improved the text. The author wishes to thank especially Helen Free, for agreeing to be interviewed and for reading this brochure in draft form. Needless to say, any remaining errors are the author's alone.

Designed by MSK Partners, Hunt Valley, Maryland

ACS Chemistry for Life®

American Chemical Society
Office of Public Affairs
National Historic Chemical Landmarks Program
1155 Sixteenth Street, NW
Washington, DC 20036
202-872-6214
800-227-5558 ext. 6214
www.acs.org/landmarks